



MOLDOVA STATE UNIVERSITY
Center of Functional Genetics, Faculty of Biology and Pedology
Doctoral School in Biological, Geonomic, Chemical and Technological Sciences
Scientific Association of Geneticists and Breeders of the Republic of Moldova

The National Conference with international participation

**LIFE SCIENCES IN THE DIALOGUE OF
GENERATIONS: CONNECTIONS
BETWEEN UNIVERSITIES, ACADEMIA
AND BUSINESS COMMUNITY**

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SUMMARY

A. PLANT GENETICS, BREEDING AND PROTECTION		
1.	Anton F. G., Joita-Pacureanu M., Risnoveanu L. SOURCES OF RESISTANCE AT SUNFLOWER DOWNY MILDEW	20
2.	Bahsiev A., Zamorzaeva I. MOLECULAR DIAGNOSIS OF PHYTOPLASMA IN THE WILD TOMATO SPECIES <i>SOLANUM HABROCHAITES</i>	21
3.	Balan P., Talpalaru D. THE GROWTH AND FRUCTIFICATION OF APPLE VARIETIES DEPENDING ON VARIETY AND THE PRUNING SYSTEM	22
4.	Balan P., Talpalaru D., Bilici I. THE PRODUCTIVITY OF APPLE ORCHARDS DEPENDING ON VARIETY AND PRUNING SYSTEM	23
5.	Balici E., Grajdieru C. EVALUATION OF RESISTANCE TO TOXIGENIC FUNGI IN SEVERAL MAIZE INBRED LINES	24
6.	Balmuş Z., Cotelea L., Butnaraş V. PERFORMAND NEW VARIETIES OF <i>SALVIA SCLAREA</i> (CLARY SAGE)	25
7.	Batiru Gr., Comarova G., Adamciuc A., Bounegru S., Cojocari D., Rotari E. PECULIARITIES OF STORAGE PROTEIN POLYMORPHISM IN THE ENDOSPERM OF MUTANT MAIZE LINES	26
8.	Бречко Е.В. УСОВЕРШЕНСТВОВАНИЕ СИСТЕМЫ МЕРОПРИЯТИЙ ПО ЗАЩИТЕ ЗЕРНА ОТ ЧЛЕНИСТОНОГИХ В БЕЛАРУСИ	27
9.	Burcovschi I. IMPACT OF ENVIRONMENTAL CONDITIONS ON THE PRODUCTIVITY OF SUNFLOWER HYBRIDS	28
10.	Butnarash V., Balmush Z., Goncariuc M., Cotelea L. EVALUATION OF PERFORMANT HYBRIDS IN DIFFERENT YEARS OF VEGETATION	29
11.	Cassir P., Ghendov V. <i>LEUCOJUM AESTIVUM</i> (AMARYLLIDACEAE) – NEW SPECIES FOR THE LOWER PRUT FLORA	30
12.	Cassir P., Izverscaia T., Ghendov V. REINTRODUCTION OF <i>CRAMBE TATARIA</i> IN THE „LOWER PRUT LAKES” RAMSAR SITE	31
13.	Ciobanu R. ASSESSMENT OF THE MORPHOGENETIC AND REGENERATIVE POTENTIAL OF TRITICALE GENOTYPES IN <i>IN VITRO</i> CULTURE	32

14.	Clapco S. GENETIC DIFFERENTIATION AND RELATIONSHIPS OF <i>O. CUMANA</i> POPULATIONS FROM DIFFERENT SUNFLOWER PRODUCING COUNTRIES	34
15.	Climenco O.A. VARIABILITY OF QUANTITATIVE TRAITS OF CORN HYBRIDS AND INBRED LINES UNDER DROUGHT AND SALINITY	35
16.	Cotelea L., Gonceariuc M., Balmuş Z., Butnarus V., Botnarenco P. GENOTYPES OF <i>S. SCLAREA</i> ESTABLISHMENT OF ESSENTIAL OIL	36
17.	Cristea N., Gavzer S., Lupascu G. MANIFESTATION OF TRANSGRESSIONS ON THE TRAITS OF SPIKE PRODUCTIVITY IN F ₂ POPULATIONS OF COMMON WHEAT	37
18.	Crucean Ş., Scerbacova T., Lungu A. <i>TRICHODERMA</i> SPECIES FOR PLANTS PROTECTION AGAINST ALTERNARIOIS	38
19.	Curshunji D. VARIABILITY SOME MORPH-BIOLOGICAL TRAITS IN BREEDING MATERIAL OF CHICKPEA	39
20.	Duca M., Mutu A., Bivol I. ANALYSIS OF MOLECULAR VARIANCE (AMOVA) OF <i>OROBANCHE CUMANA</i> POPULATIONS	40
21.	Elisovetcaia D., Ivanova R., Ciorchina P., Brindza J. ADAPTIVE CAPACITY OF THE <i>FAGUS SYLVATICA</i> L. POPULATION FROM THE SLOVAK REPUBLIC	41
22.	Erhan T., Raileanu N., Jalba S., Odobescu V., Bogaciiov E. EVALUATION, ESTIMATION AND MONITORING <i>DIABROTICA VIRGIFERA</i> LE CONTE	42
23.	Gladei M. THE DEVELOPMENT OF VITICULTURE THROUGH THE REQUIREMENTS PRISM OF GREEN ECONOMY	43
24.	Grajdieru C. MOLECULAR ASSESSMENT OF <i>F. GRAMINEARUM</i> IN SEVERAL MOLDAVIAN MAIZE GENOTYPES	44
25.	Gribincea V. ASSESSMENT OF GERMPASM DIVERSITY USED IN THE MAIZE INBRED LINES DEVELOPMENT	45
26.	Gusan A., Tretiacova T., Popa A., Gladcaia A. BIOLOGICAL EFFICACY OF THE NEEM OIL FOR THE CONTROL OF <i>APHIS GOSSYPII</i>	46
27.	Makovey M.D. PHENOTYPICAL VARIABILITY OF FETAL TRAITS IN MUTANT TOMATO FORMS	47
28.	Makovey M.D. USING MUTANT <i>ls</i> AND <i>br</i> GENES OF TOMATO TO CREATE A NEW SOURCE MATERIAL	48
29.	Marii L., Andronic L., Smerea S. EVALUATION OF ROS ACCUMULATION IN TOMATO ROOTS DURING POSTSTRESS ACLIMATIZATION	49

30.	Martea R., Mutu A., Gîscă I. EXPANSION AND DIVERSITY OF THE BROOMRAPE RACES IN THE REPUBLIC OF MOLDOVA	50
31.	Mihaila V., Brinzan A. BIOMORPHOLOGICAL PECULIARITIES OF SOME SPECIES OF THE GENUS <i>CUPHEA</i> IN CONDITIONS OF INTRODUCTION	51
32.	Moldovan C., Sîrbu T. STUDY OF LAKE FUNGI BIODIVERSITY IN FROM THE <i>LA IZVOR</i> LAKE (CHISINAU MUNICIPALITY)	52
33.	Nikolić V., Žilić S., Simić M., Kandić V., Perić V., Srdić J., Kravić N. NUTRITIONAL QUALITY AND DIGESTIBILITY OF MAIZE HYBRID PLANTS FOR SILAGE	53
34.	Pintea M., Cozmic R., Borozan E. IMPORTANCE OF WALNUT (<i>Juglans regia</i> L.) WITHIN REPUBLIC OF MOLDOVA	54
35.	Pintea M., Cozmic R., Terentii P. LOCAL PLUM VARIETIES OF REPUBLIC OF MOLDOVA FOR BREEDING AND PRACTICAL PURPOSES	56
36.	Popa A., Todiras V., Gusan A. EFFECTIVENESS OF CARBECOL IN PREVENTING AND COMBATING POWDERY MILDEW ON VINE	58
37.	Port A. DIFFERENTIAL GENES EXPRESSION UNDER ANTERO- AND RETROGRADE CONTROL IN SUNFLOWER MICROSPOROGENESIS	59
38.	Raileanu N., Stratulat T., Jalba S. REVIEW OF PESTS OF THE FOREST PARK "RISHCANI", CHISINAU	60
39.	Risnoveanu L., Oprea D., Joita-Pacureanu M., Anton F. G., Sava E. BEHAVIOR OF SOME SUNFLOWER HYBRIDS AT THE ATTACK OF THE PATHOGEN <i>B.cinerea</i> Pers., IN THE SOUTH-EAST AREA OF ROMANIA	61
40.	Rotari E., Dreglea M. ENERGETIC CRISIS AND GRAIN SORGHUM VARIETY MODEL	62
41.	Rusu Iu. PRELIMINARY DATA ON THE DEVELOPMENT OF THE INVASIVE SPECIES <i>HALYOMORPHA HALYS</i> (STAL) 1855, (HEMIPTERA; PENTATOMIDAE) IN THE REPUBLIC OF MOLDOVA	63
42.	Saltanovici T., Doncila A., Andronic L., Antoci L. SCREENING FOR HEAT-RESISTANCE OF POLLEN IN PROGENY OF VIRUS-INFECTED TOMATO GENOTYPES	64
43.	Samoilova A. PHAGE EFFICIENT AGAINST FIRE BLIGHT AND FRUIT TREES BACTERIAL CANKER PATHOGENS	65
44.	Sarban V., Buză C. THE IMPACT OF THE PRUNING SYSTEM ON THE SWEET CHERRY TREE GROWTH AND FRUCTIFICATION	66
45.	Sarban V., Buză C., Ivanov I. THE IMPACT OF THE CROWN SHAPE ON THE GROWTH AND FRUCTIFICATION OF CHERRY TREE VARIETIES GROWN IN AN INTENSIVE SYSTEM	67

46.	Savranschii D., Gușan A., Tretiacova T., Popa A., Todiraș V. EVALUATION OF <i>SOPHORA FLAVESCENS</i> EXTRACT FOR COMBAT THE RED MITE (<i>TETRANYCHUS URTICAE</i>) TO THE TOMATO CROP IN GREENHOUSE	68
47.	Shleahitici V., Raileanu N., Odobescu V., Jalba S. INFLUENCE OF MINOR COMPONENTS ON THE EFFICIENCY OF APPLE WORM SEX PHEROMONE	69
48.	Spinu A. RELATIONSHIP BETWEEN RELATIVE MATURITY AND GRAIN YIELD IN EXPERIMENTED NEW MAIZE HYBRIDS	70
49.	Stingaci A., Voloshyuk L. BIOPESTICIDES - AN ALTERNATIVE AND ECO-FRIENDLY SOURCE FOR THE CONTROL OF PESTS	71
50.	Tanachi T., Roșca I. BIO-ECOLOGICAL PECULIARITIES OF SOME NEW TAXA OF <i>BERBERIS THUNBERGII</i> DC.	72
51.	Yan N., Liu Z., Zhang J., Zhao J. TRANSCRIPTOME ANALYSIS AND RESISTANCE GENE MINING OF SUNFLOWER RESISTANT <i>OROBANCHE CUMANA</i> RACE G	73
52.	Yang J., Duan R., Mandela Elorm A., Zhang W., Yun X., Du L., Duan X., Zhang J., Zhao J. FIELD RESISTANCE OF SUNFLOWER VARIETIES RESOURCES TO SEED RUST SPOTS DISEASE	74
B. PLANT, ANIMAL AND MICROBIAL BIOTECHNOLOGY		
1.	Ababii A., Țiței V., Doroftei V., Covalciuc D., Chisnicean L., Cozari S., Mocanu N. THE BIOMASS QUALITY OF CHIA <i>SALVIA HISPANICA</i> L. AND PROSPECTS FOR ITS USE IN MOLDOVA	76
2.	Artiomov L., Frunze N. THE INFLUENCE OF THE AGRICULTURAL MANAGEMENT SYSTEM ON THE SOIL MICROBIOME	77
3.	Balacci S., Balan I., Buzan V. DIRECTING THE GROWTH OF CARP LARVAE THROUGH THE APPLICATION OF THERMAL FACTOR	78
4.	Balan I., Roșca N., Buzan V., Balacci S., Moroz M., Cazacova Iu., Mereuța I., Cretu R., Bacu Gh. THE ROLE AND IMPORTANCE OF REPRODUCTIVE BIOTECHNOLOGIES IN ANIMAL BIODIVERSITY	79
5.	Bivol A., Toderăș I., Mager M., Bădărău S., Iurcu-Străistaru E., Rusu Ș., Bivol E. RESEARCH ON THE INVASIVE IMPACT OF HARMFUL INSECT COMPLEXES ASSOCIATED WITH PARASITIC NEMATODES AND PATHOGENIC VIRUS VECTORS IN PRODUCTIVE PLUM ORCHARDS	80
6.	Bîrsa M., Burțeva S., Cebotari V. VIABILITY OF DREPTOMYCES STRAINS AND ITS VARIANTS AFTER FREEZE-DRYING IN CNMN	82

7.	Bogdan V. SPECIFIC STREPTOCOCCI OF THE DIGESTIVE TRACT AND THEIR SUITABILITY FOR INCLUSION IN PROBIOTIC PREPARATIONS	83
8.	Burduja D., Buşmachiş G., Bacal S. NEW DATA ON LADYBUGS (INSECTA) FROM THE REPUBLIC OF MOLDOVA	84
9.	Chitan R., Ciorchină N., Tabăra M. INITIATION OF THE <i>IN VITRO</i> CULTURE OF THE <i>MACROCARPON VACCINIUM</i> AITON VARIETY 'EARLY BLACK'	85
10.	Ciloci A., Clapco S., Dvornina E., Labliuc S., Bulhac I., Ureche D. INFLUENCE OF SOME COORDINATION COMPOUNDS WITH POLYDENTATE LIGANDS ON THE PROTEOLYTIC ACTIVITY OF <i>FUSARIUM GIBBOSUM</i> CNMN FD 12	86
11.	Coşcodan M., Todiraş V., Prisacari S., Lungu A. MEANS OF ENHANCING THE ROLE OF BIOLOGICAL NITROGEN IN PHYTOTECHNY	87
12.	Coscodan M. THE USE OF MICROBIAL BIOPREPARATIONS IN MODERN BIOTECHNOLOGIES	88
13.	Coscodan M. BIODETERIATION OF PLASTIC MATERIALS BY PHYTOREMEDIATING MICROORGANISMS	89
14.	Curiev L. STUDY OF THE SYNERGISM BETWEEN MICROBIOLOGICAL AGENTS IN CONTROL OF APPLE SCAR	90
15.	Demcenco B., Balan I., Petcu I., Osadci N., Roşca F., Gramovici A. ANTIOXIDATIVE PROTECTION IN THE BIOTECHNOLOGY OF AGRICULTURAL BREEDING	91
16.	Frunzete M. E., Zamfirache M. M., Ivănescu L. C. <i>TAXUS BACCATA</i> L.-MORPHO-STRUCTURAL CHARACTERS IN SPONTANEOUS AND CULTIVATED TAXA	92
17.	Gherasim E. THE ROLE OF <i>PELOPHYLAX RIDIBUNDUS</i> (PALLAS, 1771) IN THE FORMATION AND MAINTENANCE OF PARASITIC ZOONOSES	93
18.	Ghereg M., Ciorchină N. ASEPTICIZATION OF PLANT MATERIAL OF SOME SPECIES OF FAM. <i>AMARYLLIDACEAE</i> L.	95
19.	Guţu N. CONDITIONALLY PATHOGENIC AGENTS OF THE FAMILY ENTEROBACTERIACEAE CAUSING ACUTE DIARRHEA DISEASES	96
20.	Lungu A. INFLUENCE OF EXTERNAL FACTORS ON THE DEVELOPMENT OF <i>S. SPINOSA</i> ON THE LIQUID MEDIUM	97
21.	Mihăşan M., Boianşu R. Ş., Guzun D., Babii C., Channaveerappa D., Aslebagh R., Dupree E., Daric C. PROTEOMICS ANALYSIS OF NICOTINE METABOLISM REGULATION IN <i>PAENARTHROBACTER NICOTINOVORANS</i>	98

22.	Mustya M. ATHERINA (<i>ATHERINA BOYERI</i> RISSO, 1810) OF KUCHURGAN RESERVOIR	99
23.	Paladi V. PRESENCE OF THE SPECIES <i>GLAREOLA PRATINCOLA</i> (CHARADRIIFORMES, GLAREOLIDAE) IN THE LOWER PRUT ARIA	100
24.	Petcu I., Balan I., Demcenco B., Osadci N., Roșca F., Gramovici A. THE INFLUENCE OF CLASSICAL BIOTECHNOLOGIES ON THE WELFARE OF AGRICULTURAL BIRDS	101
25.	Postovan R. REFERINȚE ASUPRA CONDIȚIILOR GEOGRAFICE ȘI A CERINȚELOR DE PROIECT PENTRU REPARAȚIA PODULUI PESTE RÂUL RĂUT ÎNTRE LOCALITĂȚILE USTIA ȘI ZOLONCENI DIN RAIONUL CRIULENI	102
26.	Rosca N., Balan I., Balacci S., Buzan V., Cretu R., Moroz M., Osipciuc G., Bacu Gh. ADVANTAGES OF CRYOCONSERVATION OF SPERM IN REPRODUCTIVE BIOTECHNOLOGY	103
27.	Sîrbu T. Țurcan O., Moldovan C., Timuș I. VIABILITY AND STABILITY OF AQUATIC FUNGI OF BIOTECHNOLOGICAL INTEREST AFTER LYOPHILIZATION	104
28.	Sîrbu T., Țurcan O., Timuș I. THE IMPACT OF ZnO AND Cu NANOPARTICLES SUPPLEMENTED IN THE REHYDRATION MEDIUM ON LYOPHILIZED MICROMYCETES	105
29.	Sitnic V., Caraman N., Caldari V., Sitnic V. DENSITY DYNAMICS OF RODENT SPECIES IN AGROCENOSSES IN THE REPUBLIC OF MOLDOVA IN 2021	106
30.	Stingaci A., Serbacova T., Samoiloa A., Zavtoni P., David T., Lungu A., Curiev L. COMPLEX APPLICATION OF <i>BACILLUS SPP.</i> AND BIOREGULATORS FOR THE CONTROL OF PESTS	107
31.	Timuș I. IMPACT OF NANOPARTICLES IN THE CULTIVATION MEDIUM ON THE VIABILITY AND STABILITY OF MICROMYCETES AFTER LYOPHILIZATION	108
32.	Țiței V., Doroftei V., Cozari S., Mocanu N. THE QUALITY OF FRESH AND ENSEILED BIOMASS FROM NEW CULTIVAR „MARIA” OF <i>HELIANTHUS TUBEROSUS</i>	109
33.	Tofan E., Chiselița N., Chiselița O., Beșliu A., Efremova N. VALORIZATION OF WINE YEAST SEDIMENTS AS A SOURCE OF LIPID PREPARATIONS	110
34.	Tolstenco D., Leorda A. CAPACITY OF MICROBIAL MULTIPLICATION DEPENDING ON THE FORM OF PREPARATION OF SOME ONCOPROTECTIVE PLANTS	111
35.	Țugulea C., Bușmachiu G. NEW FAUNISTIC RECORDS OF THE ENDANGERED SPECIES <i>PROSERPINUS PROSERPINA</i> (LEPIDOPTERA: SPHINGIDAE) FROM THE REPUBLIC OF MOLDOVA	112

36.	Turcan O. DYNAMICS OF ACCUMULATION OF EXOPOLYSACCHARIDES IN CULTURAL LIQUID AT THE CULTIVATION OF <i>SPIRULINA PLATENSIS</i> SUPPLEMENTED WITH COORDINATIVE COMPOUNDS OF CU (II)	113
37.	Turcan O., Sîrbu T. METHODS OF CONSERVATION OF MICROALGAE AND CYANOBACTERIA	114
38.	Volosciuc L., Lungu A. ACTINOBACTERIA AS BIOCONTROL AGENTS FOR COMBATING PEST INSECTS	115
39.	Zamornea M., Rusu Ș., Erhan D., Chihai O., Gliga O., Botnaru N. HELMINTOFAUNA IN PHEASANT (<i>PHASIANUS COLCHICUS L.</i>) MAINTAINED IN CAPTIVITY IN MOLDOVA	116
40.	Zosim L., Bivol C., Elenciuc D. PRODUCTIVITY, CAROTENOID AND GLYCEROL CONTENT OF <i>DUNALIELLA SALINA</i> CULTIVATED IN THE PRESENCE OF GEO ₂ WITH VARYING LIGHTING REGIME	117
41.	Zosim L., Trofim A., Balan G., Rudic V., Elenciuc D. ANTIMICROBIAL ACTIVITY OF POLYSACCHARIDE-CONTAINING SPIRULINA EXTRACTS	118
C. MOLECULAR BIOLOGY AND BIOMEDICINE		
1.	Andriuță C. NUTRIGENETICS-THE FUTURE OF PERSONALIZED NUTRITION	120
2.	Babileva A. THE INFLUENCE OF NUTRIENTS ON THE METABOLISM DEPENDING ON THE TYPE OF STRESS REACTIVITY	121
3.	Bacalov Iu., Crivoi A., Chirița E., Bîrsan A., Druța A., Barbăroș M., Revenco A., Popușoi Iu. IMPACT OF THE PHYTOPREPARATUS APUSET-6 ON THE ENDOCRINE PANCREAS-THYROID AXIS	122
4.	Baciu A.Ja., Mereuta I., Fedas V., Listopadova L. PROPHYLAXIS OF RESPIRATORY SYNDROMES BY NEUROIMMUNOMODULATORY ACTION OF NATURAL ENVIRONMENTAL FACTORS	123
5.	Boiciuc C., Blanita D., Hlistun V., Leferber D., Usurelu N. DIFFERENTIAL DIAGNOSIS OF CDG THROUGH MOLECULAR ANALYSIS OF GALT AND ALDOB GENES	124
6.	Brînză I., Stache A., Eldahshan O., Gorgan L., Mihasan M., Hritcu L. BAICALEIN PREVENT SCOPOLAMINE-INDUCED MEMORY IMPAIRMENT IN ZEBRA FISH BY INCREASING THE CREB PROTEIN LEVEL AND THE mRNA EXPRESSION OF <i>BDNF</i> AND <i>CREB</i> GENE	125
7.	Coliban Iu., Blăniță D., Hadjiu S., Ușurelu N., Revenco N., Sacară V. THE LINK BETWEEN CLINICAL MANIFESTATIONS OF SMA AND UNBALANCED GENOMIC CHANGES	126
8.	Dorif A., Sacara V. DEVELOPMENT OF METHOD FOR SHORT REPEATS EXPANSION	127

	CAUSED ATAXIAS DIAGNOSTIC	
9.	Duca M., Bivol I. EVALUATION OF POLYMORPHISM INFORMATION OF GENETIC DIVERSITY IN BROOMRAPE FROM BULGARIA	128
10.	EISabeh A., Honceriu I., Kallabi F., Boiangiu R., Mihășan M. GENOME OF THE NICOTINE-DEGRADING <i>PAENARTHROBACTER NICOTINOVORANS</i> ATCC 49919	129
11.	Frunze N. MOLECULAR ANALYSIS OF PROKARYOTIC MICROBIAL COMMUNITIES IN A TYPICAL CHERNOZEM	130
12.	Garbuzneac A., Sheptitsky V. CONDITIONED REFLEX LEARNING AND MEMORY OF WHITE RATS OF DIFFERENT AGES UNDER THE INFLUENCE OF THE BIOMASS OF STREPTOMYCETES ISOLATED FROM THE SOILS OF THE REPUBLIC OF MOLDOVA	131
13.	Grosul-Raileanu O., Sheptitsky V. ACTIVITY OF DIGESTIVE ENZYMES OF THE SMALL INTESTINE OF RATS WITH DIFFERENT CONSTITUTIONAL STRESS REACTIVITY	132
14.	Mangul O., Sheptitsky V. MEMBRANE HYDROLYSIS OF MALTOSE IN THE SMALL INTESTINE UNDER THE INFLUENCE OF DIETS WITH DIFFERENT CONTENT OF CARBOHYDRATES IN EARLY POSTNATAL ONTOGENESIS	133
15.	Mereuță I., Leorda A., Poleacova L. THE PERSPECTIVE OF THE USE OF PREBIOTICS IN METABOLIC DISORDERS	134
16.	Mihasan M. 3D PRINTING MACROMOLECULAR MODELS FOR TEACHING AND DEMONSTRATION	135
17.	Musteata V., Musteata L., Coman D. MANAGEMENT ISSUES OF CHRONIC LYMPHOCYTIC LEUKEMIA	136
18.	Poleacova L. THE MODIFICATION OF FREE AMINO ACIDS RATS FED WITH PROTEIN RATION	137
19.	Pușica Z. PSYCHOGENIC STRESS IN HOSPITALIZED PERSONS	138
20.	Racoviță S., Moșin V., Sprincean M. ANALYSIS OF Y CHROMOSOME MICRODELETIONS AND CFTR GENE MUTATIONS AS GENETIC MARKERS IN MALE INFERTILITY	139
21.	Rotaru L., Rotaru T. MOLECULAR-GENETIC ASPECTS OF MIGRAINE	140
22.	Sacara V., Dorif A. USE OF AI-ASSISTED TOOLS IN HEREDITARY DISEASE DIAGNOSTIC	141
23.	Secu D., Ușurelu N., Blăniță D., Sacară V. OVERLAP OF CLINICAL MANIFESTATIONS IN MITOCHONDRIAL DISEASES	142
24.	Secu Gh., Secu D., Roșca T. THE BENEFITS OF L-ARGININE IN PREECLAMPSIA	143

25.	Sitnic V. COMPARATIVE <i>IN SILICO</i> ANALYSIS OF PRIMERS USED FOR VERTEBRATE METABARCODING	144
26.	Usurelu D.-C., Scurtul M., Boiciuc C., Blanita D., Sacara V., Usurelu N. DIFFERENTIATION OF THE FORMS OF PHENYLKETONURIA BASED ON GENETIC DETERMINISM	145
D. ENVIRONMENT PROTECTION AND NATURAL RESOURCES MANAGEMENT		
1.	Agapi I. DENDROLOGICAL ASPECTS OF VEGETATIVE REPRODUCTION OF SOME WALNUT GENOTYPES	147
2.	Anastas A. GROUNDWATER CHEMISTRY OF SARMATIAN AQUIFERS IN RABNITA AND DUBASARI DISTRICTS	148
3.	Bacal S., Buşmachiu G. CONTRIBUTIONS TO THE STUDY OF SAPROXYLIC BEETLES (INSECTS: COLEOPTERA) FROM THE REPUBLIC OF MOLDOVA	149
4.	Banari E. STUDY IN THE FIELD OF REDUCING THE HARMFULNESS OF A BIODIESEL POWERED ENGINE	150
5.	Bejan A. EVALUATION OF TRENDS IN CHANGING THE LANDSCAPE STRUCTURE ON THE EXAMPLE OF THE CUBOLTA HYDROGRAPHIC BASIN	151
6.	Belous S. <i>IRIS VARIEGATA</i> (IRIDACEAE) IN THE FLORA OF LANDSCAPE RESERVE "CĂRBUNA"	152
7.	Blonschi V., Gladchi V. ADAPTATION OF THE METHOD FOR DETERMINING VITAMIN B ₉ IN AQUEOUS SOLUTIONS	153
8.	Bogdan N., Sirbu T., Bîrsa M., Cernăuţeanu V., Cojocaru I. OPEN SCIENCE PRACTICE USED IN STRENGTHENING THE NATIONAL COLLECTION OF NON-PATHOGENIC MICROORGANISMS	154
9.	Bogdan N., Slanina V. BACTERIAL VIABILITY AFTER 15 YEARS STORAGE	155
10.	Boian I. THE RISK OF LATE SPRING FROSTS FOR AGRICULTURE OF THE REPUBLIC OF MOLDOVA	156
11.	Borodaev R. ON THE MECHANISM OF CHEMICAL SELF-PURIFICATION OF VARIOUS WATER SYSTEMS OF THE LOWER DNIESTER BASIN	157
12.	Botnari A. SPATIAL MODELING OF DANGEROUS FROSTS IN THE REPUBLIC OF MOLDOVA	158
13.	Calestru L., Belova V. CONTRIBUTIONS TO THE KNOWLEDGE OF LEAF BEETLES	159

	(CHRYSOMELIDAE) FROM ALFALFA	
14.	Caraman N. INERESPECIFIC BEHAVIOUR BETWEEN <i>APODEMUS FLAVICOLLIS</i> AND <i>APODEMUS SYLVATICUS</i> FEMALES FROM URBAN ECOSYSTEMS OF CHIȘINĂU CITY, REPUBLIC OF MOLDOVA	160
15.	Cazac V. CLIMATIC AND HYDROMETEOROLOGICAL PHENOMENA OF RISK ON THE TERRITORY OF THE REPUBLIC OF MOLDOVA	161
16.	Chihai O., Nistreanu V., Larion A., Rusu Ș., Zamornea M., Melnic G. PARASITE FAUNA IN PYGMY FIELD MOUSE FROM VARIOUS BIOTOPES OF THE REPUBLIC OF MOLDOVA	163
17.	Cîrlig N., Țiței V., Guțu A., Iurcu-Străistaru E. CONTRIBUTION TO THE STUDY OF <i>FAGOPYRUM ESCULENTUM</i> MOENCH IN THE REPUBLIC OF MOLDOVA	164
18.	Codreanu L. CYANOBACTERIUM <i>NOSTOC LINCKIA</i> GROWTH UNDER DIFFERENT CONCENTRATIONS OF COPPER(II) IONS	165
19.	Colibaba L. THE ROLE OF CLIMATE FACTORS IN THE PROCESS OF AGRICULTURAL PRODUCTION IN MOLDOVA	166
20.	Corcimaru, S., Mereniuc, L., Sîtnic, F., Mereniuc, R. MICROBIOLOGICAL TOOLS FOR ASSESSING IMPACTS ON SOIL ORGANIC MATTER CONTENT	167
21.	Corlateanu L., Ganea A., Leatamborg S. STORAGE POTENTIAL OF TRITICALE ACCESSIONS - INDICATOR OF THEIR VIABILITY UNDER <i>EX SITU</i> CONSERVATION	168
22.	Domenco R. THE IMPACT OF PRECIPITATION ON THE SUNFLOWER CROP IN THE NORTHERN REGION OF THE REPUBLIC OF MOLDOVA	169
23.	Dudnicenco T. ECOTOURISM POTENTIAL OF BIODIVERSITY FROM CRIULENI DISTRICT, REPUBLIC OF MOLDOVA	170
24.	Dudnicenco T. ASPECTS REGARDING THE ECOLOGICAL SITUATION OF SOME PROTECTED AREAS IN CHISINAU MUNICIPALITY	171
25.	Eroshenkova V.A., Bulimaga C.P., Doroftei S.D., Demchukova N.V. ASSESSMENT OF THE ANTHROPOGENIC IMPACT ON THE WATER QUALITY OF THE LOWER DNIESTER TRIBUTIES	172
26.	Eroshenkova V.A., Bulimaga C.P., Lukasheva N.V., Medvedeva N.L. MICROBIOLOGICAL POLLUTION OF SMALL TRIBUTIES OF THE LOWER PART OF THE DNIESTR	173
27.	Frunză M., Bîrsan A. CYTOTOXIC EFFECT OF NANOCOMPOSITES BASED ON VANADIUM OXIDES	174
28.	Gladchi V., Blonschi V. PARTICIPATION OF THE VITAMIN B COMPLEX IN PROCESSES OF CHEMICAL SELF-PURIFICATION OF THE AQUATIC ENVIRONMENT	175

29.	Grozdeva S. NEW SPECIES OF <i>MACROSTELUS</i> (HEMIPTERA, AUCHENORRHYNCHA) IN THE REPUBLIC OF MOLDOVA	176
30.	Indoitu D. SOME AGROCHEMICAL PROPERTIES OF CARBONATE CHERNOZEM AND DIFFERENT SOIL USE	177
31.	Iurcu-Străistaru E., Bivol A., Meleca A., Criucikov O., Rusu Ș., Cîrlig N., Bivol E. THE ASSOCIATIVE AND INVASIVE IMPACT CAUSED BY COMPLEXES OF PARASITIC INSECTS AND NEMATODES WITH THE APPLICATION OF CHEMICAL MANAGEMENT IN MAIZE PLANTATIONS UNDER THE CONDITIONS OF THE REPUBLIC OF MOLDOVA	178
32.	Jigău Gh., Ciolacu T., Turchin B., Stadnic A., Dobrojan S. CLIMATE-OPTIMIZED ADAPTIVE-LANDSCAPE TECHNOLOGIES: CONCEPTUAL-THEORETICAL AND APPLICATIVE SUPPORT	180
33.	Marareskul V.A. FOSSIL FELIDAE (GENUS <i>METAILURUS</i>) IN THE MIOCENE OF THE REPUBLIC OF MOLDOVA	182
34.	Mînzat C. CONTRIBUTIONS TO THE STUDY OF THE ANTS (INSECTS: FORMICIDAE) FROM THE REPUBLIC OF MOLDOVA	183
35.	Muntean A. DEVELOPMENT OF A MATHEMATICAL MODELS SYSTEM FOR REMOTE ASSESSMENT SOIL EROSION	184
36.	Nistoreanu V. Caldari V., Dibolscaia N., Larion A. BAT FAUNA (MAMMALIA, CHIROPTERA) FROM LIMESTONE MINES OF MOLOVATA NOUĂ	185
37.	Nistoreanu V., Larion A. TROPIC SPECTRUM OF THE LONG-EARED OWL (<i>ASIO OTUS</i>) IN THE REPUBLIC OF MOLDOVA	186
38.	Portarescu A., Certan C., Grabco N., Bulimaga C. THE BIOECOLOGICAL SPECTRUM OF HERBACEOUS SPECIES IN THE CENTER OF BALTI CITY	187
39.	Roman C., Mapelli C., Arsene C., Bejan I.G., Dillon T.J., Olariu R. I. ATMOSPHERIC BREAKDOWN CHEMISTRY OF THE NEW GREEN SOLVENT 2,2,5,5- TETRAMETHYLOXOLANE: GAS-PHASE RATE CONSTANTS EVALUATION TOWARDS OH AND CL RADICALS AT 298 K AND 1 BAR	188
40.	Romanovich N.A. APPLICATION OF BIOTECHNICAL STRUCTURES TO IMPROVE BREEDING CONDITIONS OF WETLAND BIRDS IN THE YAGORLYK NATURE RESERVE	189
41.	Sasco E. GROWTH CHARACTERISTICS OF FUNGAL PATHOGENS IN CONDITIONS OF WATER RESTRICTIONS	190
42.	Shova E. THE IMPACT OF REGIONAL CLIMATE CHANGE ON THE	191

	SUSTAINABLE DEVELOPMENT OF THE AGRICULTURAL SECTOR IN THE REPUBLIC OF MOLDOVA	
43.	Sochircă V., Nagacevschi T. GEOPAEDOLOGICAL INVESTIGATIONS AT THE ARCHAEOLOGICAL SITE LIPOVENI II-LA NISIPĂRIE FROM CIMISLIA DISTRICT	192
44.	Stadnic A. ELEMENTS OF AGROGENIC EVOLUTION OF THE AGGREGATIC STRUCTURE OF CLAYEY-LOAMY AND LOAMY-CLAYEY MODERATE HUMIFERUS CHERNOZEMS	193
45.	Stratan L., Răileanu V. CLIMATE INFLUENCES ON THE LANDSCAPES OF THE NARNOVA HYDROGRAPHIC BASIN	195
46.	Tabara M., Scortesco F., Ghendov V. EX-SITU AND IN-VITRO PROPAGATION OF <i>GENISTA TETRAGONA</i> IN REPUBLIC OF MOLDOVA	196
47.	Tasca C., Duca Gh., Covaci E. THE INFLUENCE OF TOMATIN BAC ON THE PROCESS OF ALCOHOLIC FERMENTATION OF WASTE BIOMAS	197
48.	Țugulea A. FIXED SOURCES OF ATMOSPHERIC AIR POLLUTION IN THE BALTI URBAN ECOSYSTEM	198
49.	Yushin N., Zinicovscaia I., Cepoi L., Chiriac T., Rudi L. EUROPIUM BIOACCUMULATION BY <i>ARTROSPIRA PLATENSIS</i> AND ITS EFFECT ON BIOMASS	199
50.	Zakharov D. S. FINDING OF THE REMAINS OF A FROG AND VARAN FROM THE PLIOCENE LOCALITY PRIOZERNOE	200
E. CHEMISTRY AND CHEMICAL COMPOUNDS IN BIOLOGY, AGRICULTURE AND MEDICINE		
1.	Barbulescu I.D., Frincu M., Begea M., Teodorescu R.I., Dumitrache C., Marculescu I. S., Cîric A.I., Banita C-D., Vrinceanu C-R., Soreanu G., Cretescu I. OBTAINING ACTIVE DRY WINE YEAST BIOMASS AT THE MICROPILOT LEVEL	202
2.	Bîrcă N., Barba A., Kulcițki V. THE USE OF qNMR SPECTROSCOPY FOR ANALYTICAL EVALUATION OF LAVENDER EXTRACTS. DETERMINATION OF ROSMARINIC ACID.	203
3.	Bîrcă N., Jian M., Cobzac V., Morarescu O., Cotelea T., Cirimpei O., Nacu V., Kulcițki V. SELECTIVE EXTRACTION OF POLYPHENOLIC COMPOUNDS FROM <i>HIPPOPHAE RHAMNOIDES</i> SEEDS	204
4.	Bolocan N., Duca Gh. STOPPED-FLOW STUDIES OF THE INTERACTION OF DFH ₄ AND ITS DERIVATIVES WITH DPPH'	205

5.	Bolocan N., Duca Gh. ESTIMATION OF ADMET PROPERTIES OF DFH4 AND ITS NOVEL DERIVATIVES	206
6.	Calalb T. CAROTENOID CONTENT IN PLANT PRODUCTS OF <i>CASSIA OCCIDENTALIS</i> (L.) LINK SPECIES	207
7.	Calalb T., Ciorchina N. CONTENT OF TANNINS IN PLANT PRODUCTS OF SOME SPECIES FROM GENUS ACTINIDIA	208
8.	Ciocarlan A., Aricu A., Lungu L., Blaja S., Popescu V., Zinicovscaia I., Carrupt V. PRELIMINARY PHYTOCHEMICAL ANALYSIS OF CRUD EXTRACT FROM <i>TANACETUM CORYMBOSUM</i> (L.) SHI. BIP.	209
9.	Ciursin A., Rusnac R., Gulea A. SYNTHESIS OF N-CYCLOHEXYL-2-[(3-ETHOXY-2-HYDROXYPHENYL) METHYLIDENE] HYDRAZINE-1-CARBOTHIOAMIDE	210
10.	Duan R., Liu Z., Guo X., Zhang Z., Zhang W., Zhang J., Zhao J. JINMIAO TARGET INHIBITION OF <i>O. CUMANA</i> THE MECHANISM OF PARASITIC SUNFLOWER	211
11.	Erhan T., Gulea A., Garbuz O. STUDY OF THE ANTIOXIDANT PROPERTIES OF SOME METHYLPHENYLTHIOSEMICARBAZONES	212
12.	Ivanova R., Brindza J. NATURAL DYES FROM POKEWEEED BERRIES: EXTRACTION PROCEDURES AND STABILITY	213
13.	Lungu L., Blaja S., Cucicova C., Ciocarlan A., Aricu A. SYNTHESIS OF NEW POTENTIAL ACTIVE HOMODRIMANE SESQUITERPENOIDS WITH BENZIMIDAZOLE FRAGMENT	214
14.	Mîndru A. IRON(III) CLUSTERS WITH 3-FORMYLSALICYLIC ACID	215
15.	Neguța A. SYNTHESIS OF SOME THIOSEMICARBAZONES DERIVED FROM 1-PYRIDIN-2-YL-3-PHENYL-PROP-2-EN-1-ONES (CHALCONES) SUBSTITUTED IN THE PHENYL RING	216
16.	Olaru E.-I., Olaru Ș. M., Zamfirache M.-M., Ivănescu L. C. MICROMORPHOLOGICAL AND ANATOMICAL CHANGES INDUCED BY SALINE STRESS IN <i>OCIMUM BASILICUM</i> L. (VAR. GENOVESE), IN EXPERIMENTAL GROWTH CONDITIONS	217
17.	Popescu V. CHEMICAL COMPOSITION OF DAMASK ROSE (<i>ROSA DAMASCENA</i> MILL.) CONCRETE OF MOLDAVIAN ORIGIN	219
18.	Popușoi A. SYNTHESIS AND STUDY OF Zn TETRA-SUBSTITUTED PHTHALOCYANINE WITH CHALCONIC GROUPS	220
19.	Raischi I. DESIGNING OF THE BEST SLEEP PROTOCOL ACCORDING TO THE PSYCHOPHYSIOLOGICAL NEED	222

20.	Rusnac A., Garbuz O., Shova S., Gulea A. COPPER COMPLEXES WITH N ⁴ (2-ETHYL BENZOATE) THIOSEMICARBAZONE OF 2-ACETYLPIRIDINE	223
21.	Scraliuc M. AUTOPHAGY ROLE IN MODULATING INFLAMMATORY MARKERS RESPONSE IN KERATINOCYTES	224
22.	Suhodol N., Deseatnicov O., Ghendov-Moșanu A., Covaliov E., Sturza R. OPTIMIZATION OF THE EXTRACTION PROCESS OF BIOACTIVE COMPOUNDS FROM PEACHES	225
23.	Talmaci N. Cu(II) COMPLEXES WITH 1,5-BIS(2-HYDROXY-3-METHOXYBENZYLIDENE)CARBONOHYDRAZIDE	226
24.	Ulchina Ia., Graur V., Tsapcov V., Celac M., Garbuz O., Gulea A. CU(II) COMPLEXES WITH 4-ALLYLTHIOSEMICARBAZONE AS POSSIBLE ANTIOXIDANT AGENTS	227
25.	Vasiliev A., Gîrbu V., Morarescu O., Kulcitki V. SYNTHESIS OF NEW <i>EPI</i> -MANOYLOXIDE DERIVATIVES WITH AZIDE AND □-LACTAM FUNCTIONAL GROUPS	228
26.	Vicol C., Duca Gh. PHENOLIC COMPOUNDS AND ANTIOXIDANT ACTIVITY OF SOME MOLDOVAN WHITE AND RED GRAPE VARIETIES AT DIFFERENT MATURATION PERIODS	229
27.	Cepoi L. COPPER COMPOUNDS AS STRESS FACTORS AND REGULATORS IN PHYCOBIOTECHNOLOGY	230
28.	Pacureanu M.J., Popescu G., Ciornei L., Risnoveanu L., Sava E. IMPROVING SUNFLOWER CROP BIODIVERSITY, BY CREATING NEW, MORE PERFORMANT GENOTYPES	231
	AUTHOR INDEX	232

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Session A

PLANT GENETICS, BREEDING AND PROTECTION

SOURCES OF RESISTANCE AT SUNFLOWER DOWNY MILDEW

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Pathogen *Plasmopara halstedii* causes low seed yield at sunflower genotypes, in year with high precipitation and low temperature in first stage of development of sunflower plant. In year 2021, at Fundulea location, sunflower downy mildew has a big attack degree of infection in first stage of development, due to weather conditions and in year 2022, degree attack was very low. In year 2021, only differential line for downy mildew HA 288 (gene *Pl²?*) from Institute of Field and Vegetable Crops Novi Sad, Serbia and differential lines RHA419 (gene *Pl_{Arg}*), accession PI 619204 and HA-R5 (gene *Pl₁₃*), accession PI 650763 from United States National Plant Germplasm System, has 0% attack degree of pathogen *Plasmopara halstedii*.

In year 2022, only differential line for downy mildew RHA274(gene *Pl₂/Pl₂₁*), accession PI 599759, from United States National Plant Germplasm System and differential lines DM 2 93CA from Institute of Field and Vegetable Crops Novi Sad, Serbia and RHA340 (gene *Pl₈*), HA 335 (gene *Pl₆*), has sunflower plant infected with pathogen *Plasmopara halstedii*. Year 2021 was the reference for selection of resistant sunflower genotypes at downy mildew. In year 2021, in Fundulea location, only three sunflower genotypes created at NARDI Fundulea, SPL 21-2X *Helianthus maximiliani* (F₂ generation), SPL 14-3 and 17-3 P. sint, was resistant at races of downy mildew present in this area. These three sunflower genotypes represent sources of resistance at pathogen *Plasmopara halstedii*.

Keywords: downy mildew, sunflower genotypes, differential lines.

MOLECULAR DIAGNOSIS OF PHYTOPLASMA IN THE WILD TOMATO SPECIES *SOLANUM HABROCHAITES*

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Phytoplasmas are microorganisms without a cell wall that colonize the phloem of many crops. Diseases of plants caused by phytoplasma lead to decreased yields. In the Republic of Moldova, phytoplasma mainly affects tomato and grapevine. Phytoplasmosis of local tomato is associated with the presence of the pathogen ‘*Candidatus Phytoplasma solani*’. The infection causes dwarfism of the plants, the sepals are completely joined, the petals are green, the flowers are often sterile, and the fruits, if formed, are small. All this leads to a decrease in agricultural production up to 70-100%.

The aim of the research was to test the presence of ‘*Ca. P. solani*’ in the wild tomato *Solanum habrochaites*. Molecular diagnosis of this pathogen was made during two years, 2019 and 2020. The determination of phytoplasma was carried out on 10 individual plants in both years. In 2019, the analysis of the presence of phytoplasma was made during the entire vegetation season: in July, August and September. In 2020, this research was realized in August and September. DNA was isolated using express method by boiling of thin sections of tomato peduncles in alkaline solution. Nested-PCR analysis was made using specific to ‘*Ca. P. solani*’ chaperonine primers. Molecular diagnosis of plants of the wild tomato species *Solanum habrochaites* did not reveal the presence of the ‘*Ca. P. solani*’ during both growing seasons of the study: neither in 2019 nor in 2020. This result in the lack of ‘*Ca. P. solani*’ in the plants of the wild species in conditions of both years allowed to assume the high resistance of *Solanum habrochaites*. To evidence this, it was necessary, firstly, to confirm the presence of ‘*Ca. P. solani*’ in the tomato field and, secondly, to compare data obtained on the wild species *Solanum habrochaites* with the results of the diagnostics of phytoplasma spread in plants of the species *Solanum lycopersicum*. So, plants of four local varieties of *Solanum lycopersicum* were analyzed at the end of the growing seasons (mid-September) of 2019 and 2020. The scheme of molecular procedures was the same as for the wild species. It was found that the total proportion of plants affected by phytoplasma in the four tomato varieties in 2019 was significant and reached 69.6%. On the contrary, the percentage of infected plants of these four varieties of tomato in 2020 was lower, amounting to only 14.5% at the end of the growing season. Thus, a different level of phytoplasma distribution was registered in *Solanum lycopersicum* plants in 2019 and 2020. Plants of *Solanum habrochaites* were resistant to phytoplasma in both years of the study, manifesting the absence of the infection. Resistance of this wild species to ‘*Ca. P. solani*’ may be due to some morpho-physiological characteristics. First of all, a dense pubescence of stems and leaves of *Solanum habrochaites* is a mechanical barrier for insect vectors. In addition, the eventual production of plant-specific repellents could prevent the infestation through insects. Therefore, *Solanum habrochaites* can be used by breeders for comparative evaluation and creation of tomato varieties and hybrids resistant to phytoplasma.

Keywords: *Solanum habrochaites*, pathogen, Nested-PCR analysis, varieties.

THE GROWTH AND FRUCTIFICATION OF APPLE VARIETIES DEPENDING ON VARIETY AND THE PRUNING SYSTEM

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The aim of the research: to find ways to increase the productivity of new apple tree varieties and of fruit quality by developing new methods of tree crown maintenance and pruning. Apple trees, aged 11-12, of Gala Delicious, Golden Delicious and Granny Smith varieties, grafted on M9 rootstock, planted in the meadow of the Dniester River, namely at the Spica-N Agro Farm Cooperative, in the village of Onitcani, the district of Criuleni, served as the object of the research. The orchard had a planting density of 2857 trees/ha.

Two apple tree pruning systems were used during the research period, namely in G1 (the control group) – the maintenance and thinning pruning; G2 – the maintenance and thinning pruning without the shortening of main branches. The experiment was carried out using four groups of eight trees each. The soil was artificially sown with grass between the rows and ploughed on the rows of trees. The pruning of trees was carried out according to the scheme of the experiment, i.e. pruning that permit to keep the trees in physiological balance of growth and fruiting. The fight against diseases and pests was carried out according to the guidelines issued by the Chisinau Station for Plant Protection and via direct observations made at the weather station directly in the orchard. Foliar application of urea and some micronutrients to fruit trees, i.e. foliar fertilization, as well as balanced fertigation using the drip irrigation as needed, were regularly carried out in the orchards during the vegetation period.

The thinning pruning (G1) allowed for higher annual growth as compared with the maintenance and thinning pruning without the shortening of main branches (G2). If the main branches are not shortened (G2), but the length of the annual branches decreases, the crown of the tree develops in a balanced and natural way along the height of the tree and are able to form optimal crops for such orchards in a super-intensive system. Leaf area per tree and per unit area was almost the same for both trees pruning methods. The difference in leaf area, depending on the pruning method used, consisted in an increase in leaf area on shoots in G1 and an increase in leaf area on fruit formations in. The Gala Delicious, Granny Smith and Golden Delicious varieties, grafted on the M9 rootstock, were productive; after 11-13 from their planting, they produced between 19.6-48.2 t/ha in 2020, and between 51.0 - 65.4 t/ha in 2019. The Granny Smith variety showed a higher weight of the fruit as opposed to the Gala Delicious and Golden Delicious varieties. The firmness of the fruit, the titratable acidity and the dry soluble matter in the varieties studied are indicators that depend on the biology of the variety and less on the agrotechnics used in the orchard.

The maintenance and thinning pruning without the shortening of main branches, exerted a major impact on the increase of the number of flower buds, the obtaining of stable yields of 30-40 t/ha, and the generation of a profit from the sale of production of over 100,000 lei/ha with a profitability of about 150%.

Keywords: productivity, varieties, fruit quality.

THE PRODUCTIVITY OF APPLE ORCHARDS DEPENDING ON VARIETY AND PRUNING SYSTEM

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The aim of the research was to find ways to increase the productivity of apple orchards by developing methods for pruning trees that maintain a physiological balance between growth and fruiting. The research was conducted in the experimental apple orchard of the Spica-N Agro Farm Cooperative in the village of Onitcani, the district of Criuleni. The orchard was planted in 2009 with apple trees of Gala Delicious, Golden Delicious and Granny Smith varieties, grafted on the dwarfing rootstock M9. The orchards were laid out with 1m between trees in the row, and 3.5 m between rows. The tree crowns had the shape of an improved slender spindle.

During the experiment, two pruning methods were studied: G1 – the maintenance and thinning pruning (the control group); G2 – the maintenance and thinning pruning without the shortening of main branches. The experiment was performed using four groups of eight trees each. The maintenance and thinning pruning (G1 – the control group) was performed according to the methodical guidelines on modern orchards in the Republic of Moldova. The care and phytosanitary protection of the trees were carried out in accordance with the provisions of the technology for growing apple trees. The soil in the orchard was maintained by its artificial grassing and the utilization of herbicides. Foliar application of urea and some micronutrients to fruit trees, i.e. foliar fertilization, as well as fertigation using the drip irrigation as needed, were regularly carried out in the orchards. The apple trees of the studied varieties reached a maximum yield of 18-20 kg/tree in 2019. The trees in G2, were used, produced a higher crop as compared with the control group. When the Gala Delicious apple trees reached the age of eleven, they produced 60.9 t/ha in G1 and 65.4 t/ha in G2. The same legitimacy was registered for the Golden Delicious and Granny Smith varieties, i.e. the harvest increased when the maintenance and thinning pruning without the shortening of main branches were used. In 2020, the fruit harvest decreased considerably, as opposed to the previous year, and was only between 19.6-48.3 t/ha. The Gala Delicious and Golden Delicious varieties produced higher crops in contrast with the Granny Smith variety. The maintenance and thinning pruning without the shortening of main branches also increased the fruit production by 5-9 t/ha as compared to the maintenance and thinning pruning used in the control group. In 2021, the fruit yield was influenced not only by the pruning system, but also by the climatic conditions during the flowering of the trees. The Gala Delicious variety produced between 35.9-38.8 t/ha, the Granny Smith variety – between 39.2-42.7 t/ha and the Golden delicious variety – between 27.1-28.3 t/ha.

In conclusion, it can be emphasized that the maintenance and thinning pruning without the shortening of main branches of the apple trees of the Golden Delicious, Gala Delicious and Granny Smith varieties, grafted on the semi-dwarf rootstock M9, is a technological process that is worth using in modern apple orchards.

Keywords: apple orchards, productivity, growth, varieties.

EVALUATION OF RESISTANCE TO TOXIGENIC FUNGI IN SEVERAL MAIZE INBRED LINES

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Maize is attacked by a wide range of pathogenic fungi that cause devastating diseases and lead to significant yield losses. Precise identification of fungi in agricultural products is the key aspect of any pest prevention and management program, providing essential information for crop health and food safety. The aim of this paper was to evaluate new maize genotypes based on resistance to fungal phytopathogens. It allows creating the specialized collection within the active corn collection of the Laboratory of Plant Genetic Resources of IGPPP.

Twelve new inbred lines obtained from synthetic populations were used in the study. The collection samples were evaluated in the field under climatic conditions of 2021. During harvesting, the number of cobs with symptoms of *Fusarium* spp. infection was determined. The analysis of the obtained data revealed a genotypic difference in the degree of attack and the intensity of disease development. The rate of plants infected with *Fusarium* ear rot ranged from 0 to 100% depending on the genotype. The mean value of the infection rate was 26.3%. The maximum level of plant damage (70% and 100%) was recorded in MAN2425 and MAN2448 respectively, which allowed them to be classified as highly susceptible. Four lines with an infection rate of 30 to 50% (MAN2424, MAN2461, MAN2281 and MAN2451) were considered as the moderately susceptible group. Genotypes with a disease prevalence of less than 30% were classified as poorly susceptible (MAN2308, MAN2459, MAN2526, MAN2413, MAN2452 and MAN2414). Molecular identification of fungi was performed using nested-PCR with a set of primers developed in the Molecular Genetics Laboratory based on genomic sequences, associated with the synthesis of trichothecenes, aflatoxins and fumonisins from the GenBank database. Positive amplification was marked by a band of 152 bp for *A. flavus*, 120 bp for *A. parasiticus*, 147 bp - *F. graminearum*, 179 bp - *F. verticillioides*, 123 - *F. proliferatum*. Thus, all five species of toxigenic fungi were identified in MAN2459 and MAN2461 kernels. *F. proliferatum* was absent, and only *A. flavus*, *F. graminearum* and *F. verticillioides* were identified in MAN2451 kernels. The toxigenic race of *F. graminearum* was absent in MAN2308 and MAN2526 kernels. In two lines (MAN2425 and MAN2452) only toxigenic *A. flavus*, *F. verticillioides* and *F. proliferatum* were identified. Only *F. graminearum* was absent in the MAN2414 line, and in the case of the MAN2413 genotype two *Fusarium* species were absent. *F. graminearum* and *F. proliferatum* were absent in MAN2424 kernels, as well as four species of toxigenic fungi were identified in MAN2448 inbred line.

The most infected corn lines were MAN2459 and MAN2461, in whose kernels 5 species of toxigenic fungi were detected. And the least infected was the MAN2452 line - only *F. verticillioides* was identified in grain samples.

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Keywords: *Fusarium* spp., inbred lines, degree of attack, maize, nested-PCR.

PERFORMAND NEW VARIETIES OF *SALVIA SCLAREA* (CLARY SAGE)

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In Moldova, the varieties of hybrid origin of *Salvia sclarea* L., which bloom and produce raw materials and essential oil in the first year of vegetation and the plantations can be exploited three years, are created and homologated. The varieties are distinguished by a high production of raw materials and essential oil, different periods of technical maturity, resistance to drought, frost and wintering. The specificity of the research consists in the creation and implementation of the varieties resistant to frost, winter, diseases, performances that ensure a high productivity and a high quality, thus diminishing the negative consequences of climate change. In the first year of vegetation, the varieties *Ambriela* and *Parfum Perfect* form floral stems which are from 109.5 cm to 120.6cm tall, respectively, with long inflorescences (52.7 to 58.8 cm), compact, with a number of ramifications (14.5 to 18.4). These characters favor the yield structure; contribute to the synthesis and accumulation of large amounts of essential oil. The content of essential oil in the first year of vegetation has values between 1.037% (dry matter) and 1.636% (dry matter). In 2020, under conditions of severe drought, the tested varieties, that were in the second year of vegetation, synthesized and accumulated a high content of essential oil, from 1.320% (dry matter) – the variety *Ambriela*, up to 1.424% (dry matter) the variety *Parfum Perfect*. The quality of essential oil is excellent due to the high content of linalyl acetate and sclareol. The variety *Ambriela* is a complex hybrid with constant heterosis. distinctive by: Physiological properties: Very good resistance to wintering; very high resistance to drought; Resistant to foliar diseases and root system diseases. Quality properties: Essential oil content: First year of vegetation: 0.353% (standard humidity, 70%); 1.175% (dry matter); Second year of vegetation: 0.335% standard humidity; 1.185% dry matter. Major components in essential oil: Linalyl acetate 61.06%, linalool, 8.59%, sclareol, 5.25%.

The variety of *Salvia sclarea* L. *Parfum Perfect* is a triple hybrid with a constant effect of heterosis on a number of quantitative characters, including the essential oil content. Productivity in 2 years of operation of the plantation: inflorescences: 26.9 t/ha (13.8 t/ha, first year; 13,1 t/ha, second year); essential oil content 1.267 -1.424% (dry matter). The productions of essential oil constitute 73.5 kg/ha (8.7 kg/ha, year I; 64.8 kg/ha, year II). Efficiency: 3.8 kg of essential oil per ton of inflorescences.

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Keywords: *Salvia sclarea*, hybrid, quantitative characters.

PECULIARITIES OF STORAGE PROTEIN POLYMORPHISM IN THE ENDOSPERM OF MUTANT MAIZE LINES

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The scientific and research school of Professor Andrei Palii vividly illustrates the effectiveness of using endosperm genes in heterosis breeding of *Zea mays* L. that improve the quality of maize grain. The high-lysine hybrids obtained at the State Agrarian University of Moldova (SAUM) were the optimal model for the development of a new research vector related to the production of tetraploid maize forms containing the opaque-2 (*o2*) gene in their genotype. A new reserve for expanding the spectrum of genetic variability in terms of protein metabolites of maize grain quality was experimentally demonstrated. In the period of 2015-2018 a study was carried out at the heterozygous level of the effect of the influence of polyploidy on the phenotype and expression of specific genes in grain according to the end products of protein metabolism. However, for maize, the success of heterosis breeding for quality is determined primarily by the use of specific parental lines. In the direction under discussion, these are lines that combine gene and genomic mutations, the study of which requires the use of modern methodological tools. Therefore, the main goal of the presented work was to study the features of polymorphism of storage proteins (zeins) of gene (*o2* gene) and genomic mutant (tetraploid) maize lines. 32 maize genotypes, created on the basis of 10 maize lines according to the principle of "unity of differences" (*o2*/normal; 2n/4n), were used as germplasm. The studies were carried out using the storage protein electrophoresis method according to the national standard SM-2003, the FOREZ-2 computer program, the technique of direct and reverse marking of gene and genomic mutations, as well as the principles of quantitative analysis of the mutational effect on zein polymorphism. It has been established that the use of automatically synthesized electrophoretic (EF) spectra from matrices of protein profiles in reciprocal combinations makes it possible to study and interpret the effects of enrichment and elimination of the action of the *o2* gene and colchicine polyploidization on the molecular forms of zein (MPS). The predominant effect of the eliminating (reducing) action of the *o2* gene and the enriching effect of colchicine polyploidization on the zein polymorphism of the endosperm of the studied maize lines was revealed. The prevailing elimination effect of the *o2* gene on the zein fraction of the grain was established: it is most pronounced in the EF zone of medium migration. The elimination effect of colchicine polyploidization according to the protein profiles of the EF spectra is also most clearly manifested in the zone of average migration of molecular forms of zein (MFZs) markers. This fact is of considerable interest for future research in the field of selection at the level of protein marking of maize source material for mutational selection.

Keywords: *Zea mays* L., polymorphism, zein, *o2* gene, metabolism.

УСОВЕРШЕНСТВОВАНИЕ СИСТЕМЫ МЕРОПРИЯТИЙ ПО ЗАЩИТЕ ЗЕРНА ОТ ЧЛЕНИСТОНОГИХ В БЕЛАРУСИ

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В Республике Беларусь предпосылками для распространения вредителей запасов в техноценозах зернохранилищ являются потепление климата, наличие зернохранилищ разных конструкций, ограниченный ассортимент препаратов. В связи с этим усовершенствована система мероприятий по защите зерна от членистоногих.

Исследования, проведенные в 2019–2020 гг., показали, что в складских помещениях распространение получили 17 видов вредителей из отрядов Жесткокрылые (*Coleoptera*), Сеноеды (*Psocoptera*), Акариформные клещи (*Acarina*).

Система включает два этапа: подготовка складских помещений до загрузки и защита сельскохозяйственной продукции во время хранения. Она базируется на рациональном использовании препаратов различными способами (влажная и аэрозольная обработки, фумигация) с учетом фитосанитарной ситуации по видовому составу, структуре доминирования вредителей, конструкции (герметичности) зернохранилищ, целевого назначения и способа хранения продукции, температурно-влажностного режима, механизма действия препаратов с антирезистентной направленностью.

Влажная обработка инсектицидами и инсектоакарицидами из химических классов фосфорорганические, комбинированные препараты и пиретроиды обеспечивала биологическую эффективность от насекомых и клещей на уровне 83.9–100 %. Эффективность аэрозольной обработки зависела от способа хранения сельскохозяйственной продукции (в мешках, в насыпи). Фумигация определялась выбором препаратов, нормы расхода, экспозиции с учетом температурного режима. Биологическая эффективность против вредителей составляла 80.5–100 %. Согласно антирезистентной стратегии препараты следует чередовать с учетом чувствительности популяций вредителей к токсикантам.

По результатам проведенных исследований разработана блок-схема мероприятий по защите зерна от вредителей запасов, позволяющая оперативно принимать решения.

Keywords: членистоногие, зерна, фумигация, инсектоакарицид, инсектицид, защитные меры.

IMPACT OF ENVIRONMENTAL CONDITIONS ON THE PRODUCTIVITY OF SUNFLOWER HYBRIDS

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In the face of climate change, the risk of decreasing agricultural production, including sunflower seeds, is increasing. An important role in mitigating climate risks is reserved for the creation, evaluation, and introduction of drought-resistant hybrids. Given that the productivity and quality of the sunflower crop are strongly influenced by genotype, environmental conditions, and their interaction. The aim of this study was to evaluate how sunflower production is affected by the Genotype-Environment interaction.

For this, in the period 2015-2020, 21 sunflower hybrids were studied in five localities on the experimental sectors of the State Commission for Testing Plant Varieties located in different agro-climatic areas of the Republic of Moldova such as Visoca, Pelenia, Băcioi, Grigorievca, and Svetlii.

During the investigated period, two of the investigated years 2015 and 2020 were characterized by droughts of different intensities. If in the dry year 2015 the small amount of precipitation during the vegetation period is partially supplemented by the precipitation in the cold season before the growing season, in 2020, the effects of drought were more pronounced due to insufficient rainfall in the cold season, and higher thermal average.

The best conditions for sunflower growth were in 2017, when an average harvest of 3.36 t / ha was recorded. In 2015 and 2020, the average harvest was 2.99 t / ha and, respectively, - 2.81 t/ha, decreasing by about 13% compared to the average multiannual data.

The two-factors variance analysis identified the relative contributions of the specific climatic conditions of the year on productivity, regardless of the agro-climatic zone of which the given location is part. The average in all five locations of the relative contributions of meteorological conditions of the year in the yield variance is about 38%. It should be noted that the relative contributions of genotype & year interaction in some localities such as Visoca and Svetlii exceeds 50%. The relative contributions of the “growing year” factor varies from 51,4% in the North to 26,4% in the South, and the relative contributions of the “location” factor varies from 68.3% in 2020 to 25.3% in 2016. The relative contributions of the „genotype” factor contribution is usually less than 20% in most locations.

So, the meteorological conditions of the location or the growing season and their interaction with the genotype, are the main sources of crop variation.

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Keywords: hybrid, productivity, sunflower, drought, growing.

EVALUATION OF PERFORMANT HYBRID HYBRIDS IN DIFFERENT YEARS OF VEGETATION

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The creation and evaluation of prospective hybrid genotypes of *L. angustifolia*, the selection of high-performance hybrids with different maturation terms, which would harmoniously combine special indices of the main biological and production characters as well as increased essential oil content, is of major importance. Research includes prospective hybrids, with remarkable quantitative traits for obtaining new lavender clone varieties.

The basic objective of lavender breeding is to create hybrids with high essential oil content in the raw material. This index varies from 4.022% (dry matter) to 6.042% (dry matter) in the hybrids from the early maturation group, from 4.042% - 6.476% the semi-early hybrids are characterized and from 4.019% to 6.012% - the late group. The most promising prospects for essential oil content were studied during the years 2019-2021. From the set of studied hybrid genotypes, with high essential oil content, 2 genotypes from the maternal forms (Fr.1, Fr.5) and 4 descending hybrids from the maternal forms Fr.8 and Cr.13 were manifested. The essential oil content in the fourth year of vegetation (2019) for lavender F1 hybrids is from 3.988% to 6.344%. Values clearly higher than this character were recorded by the hybrids 1Fr.1-3-2-30V- 5.101% (dry matter), 14Fr.8-5-15-18V-5.453% (dry matter), 4Fr.5S-8-54-5 -5.212% (dry matter), 7Cr.13S-6-12-27- 5.226%, 7Cr.13S-6-12-28-6.344% (dry matter). The content of essential oil in inflorescences in the maternal forms is significantly lower and constitutes 2.508%, 2.600% 2.978% and 3.435% (dry matter) respectively at Cr.13; Fr 5; Fr.1; Fr.8. The variation of the essential oil content in the hybrids evaluated in the fourth year (2020), registers values from 3.988% to the hybrid 4Fr.5S-8-54-10 to 6.344% (s.u.) to the hybrid 7Cr.13S -6-12-28. Maternal forms have accumulated a lower essential oil content: Cr.13 (2.508%); Fr.5 (2.600%); Fr.1 (2.978%) and Fr.8 (3.435% dry matter). From the group of hybrids studied in the 5th year of vegetation (2021), six genotypes accumulated essential oil content higher than 5% due to the favorable climatic conditions, which contributed to the synthesis and accumulation of the essential oil.

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Keywords: *L. angustifolia*, hybrid, quantitative traits, climatic conditions.

**LEUCOJUM AESTIVUM (AMARYLLIDACEAE) – NEW SPECIES FOR
THE LOWER PRUT FLORA**

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The humid zone of the Lower Prut Lakes Ramsar site is represented by typical plant communities of the floodplain, the preservation of which was facilitated by the strict regime of the border zone maintained in previous decades. The main types of vegetation are azonal communities: floodplain forests, which occupy most of its area, and lowland meadows, located in narrow fragments among trees and shrubs.

On the territory of the forest stand, in places prone to prolonged flooding, forests dominated by *Salix alba* and are most often represented by communities of *Salicetum rubosum*, in which an admixture of *Populus alba*, *Ulmus laevis* and some others. It is also characterized by an unstable moisture regime, weak density and few types of grass cover. The unevenly developed shrub layer contains: *Swida sanguinea*, *Frangula alnus*, *Ligustrum vulgare*, *Rhamnus cathartica*, *Crataegus monogyna*, *Euonymus europaea*, *E. verrucosa*. In the grass cover, *Rubus caesius* is among the most widespread species, growing abundantly in places; *Anthriscus sylvestris*, *Potentilla reptans*, *Phragmites australis*, *Ranunculus repens*, *Galium aparine*, *Poa pratensis*, as well as *Poa palustris*, *Aegopodium podagraria*, *Glechoma hederacea*, *Lysimachia nummularia*, *Symphytum tauricum*, *Myosoton aquaticum*, *Humulus lupulus*, *Equisetum arvense* are more common than others. On wet depressions under the canopy, there are spots of a rare, new for the territory species – *Leucojum aestivum* L., belonging to Amaryllidaceae family. *Leucojum aestivum* is a perennial, bulbous plant. Blooms in April-May and fructifies in June-July. Propagates by seeds and bulbs. The species is decorative and medicinal.

This Critically endangered in the Republic of Moldova vascular plant was registered in April 2022 in the outskirts of Crihana Veche village, Cahul district. The exact location of the population is – west of the village: N 45° 50' 09", E 28° 07' 42". The population covers approximately 50 m², average number of vegetative specimens is 5-8 and flowering plants are 1-2 per square meter.

This finding is new for the territory of the republic, whilst so far two locations are known: in the valley of the Prut river in the vicinity of the commune of Cioara (Hîncești district) and village Sărata-Răzeși (Leova distr.). The species is located at the north-eastern limit of its spreading area. Outside the country it is met in the Mediterranean region, the Caucasus, the Atlantic and Central and south-eastern Europe.

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Keywords: *Leucojum aestivum*, floodplain, population, preservation, medicinal specie.

REINTRODUCTION OF *CRAMBE TATARIA* IN THE „LOWER PRUT LAKES” RAMSAR SITE

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Spontaneous vascular plants in Republic of Moldova are under threat with significant loss of plant species and habitat taking place. Local and regional activities on reintroduction of rare and threatened vascular plants in our republic has been started in recent years. The repopulation programs in natural habitats of threatened with extinction species of Community interest and included in international conventions, a rare species in the republic – *Crambe tataria* Sebeók (Brassicaceae), was chosen as one of the objects for study, reproduction and repatriation to nature.

This species, in the region, is protected by the state (category II – endangered), included in the 3rd edition of the Red Data Book of the Republic of Moldova as threatened species (category EN). Internationally protected by the Berne Convention and included in Appendices II and IV of the Habitats Directive. The species is also rare on the territory of Ukraine, where it is included in the Red Book as a vulnerable species.

Crambe tataria is a typical steppe species with a Pontic-Pannonian-Sarmatian disjointed range of distribution. In the Republic of Moldova, it occurs sporadically, except in the north-eastern and southern parts. It grows as part of the grasslands of anthropogenic variants of primary steppes, steppe and limestone slopes, glades and edges of the *Quercus pubescens* forest stands. In order to make available *ex situ* collection of *Crambe tataria* and possible for population recovery and restoration programme into natural habitats, during 2020-21 numerous areas with steppe vegetation were examined, among which we chose „Slobozia Mare – Văleni” natural steppe area.

In June 2021 a number of 15 plantlets obtained from seeds (originally collected from Bugeac steppe plants in August 2020), were planted in the sector of grassland of the

*62C0 (Ponto-Sarmatic steppes) habitat. This habitat is one of the most species-rich plants communities in Europe in terms of the plant species. The location of the sector is: N 45° 35' 30", E 28° 09' 48". The surface area represents a steppic slope of western exposition with steepness of circa 30°. The plant community is *Festucetum (valesiaca) – stipeto (ucrainica + lessingiana + capillata) herbosum*, with various herbacious vascular plants: *Thymus marschallianus*, *Salvia nutans*, *Eryngium campestre*, *Salvia nemorosa*, *Euphorbia cyparissias*, *Poa angustifolia*, *Artemisia austriaca*, *Linum austriacum*, etc.

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Keywords: *Crambe tataria*, threatened species, *ex situ* collection, plantation.

**ASSESSMENT OF THE MORPHOGENETIC AND REGENERATIVE
POTENTIAL OF TRITICALE GENOTYPES
IN *IN VITRO* CULTURE**

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The main focus of research is determined by the country's current and future need for food security. The global and regional climate, energy and food crises pose great challenges to the scientific society, especially to plant genetics and physiology, to obtain new fundamental and applied knowledge in highlighting, evaluating and directing the genetic-physiological mechanisms of the production process and ecological resistance of plants, in this case triticale, by improving adaptability to adverse climatic conditions: increasing resistance to water and heat stress and obtaining healthy yields. In recent decades, thanks to genetic advances in breeding, new varieties have been developed that are much more competitive than current cereal genotypes. These varieties are better because of their high yield capacity and useful agronomic characteristics (fall resistance, resistance to unfavourable environmental conditions, grain filling, etc.). From a technical point of view, triticale has a great capacity to adapt to the most diverse climatic conditions, which gives it undeniable advantages over other cultures that are more susceptible to natural factors. The importance of triticale to the economy is explained by the chemical composition of the grain, which gives it special properties for use in food, animal feed and industry.

The development of theory and new effective methods for plant breeding requires a deep understanding of the mechanisms that determine the spectrum of variability in genetic information. Biotechnological methods, through the application of *in vitro* culture techniques, are a reliable way to obtain pathogen-free biological material, the preservation of genetic resources and the fastest way to create and multiply genotypes. In view of these aspects, we proposed to establish methods of culture that would ensure the successful *in vitro* regeneration of agronomically valuable genotypes.

The biological material used in this study was represented by 3 genotypes of triticale: Ingen 35, Ingen 93, 188TR5021. Mature embryos including 120 embryos from each genotype were used as source material. The basic nutrient substrate was Murashige and Skoog (1962) medium modified and supplemented with L-asparagine, mesoinositol, glycine, sucrose and growth regulators: 2,4-D dichlorophenoxyacetic acid, naphthylacetic acid (NAA) and Kinetin (K), thidiazuron, BAP (6-benzylaminopurine), agar, pH 5.8.

In order to investigate callusogenesis processes, callus cultures were initiated from mature embryos in all genotypes studied. Intense differentiation of explants and the appearance of callus formation on culturing media were observed on day 4, which is in agreement with literature data. Massive callus formation was observed at day 7-8. It was found that, all genotypes tested have a fairly high capacity to induce callus. The frequency of callusogenesis varied from 80.19% (188 TR5027), 92.02% (Ingen 93 standard) and 98.45% (Ingen 35) depending on the genotype. Two types of callus formation were observed. The first type of callus is embryogenic - compact, structured, yellowish colour; the second - non-embryogenic, watery, white-yellowish colour. Despite

evidence in the literature that a non-embryogenic callus is formed first, which either remains the primary callus during culture or is transformed into an embryogenic type, we observed that the non-embryogenic callus did not form meristematic foci throughout explant culture, and no somatic embryogenesis was observed.

The frequency of morphogenesis depends on the induction of embryogenesis and rhizogenesis. Genotypes Ingen 35 and 188 TR5027 formed embryogenic callus at a rate of 34.22% - 40.15%. A low morphogenetic potential is attested by genotype Ingen 93 - 29.24%. Obviously, this is due to the peculiarities of morphogenesis, which is carried out by the organogenesis pathway, with a predominance of the rhizogenous type.

The frequency of rhizogenesis compared to embryogenesis was found to be high with an average of 57.35%. Only in 34.53% of morphogenic callus, the development was embryogenic. This denotes that, the achievement of morphogenesis is determined by the genetic and physiological characteristics of the explant.

Stable plant regeneration is a prerequisite for the practical use of *in vitro* culture. The regeneration frequency averaged 35.07%. Regeneration potential varied significantly by genotype. If for genotypes Ingen 35 (49.03%) and Ingen 93 (40.70%) then for genotype 188 TR5027 it was only 15.50%, a low regeneration rate. The number of regenerants obtained from morphogenic callus was 0.2 and 1.3 pcs per callus. This is due to the fact that a large number of morphogenic callus developed by the rhizogenesis pathway, which precludes the possibility of obtaining plants.

Analysis of variance made it possible to identify significant factors influencing callusogenesis, morphogenesis and regeneration. The frequency of callusogenesis usually depends on a number of random factors. One significant factor for this indicator is genotype. Comparing the contribution of different factors and their interaction with each other, it should be noted that genotype plays a significant role not only in the process of callusogenesis, constituting 76.04%, but also determines regeneration capacity, showing an influence power of 69.15% with significant differences for $P < 0.05$.

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Keywords: triticale, varieties, agronomic characteristics, callusogenesis, regeneration, *in vitro* cultures.

**GENETIC DIFFERENTIATION AND RELATIONSHIPS OF
OROBANCHE CUMANA POPULATIONS FROM
DIFFERENT SUNFLOWER PRODUCING COUNTRIES**

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Orobanche cumana Wallr. is an important species of obligate parasitic weed, naturally distributed from central Asia to south-eastern Europe, where it parasitizes wild *Asteraceae*. Until now, broomrape was recorded in more than 60 countries around the world, causing significant economic damage in crop production especially in Central and Eastern Europe, Spain, Turkey, Israel, Iran, Kazakhstan, and China.

Accelerating rate of parasite evolution and distribution could rapidly diminish the effectiveness of control measures and strategies related to the deployment of resistance genes. Therefore, knowledge about the population structure of parasite are critical to the design of appropriate resistance breeding programs and effective weed control.

Thus, the present study focused on the assessment of the genetic diversity among and within 33 broomrape populations belonging to different sunflower producing countries (Moldova, Romania, Bulgaria, Serbia, China, Turkey) and races (E, G, H) employing microsatellite molecular markers (SSR). The structure of populations was analyzed using different methods (AMOVA, Nei statistics, PCA), based on GenAlEx 6.503 and XLSTAT 2014.5.03 software.

The genetic differentiation (Φ_{ST}) between pairs of *O. cumana* populations with different origins indicate values between 0.170 and 0.686 ($p < 0.001$), revealing a strong and very strong differentiation between the populations from different countries. The highest differences were found between populations from China and other countries, indicating that geographic distance is a substantial barrier to gene exchange between populations. AMOVA analysis showed significant genetic differences between groups (countries), as well as between populations within groups and individuals within populations. So, 36% of total genetic diversity was attributable to divergence between groups, 30% to population differentiation within groups and 34% to individual differences within populations. The first 2 components from a PCA explained about 41% of the total variation in the data. Cluster analysis showed that the majority of populations from the Republic of Moldova, Romania and Bulgaria, which are geographically close, were included in one group, Serbian populations formed a separate subcluster, and populations from Turkey and China grouped together. As a result, new data of broomrape population structure were accumulated.

Acknowledgments: This study was supported by the research project 20.80009.5107.01 „Genetico-molecular and biotechnological studies of the sunflower in the context of sustainable management of agricultural ecosystems”, funded by the NARD.

Keywords: *Orobanche cumana* Wallr., genetic diversity, molecular markers SSR, races, population genetic structure.

VARIABILITY OF QUANTITATIVE TRAITS OF CORN HYBRIDS AND INBRED LINES UNDER DROUGHT AND SALINITY

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The variability of the quantitative characters studied at the initial stages of plant development under stressful conditions in hybrids mostly depended on the factor "genotype" ($\eta^2=73.44\%$ *** - 88.92% *** (salinity) and 51% ** - 82.9% ** (drought). The hybrids were characterized by higher values of dependence from the "genotype" factor than inbred lines. Lines A285, P346 and hybrids (with maternal genotypes - combinations of lines A285, N6, W23) proved to be the most resistant. The variability of the characters "seedling length" (LP), "root length" (LR) at the stage of immature embryos depended significantly from the factor "genotype" (inbred lines were characterized by higher values than the hybrid combinations). In hybrids the significant dependence of the variability of productivity traits on the "genotype" factor ($\eta^2=39.7\%$ *** - 60.94% ***) were found out and the highest values of these traits were recorded in hybrid combinations with maternal genotypes A285xRf7 and A285xW23. Inbred lines were characterized by the highest value of the dependence of the character "diameter of the pollen grain" from the factor "genotype" ($\eta^2=39.84\%$ ***), but hybrids had the highest value of the dependence from the factor "abiotic stress" ($\eta^2=27.7\%$ ***). The complex evaluation (at diploid and haploid level) of the variability of quantitative characters at different stages of ontogenesis (under osmotic and saline stresses) allowed the highlighting of lines (A285, P346) and hybrids (maternal genotypes A285xN6, A285xW23) with increased resistance. These genotypes can be used in selection schemes to obtain heterotic and productive hybrids. It should be noted that lines L1866, L276, W23, N6, P165, Rf7 can be used as parental genotypes in hybrid combinations with stress resistance.

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Keywords: corn, hybrids, variability, quantitative traits, drought, salinity.

**GENOTYPES OF *SALVIA SCLAREA*
ESTABLISHMENT OF ESSENTIAL OIL**

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Special attention has recently been paid to the species *Salvia sclarea* L., due to the multiple uses of the essential oil, which is contained in inflorescences and is used in medicine, the perfumery industry, the brewing industry, etc. At the Institute of Genetics, Physiology and Plant Protection, a series of amelioration researches are carried out, by genetic methods, in order to create new genotypes, which would synthesize and accumulate the highest possible content of high-quality essential oil.

The aim of the research is to evaluate and select hybrids of *Salvia sclarea* L., with high and very high content of essential oil.

The biological material used is represented by 84 F₁-F₂ of different types of *Salvia sclarea* L., in the second year of vegetation.

Phenological, biomorphological evaluations were performed according to the methods in force. The essential oil content was determined by hidrodistillation in Ginsberg apparatus.

Research has shown that the hybrids created, evaluated, are resistant to sect, frost, wintering and disease. They are valuable through a string, which ensures increased productivity and high essential oil content. In extreme conditions, the genotypes of *Salvia sclarea* have developed plants with a height of over 91 cm, in some hybrids reaching up to 133.2 cm. Large inflorescences, 42-72 cm long. The number of first-degree branches of the inflorescence ranged from 12.0 to 18.4. Second-degree ramifications - ranged from

18.0 to 44.4 at different genotypes. All these characters facilitated the synthesis and accumulation of a fairly large amount of essential oil. The determination of the essential oil content in the inflorescences, recalculated to dry matter, showed that some hybrids due to drought synthesized and accumulated a relatively low amount of oil - 0.620-0.800%. We specify that 57% are hybrids with a high essential oil content of over 1.0%, and 21.4% are hybrid genotypes with a very high content of essential oil 1,301-1,906% (dry matter).

As a result of the evaluations, 18 exceptional hybrids of the most important character were selected, identified, they synthesize and accumulate 1,301-1,906% (dry matter) essential oil.

Acknowledgments: Research was carried out within the project of the State Program 20.80009.5107.07 "Reducing the consequences of climate change by creating, implementing varieties of medicinal and aromatic plants drought, frost, winter, disease resistant, which ensures sustainable development of agriculture and guarantees high quality raw material predestined to the perfumery, cosmetics, pharmaceuticals and food industry", financed by the National Agency for Research and Development.

Keywords: *Salvia sclarea* L., hybrids, productivity, inflorescence, essential oil content.

**MANIFESTATION OF TRANSGRESSIONS ON THE TRAITS
OF SPIKE PRODUCTIVITY IN F₂ POPULATIONS
OF COMMON WHEAT**

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Transgressive segregation in plant populations is an important factor contributing to the emergence of forms with new characteristics that are superior to the "best" parent. The mechanisms of transgressions have not yet been fully established, but the phenomenon itself is widely used to identify and select plant forms with economically valuable traits that are resistant to pathogens, pests, and unfavorable abiotic factors. Our research is devoted to the study of the influence of the parental forms on the elements of the productivity of the spike of soft winter wheat using the example of 8 F₂ hybrid populations obtained in reciprocal crosses. Positive and negative transgressions were revealed for the traits spike length, number of spikelets per spike, number of grains per spike, weight of one grain, weight of the grains from one spike.

The most significant positive transgressions based on the degree (T_g) and frequency (T_f) were recorded in the case of *the grain mass* character (T_g = 9.32-20.97%; T_f = 31.67-48.33%) in the combinations of wheat Moldova 11 (M11) x Moldova 16 (M66), Moldova 16 (M16) x M11, M16 x Basarabeanca. The combinations Basarabeanca x L M / M3, L M / M3 x Basarabeanca, and of the mass were noted by relatively high indices (T_g = 6.11- 7.50%; T_f = 12.5-25.0%) *grains per spike* - combinations M11 x M16, M16 x M11, Kuialnic x M66, M66 x Kuialnic: T_g = 6.70-15.1%; T_f = 8.33-26.67%. Insignificant positive transgressions were found for the character of *the number of grains per spike* in 5 combinations, and for the number of *spikelets per spike* - only one case (T_g = 4.29%, T_f = 10.83%) in the Kuialnic x M66 combination.

According to the studied traits, genetic distances between parental forms did not affect the manifestation of transgressions. It was found that in F₂ hybrids, obtained on the basis of reciprocal F₁ hybrids, the direction of crossing influenced quite significantly the degree and frequency of transgressions in some combinations, without changing their direction. For example, the combination M16 x M11 recorded for *the mass of a grain* T_g = 11.56%; T_f = 31.67%, and M11 x M16 - T_g = 20.97%; T_f = 35.83%.

This indicates that the successful selection of crossbreeding components and revealed specificity of the manifestation of transgressions in common wheat F₂ hybrids can contribute to the expansion of transgressive variability on the elements of spike productivity in order to create valuable forms.

Keywords: common wheat, hybrid, populations, traits productivity.

TRICHODERMA SPECIES FOR PLANTS PROTECTION AGAINST ALTERNARIOIS

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Pathogenic species of *Alternaria* can infect all parts of plants. Worldwide there is an increased presence of *Alternaria spp.* mycotoxins in raw food materials and foods. To reduce their pathogenicity, it is important to use biological methods for plants protection. The aim of the research is to select strains of *Trichoderma* micromycetes for the creation of the microbial control agents against pathogenic species of *Alternaria*.

The research objects presented 20 strains of the *Trichoderma spp* fungi from laboratory's collection. The antagonistic activity of mycelial cultures of *Trichoderma* against the pathogen *A. alternata* was studied by dual culture method. The indicator of inhibition of the pathogen by *Trichoderma spp.* was calculated (%) and assessed (in points) by the degree of overgrow of the *Alternaria spp* colonies (Поликсенова В.Д., 2004). The antifungal activity of the culture liquid was evaluated by agar diffusion method. (Еропов Н.С., 2004).

On the 10th day of the experiment, two strains; 2N and 13N have given high antagonistic activity with inhibition rates of 94% and 83% respectively. The other *Trichoderma* fungi strains have given less activity with inhibition rates from 59% to 81%. The degree of overgrow of *Trichoderma spp* fungi of the colony of the pathogen for 9 strains consisted 4 points, 4 strains - 3 points, 5 strains populated the colonies by half - 2 points. The colonization of the pathogen by the *T. virens* strain CNMNEFD-13 was 25% (or 1 point) and there was no growth reduction of the pathogen by the strain 8N (0 points). At the same time, no signs of colonization of the *Trichoderma* culture by the pathogen were seen. Evaluation of the fungicidal activity of the three *Trichoderma spp* liquid cultures was performed by the agar diffusion method. All tested strains have inhibited growth of the pathogen with the formation of the sterile zones with a radius of 13-16 mm.

As a result of the research, two strains of *Trichoderma* fungi, 2N and 13N, from the laboratory's collection, were proposed for further elaboration of the microbial control agents against *Alternaria* fungi.

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Keywords: *Trichoderma micromycetes*, microbial agents, mycelial cultures, antifungal activity.

VARIABILITY SOME MORPH-BIOLOGICAL TRAITS IN BREEDING MATERIAL OF CHICKPEA

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This report presents the results of studies of the variability of some morph-biological traits of the intraspecific hybrid combination chickpea ♀ Botna (desi, brown seed coat) × ♂ Ichel (kabuli, cream seed coat) in the F₃ generation. Desi × kabuli crosses have consistently produced high-yielding progenies and have been the source of many new cultivars. Initial steps of breeding programmers are as follows: Creation of genetic variation and selection of genotypes from this bulk.

The research material was 195 individual F₃ selections from 20 offspring obtained from the 20 individual selections. The study of the material was carried out in 2021 on the experimental fields of the Institute. With regard to the characteristics of the seeds, it should be noted the formation of intermediate gulabi (or pea shaped) seeds, which accounted for about half of the selections (97). In 13 out of 15 pigmented offspring, seed segregation by pigment (brown/cream colored) was noted, the same number with seed type segregation (desi/gulabi or desi/gulabi/kabuli). In light-seed offspring, segregation is only to the type of seeds and heterogeneity to the size of the seeds.

Seed size (weight of 100 seeds) is a key determinant of genetic variation. In the aspect of the trait, the weight of 100 seeds in this hybrid combination is marked by high variability. The minimum values of the weight of 100 seeds in the progeny A7 (27.36g (25.4 – 29.24) CV=5.1), the only cream/orange-colored seeds. Maximum values in A18 offspring (34.7g (32.6 – 37.9) CV=4.3), all offspring gulabi and brown colored. Minimum variability in A9 offspring ((CV=3.3 (32.8g (31.5-34.6)). Maximum variability in A15(CV=9.1(30.92g (24.1 - 34.7) followed by A4 (CV=8.8 (30.9g (27.4 -37.5)), A10 (CV=8.4 (33.0g (28.4 -38.2) compared to Botna (CV=0.61) and Ichel (CV=0.63) Thus, for the weight of 100 seeds, there is a continuous series of variability, with the presence of positive and negative transgression, Botna var. (28.6 g/100s) and Ichel var. (32.5g/100s).

Plant height variability in progeny is within: CV= 3.7(A9) – CV= 6.5(A15), in variety Botna - CV=2.3. The minimum and maximum plant heights (mean value) are 51.4cm (A7) - 66.8cm (A18), for the Botna variety - 55.7cm, for the Ichel variety - 64.2cm. Ripening obtained offspring within July 30-31 (A7) - August 7-8 (A18). Ripening varieties Botna and Ichel was on August 2 and 6, respectively. Variability of number seeds/plant within CV=17.2 (A18) - CV=31.4(A3) and seeds weight/plant within CV=16.3(A18) - CV=31.1(A3). Maximum value number of seeds (mean) 69.8 and seeds weight /plant 23.5 g (A4).

Keywords: hybrid, variability, morphological traits, seeds, chickpea.

ANALYSIS OF MOLECULAR VARIANCE (AMOVA) OF *OROBANCHE CUMANA* POPULATIONS

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The first reports that attest the presence of *Orobancha cumana* Wallr. (broomrape) in sunflower fields date back to 1866 in Russia. Then, due to the expansion of sunflower cultivated area, in 100 years this parasite has spread in Bulgaria, Moldova, Romania, Serbia, Spain, China and in the last 50 years – in the main sunflower producing countries around the world. Until now, eight races of *O. cumana* have been identified, from A to H.

The objective of this research was to study the genetic diversity of 33 *O. cumana* populations originated from 6 countries (Bulgaria, Moldova, Romania, Serbia, China, Turkey) using microsatellite molecular markers like SSR and ISSR. The evaluations were conducted in artificially infected pots in greenhouse conditions. The most highly virulent broomrape races, G and H, was revealed in the majority of studied countries, except of Serbia (race E or less virulent than E). The study of genetic variability of 33 populations was performed based on the analysis of molecular variance method (AMOVA). The results showed for the both marker systems (SSR: within populations 37%, among populations 63%; ISSR: 41% and 59%, respectively), that the most of the total variance was attributed to the differences between populations. At the country level, AMOVA dates for Bulgarian broomrape populations also presented a higher variability among populations while for the populations from Moldova, Serbia, China and Turkey, the molecular variations within populations were a major source of genetic variation. AMOVA results showed that both marker sets, based on molecular data SSR and ISSR, keep the same tendency of distribution of genetic variations.

The molecular differences between identified broomrape races (\leq E, G, H) very little contributed to the total variability (SSR: among races 10%; ISSR: 11%, respectively). According to the SSR markers (separately for three groups of races) genetic variations within populations are higher for the broomrape populations race H (53%) and lower for those from race G (30%) and \leq E (36%). In the case of ISSR markers, molecular variations were almost equally divided between intra- and interpopulation diversity, with predominance of diversity between populations for all groups.

In this context, our results indicate that *O. cumana* specie has a very great genetic potential and a high degree of genetic variations which can determine the evolution of more virulent races which is important for creating some sunflower crop breeding strategies and their control.

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Keywords: *Orobancha cumana* Wallr., AMOVA, populations, races, genetic diversity.

ADAPTIVE CAPACITY OF THE *FAGUS SYLVATICA* L. POPULATION FROM THE SLOVAK REPUBLIC

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The European beech *Fagus sylvatica* L. is one of the most important tree species not only in the Republic of Moldova, but throughout the European continent, both economically and ecologically. Beech forests in the Republic of Moldova need to be restored with populations that are resistant to globally changing climatic conditions. In this investigation, we studied the possibility of increasing the natural adaptive potential of beech population from the Slovak Republic, Nitra region, Topoľčianky, through the use of plant growth bioregulators capsicoside and genistifolioside at a concentration of 0.001%. The gibberellic acid was used as standard regulator of plant growth. Seeds were stratified at a temperature of +4±2 °C. After the germination, they were sown in solarium with drip irrigation, but without regulation of the air temperature, which in summer rose above +35 °C, sometimes reaching +55 °C. The viability of *Fagus sylvatica* seeds collected in autumn 2019 averaged 65.6%. Treatment of seeds with solutions of capsicoside and genistifolioside before germination leads to stimulation of daily seed germination by 18.5-22.2% and to significant reduction in the period of total seed germination by 20-22 days. As a result, 87.01±0.03% of seeds germinated in the experiment with bioregulators, which was significantly higher than in the control and standard. The adaptation of germinated seeds to solarium conditions after treatment with bioregulators was 2.5-2.9 times higher than in the standard and 3.0-3.3 times higher than in the control. The survival of seedlings in experimental variants at supraoptimal temperatures was on average 2.7-3.0 times higher than in the standard and control. Beech seedlings treated with bioregulators had the leaves with significantly bigger leaf surface area, the leaves were larger by 2.3±0.7 cm and wider by 0.8±0.4 cm that contributed to the greatest photosynthetic process. The relative chlorophyll index in the phase of three pairs of true leaves of seedlings in the variants capsicoside and genistifolioside 0.001% was 144.9±3.5 and 145.6±3.7 g/m², respectively, which significantly exceeded the control (135.7±3.2 g/m²) and the variant with gibberellic acid (121.5±2.9 g/m²). Two-year-old seedlings were transferred from the solarium to natural conditions, in mixt hornbeam/beech forest of the “Plaiul Fagului” Scientific Reservation. The use of plant growth bioregulators led to a high degree of adaptability of seedlings, 90.17% of transferred plants survived.

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Keywords: *Fagus sylvatica* L., adaptive potential, germination, viability, bioregulators.

**EVALUATION, ESTIMATION AND MONITORING
DIABROTICA VIRGIFERA VIRGIFERA LE CONTE**

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In the Republic of Moldova, corn (*Zea mais*) is one of the most important cereal crops for the food industry and feed reserves. Annually, this crop occupies an area of over 270.0 thousand ha, which constitutes approximately the 5th part of the area of arable land. The emergence of one of the most dangerous corn pests, such as the western corn rootworm (*Diabrotica virgifera virgifera* Le Conte) DVV, which has already managed to spread throughout the country, creates a real danger to corn production and the country's economy in general. Although, in the countries of the European Union, DVV was considered a harmful organism, regulated with a state of quarantine, but its spread could not be prevented. A number of recommendations have been developed based on the application of crop rotation, adequate monitoring of pest populations and other relevant measures to prevent the spread of harmful organisms and control measures.

Traps with synthetic sex pheromone have proven to be an effective tool for both monitoring the pest population (accumulating specimens for study) and a total control method, such as sterilization and mass capture. The active component of the sex pheromone of DVV *Diabrotica virgifera virgifera* Le Conte, it is 1,7-dimethylnonyl propanoate. The optimization of the synthesis scheme, the obtaining of the high purity substance as well as its analysis, is important and necessary.

Subsequently, the synthesized substance was impregnated on synthetic rubber septa with a dose of: 1 mg per dispenser. DVV synthetic sex pheromone dispensers were fixed on the "open-type" trap - a piece of laminated cardboard with entomological glue and then attached directly to the plant, in the corn field, with a distance of about 20m between them and further verification were made 1 -3 times a week. As usual, the first catches of the imago phase of the pest were recorded in the second half of June. The peak of the pest's flight falls in mid-July, with a considerable decrease in the number of pests caught by mid-August.

The economic damage threshold is 5,5 imago per plant. In the northern part of the country, in the village of Ocnita, the village of Girbova, the pest exceeded 7 times the economic threshold of damage, which is explained by the relief shape of the field, the rotation of crops and the location of the corn field compared to others. Pests have been recorded in technical corn and seed fields, where the principle of monoculture is followed. The effectiveness of protection measures is hampered by high pest migration capacity and an effective control scheme.

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Keywords: *Diabrotica virgifera virgifera*, crop rotation, corn, 7-dimethylnonyl propanoate.

THE DEVELOPMENT OF VITICULTURE THROUGH THE REQUIREMENTS PRISM OF GREEN ECONOMY

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The primary imperative of the sustainable development of the wine sector is to obtain high production, with low consumption of resources, in conditions of increased economic efficiency and the use of technological links that contribute to reducing energy consumption. High quality wine derivatives can be ensured, if three main factors are taken into account: the genotype (variety), location of the plantation (soil and climatic conditions) and applied technology (cultivation and processing).

The intraspecific genotypes have a wide capacity for use, but at the same time do not ensure the overcoming of climate change barrier. That is why, taking into account the functionality of genotypes and the use of technical algorithms and interspecific hybridization methods, more plastic rhizogenic interspecific genotypes should be created in terms of their adaptation to climate change, with beneficial repercussions on the sustainable development of the wine sector. As a result of crossing *V. vinifera* L. with *M. rotundifolia* Michx. were obtained and identified interspecific vine genotypes that allow the expansion of the vine cultivation area to the northern areas, while reducing the number of chemical treatments, which will contribute to obtaining ecological products and will improve environmental protection. The rhizogenic interspecific genotypes have an early period of grape ripening, can be multiplied by cuttings, without grafting, thus obtaining rhizogenic propagating material that contributes to reducing the costs of setting up vineyards.

They were approved as table grape varieties like: "Malena", "Nistreana" and "Algumax" and grape varieties for fresh consumption and processing: "Augustina", "Alexandrina" and "Amethyst". By creating plantations, will contribute to the extension of the area to the northern limit of vine cultivation.

Keywords: area, genotype, green economy, viticulture.

MOLECULAR ASSESSMENT OF *F. GRAMINEARUM* IN SEVERAL MOLDAVIAN MAIZE GENOTYPES

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Fusarium graminearum is a typical pathogen of many cereal crops, causing grain, stalk and seedling rots. In maize, this pathogen causes Giberella ear blight and is able to infect kernels through the stigma. Optimal conditions for the development of the fungus include moderate temperatures around 25°C and high humidity, and the species is distributed throughout the maize cultivation region. It has been established that some strains of *F. graminearum* can synthesize DON mycotoxin, which is highly toxic and provokes alimentary-toxic aleukia. Non-compliance with the rules of corn agrotechnology, in particular, non-compliance with crop rotation, insufficient tillage, too high planting density, irrational use of fertilizers and pesticides can induce DON synthesis in toxigenic *F. graminearum* strains.

Molecular diagnostics of toxigenic strains of *F. graminearum* was carried out in kernel samples of several local maize genotypes grown under field conditions in the Republic of Moldova. The fungus was identified using the nested-PCR method with primer pairs based on the *F. graminearum* genomic cluster of the fungus involved in DON synthesis (*Fusarium graminearum* isolate 23-4 Tri core gene cluster, complete sequence). External pair: ftri8gr1 (forward) - CTCCGGTAATGTTTCTCGTCACT, ftri8gr4 (reverse) - CGCTGCTGAGGGTTTTACCAT. Internal pair: fqtri8gr2 (forward) - CTCGTCACCTTCCTTGATGACACA, fqtri8gr3 (reverse) - GGGGGCCGACATTCACTTC. Comparison of the frequency of infection of the studied samples with the control genotype B73 was carried out using the Student's test for two independent samples ($p < .05$).

In 2020, there was a decrease in the frequency of infection by the fungus for all genotypes, which was due to high temperatures and low rainfall in the spring-summer period. On the contrary, in 2021, which was characterized by moderate temperatures, high air humidity and heavy rainfall during the period from April to July, an increase in the infection rate of *F. graminearum* in corn kernels was observed. The highest mean frequency of infection was noted for kernels of the CP137 and CP148 genotypes. At the same time, KU123 and MK01 genotypes showed the lowest frequency of infection with

F. graminearum; the average infection frequency during the observation period did not exceed 30%. A statistically significant lower infection frequency was observed for genotypes KU123 ($t=2.38$, $p=.02$) and MK01 ($t=2.84$, $p=.01$), while for CP137 and CP148 there was no significant difference in the frequency of *F. graminearum* in comparison with the susceptible line B73. Thus, the locally bred genotypes MK01 and KU123 are of interest for breeding programs aimed at obtaining maize lines and varieties with increased resistance to *F. graminearum*.

Keywords: *Fusarium graminearum*, genotype, pathogen, nested-PCR, frequency of infection.

ASSESSMENT OF GERmplasm DIVERSITY USED IN THE MAIZE INBRED LINES DEVELOPMENT

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Diversity of germplasm sources is the raw material for breeding programs to permanently improving of agronomic performance and reduce plant vulnerability to various pathogens and pests. In hybrid maize breeding, information on germplasm genetic diversity and heterotic groups is very useful in inbred line development and planning crosses for hybrid cultivar development.

The purpose of present investigation was to estimate the phenotypic and genetic diversity of 10 maize inbred lines from the main germplasm groups, which are currently used in maize breeding programs. Phenotypic diversity was estimated by determining the phenotypic diversity index (*idf*), calculated based on 28 quantitative and qualitative characters. Genetic diversity was assessed by the mid-parent heterosis (H, %) and SCA constants (\hat{s}_{ij}) for grain yield.

The high level of phenotypic similarities, confirmed by the low values of the *idf* index were noticed between the A654 and Co125 inbred lines (*idf* = 5.2), W153R and MK390 lines (*idf* = 5.3); Co125 and F2 lines (*idf* = 5.7), MK01 and A632 inbreds (*idf* = 5.9). Among the other inbred lines, a high level of diversity was attested (*idf* > 7.0).

The high values of reproductive heterosis (H = 57.6-182.6%) noted in the crosses between inbred lines of different origin, were demonstrated their high diversity at the genetic level and confirm their different genealogical origin. The most accentuated genetic similarity was noted between the A632 and MK390 inbred lines (H = 57.6%), being demonstrated the affiliation of the MK390 line to the BSSS germplasm group. The high values of heterosis, observed in all crosses obtained with the F2, CM7 and MO17 inbreds reveal their significant differences from the other genotypes used in the study, which confirms their belonging to distinct sources of germplasm. A high degree of similarity at the non-additive gene interactions level was attested between the CO125 and P502 lines (\hat{s}_{ij} = -0.34 t / ha), A632 and MK390 inbreds (\hat{s}_{ij} = -0.31). Significant positive values of SCA constants recorded at all crosses made with Mo17 and F2 inbred lines were confirmed their genetic distance from other genotypes.

The generalized results regarding the diversity assessment methods highlighted the essential phenotypic and genetic differences of the Mo17, F2 and CM 7 inbred lines compared to the other genotypes. Therefore, these lines represent distinct groups of germplasm. Thus, the data allow the classification of the studied inbred lines in the following global germplasm groups: Lacaune (line F2), Lancaster (MO17), Iodent (MK01), Reid (A632, A654, BC27D4, CO125, P502, MK390) and Ottawa Flint (CM7 line).

These results will be useful in the systematisation, management and more efficient use of initial material in maize breeding programs for new inbred lines and high yielding maize hybrids development.

Keywords: maize, genetic diversity, germplasm, phenotypic diversity, heterosis.

BIOLOGICAL EFFICACY OF THE NEEM OIL FOR THE CONTROL OF *APHIS GOSSYPHII*

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The role of insecticides in human society is very important. Phytophagous insects can cause losses of 10 to 90% of cultivated crops. In addition to affecting crop growth, harmful organisms can subsequently damage stored crops. However, using of synthetic pesticides has a direct negative effect on human and animal health. Also, the excessive or inappropriate use of synthetic insecticides causes biodiversity loss and occurrence of pests' resistance to these substances. The use of secondary metabolites synthesized by some plant species as part of their natural self-defense against pathogens and pests seems to be an excellent alternative to synthetic insecticides.

In our study which took place during the 2021 year we tested the biological efficacy of neem (*Azadirachta indica* A. Juss) seed oil on *Aphis gossypii* populations. The treatment was performed under laboratory and greenhouse conditions using cucumber (*Cucumis sativus* L) plants. Regarding the laboratory research the cucumber seedlings were planted in a greenhouse and infected with aphid populations transferred from a natural agroecosystem. Samples were collected from cucumber leaves in the greenhouse. The leaf blades inhabited by pests were placed in Petri dishes in 4 variants and 4 replicas for each variant. Counting of dead individuals was performed next day after treating the samples with extracts working solutions. The solutions were prepared in 0% (control), 0.1% and 0.5% neem oil concentrations while 1% ecological Pelecol insecticide was used as a standard. In the greenhouse experiments the aphids inhabited cucumber plants naturally. The plants were also treated with four variants (each variant in three repetitions): experimental variants - neem oil in two doses (8 Litre per Hectare (L/ha) and 10 L/ha), standard (Pelecol EO – 10 L/ha) and control.

The laboratory experiments showed strong aphicide effects (especially the highest concentration variant) the pests dying before they could leave the treated cucumber leaves. The greenhouse studies confirmed the potent insecticide action and they showed also a moderate repellent effect. The extract inhibited aphid feeding for a period of 5-7 days but was not able to completely inhibit the consumption of plants. The neem oil in a dose of 10 L/ha recorded highest biological efficiency - 90.05% (this was slightly higher than Pelecol result - 89.09%) and has the potential to be used as an aphicide.

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Keywords: *Azadirachta indica*, *Aphis gossypii*, harmful organisms, oil, repellent effect, treatment.

PHENOTYPICAL VARIABILITY OF FETAL TRAITS IN MUTANT TOMATO FORMS

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When creating new varieties and hybrids of tomato, the fundamental role is given to the characteristics of the fruit, since it is on them that the volume and quality of the final product, which determine the price of the efforts invested by the breeder, depend.

The main characteristics of the fetus include color, shape, weight and thickness of the pericarp. Knowing the degree of their variability depending on the genotypic features of the mutant forms and environmental conditions will make it possible to use them more effectively as a new germ plasm.

Of the many traits of the fetus, the main one is its mass. This is a complex trait, determined by a large number of genes that cause its significant variation. This opinion is confirmed by our results. The average coefficient of variation in fetal weight in the studied mutants (125 forms) was 33.6%, which characterizes the trait as highly variable. The highest amplitude of trait variability (from 20.7% to 66.4%) was found in 45.7% of the studied mutant forms. The average degree of intrapopulation variability (10.2 - 19.8%) was shown by 41.4% of genotypes from the collection of mutants. Only 12.9% of genotypes were characterized by high homeostasis (2.8–9.4%). The variability of the trait "fruit weight" was high both among plants of the population of one mutant form and between fruits within the same plant. In large-fruited mutant forms with round and flat-round fruits, the variability was higher (29.7%) than in forms with oval and plum-shaped (17.8%).

Despite the revealed high degree of variability of the trait "fetal weight" in the studied mutant forms, it should be noted that the genetic features of the trait, determined by the norm of reaction to environmental conditions specific for each genotype, are preserved. A high heterogeneity of the mutant collection in terms of fruit color was revealed, which is represented by a large number of mutant genes: *o at, ep, gs, gf, hp, t, u, ug, Ip, l, r, sh, y*, which determine the nature and intensity of fruit color. The content of *lycopene* and *β-carotene* in fruits is controlled by genes *B, B^c, B^{og}, Del*. The *pat* and *pat-2* genes give the fruits fleshiness and high elasticity. The degree of pubescence of different parts of the plant, including fruits, is determined by the *Ln, p, vi, and Wo^m* genes. The presence of keeping quality genes (*rin, nor* and *alc*) ensures the preservation of fruits up to 2-3 months. The severity of these signs varies greatly depending on the growing conditions of plants.

Knowledge of the nature of manifestation and the degree of variability of the declared traits presupposes their successful use in breeding to improve existing and create new varieties and hybrids of F₁ tomato.

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Keywords: fetal weight, fruit weight, mutant forms variability, tomato.

USING MUTANT *ls* AND *br* GENES OF TOMATO TO CREATE A NEW SOURCE MATERIAL

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It is known that when mutant forms are crossed with other biotypes, there is an exchange and enrichment of the population with genetic material. Some mutant genes in the process of splitting activate the reactions of genetic recombination's and, thereby, expand the range and level of intrapopulation variability available for selection in F₂ – F₃, making it possible to obtain new valuable source material and thereby increase the efficiency of the breeding process.

To create new forms of tomato with limited formation of lateral shoots and short internodes, two genotypes were involved in the breeding process, the semimutant line 11069 and the mutant sample Mo 443, which are carriers of the *br* (*brachytic*) and *ls* (*lateral suppressor*) mutant genes that control these traits. Using these forms, and cultural lines – L28, L111, L187, L828, L556, as well as highly productive, large-fruited varieties

– MilOranj, MaKrista, Fakel, with a set of valuable quantitative and qualitative traits that have genes – *sp*⁺, *sp*[±], *sp*, *ssp*, *u*, *nor*, *rin*, *ex*, *j*-2, 8 hybrid combinations were created.

The analysis of F₁ hybrids revealed 100% dominance of the shoot-forming ability of cultivated parental forms in all the studied combinations. The number and size of lateral shoots was different and was in direct proportion to the degree of their development in parental forms. The trait “length of internodes” was inherited according to an intermediate type (mid-parent).

In the splitting F₂ populations of all hybrid combinations involving the mutant forms L11069 and Mo 443, there are very few plants with the absence or single lateral shoots (7.3 - 12.7%), they have short internodes (< 6 cm), but at the same time a large part of the flowers (from 41 to 100%) on the inflorescences of these plants with deformations of varying severity. It has been established that the less lateral shoots are formed and the shorter the internodes on the plant, the more morphological deviations in the structure of flowers and the smaller the fruits. This indicates a multiple pleiotropic effect from the influence of the *ls* and *br* genes due to their insufficient cultivation, or these side effects are due to linkage with other genes that have an indirect effect, enhancing the nature of the manifestation of mutant traits. From different hybrid combinations, genotypes with a smaller number of lateral shoots, reduced internodes, and a normally developed reproductive system were isolated to study their offspring in the F₃–F₄ generations.

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Keywords: mutant forms, tomato, breeding process, F₁ hybrids, *br* (*brachytic*) gene, *ls* (*lateral suppressor*) gene.

EVALUATION OF ROS ACCUMULATION IN TOMATO ROOTS DURING POSTSTRESS ACLIMATIZATION

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Reactive oxygen species (ROS) fluctuations in time and space can be interpreted as signals to regulate growth, development, cell death and stress responses. The microscopic analysis obtained on 4 tomato genotypes (*S. pimpinellifolium*, Rufina, Mihaela and Mary Gratefully) show that the length of the ROS area accumulations in the root apical meristem decreases significantly in development dynamics - 96, 120 and 144 hours after seed immersion in water and 24, 48, 72 hours poststress (hps). The values of the ROS area length in optimal conditions, 96 hours, varied between 42.5µm (Mary Gratefully) and 30.4µm (*S. pimpinellifolium*) and for 144 hours decrease down to 22,4 and 2,4 µm, respectively. The heat stress determined a significant decrease of 2-15 times the length of ROS area compared to the control at 24 hps. The same trend for all analyzed terms within the genotype was maintaining except for *S. pimpinellifolium*, in which the length of ROS area increased at 48 and 72 hps, comparing to 24 hps variant. In the case of drought stress, accumulation of ROS was attested at 24 hps only, the other terms no positive histochemical response was recorded for ROS. Stress also causes a redistribution of ROS accumulations in all areas root, in a manner specific to the type of stress and genotype. Elongation of the root at 72 hours post-heat stress indicated higher values for the genotypes that marked and higher indices of the area with ROS accumulations in the following order: Mary Gratefully, Rufina, Mihaela and *S. pimpinellifolium*.

The analysis of the variance of ROS area length established a significant contribution of all analyzed factors and their interaction, the highest share of variation being due to *stress* (17%), *hours poststress* (13%), their interaction (11%), and of *genotype* (9%).

Evaluation of root cell viability by Evans Blue test established that germs exposed to heat or drought stress, compared to the optimal variant, microscopically marked penetration of the reagent in almost all meristem area at all evaluation terms -24, 48 and 72 hps. Thus, inhibition of main root growth under stress as result of loss cell integrity in the meristematic zone was accompanied by stimulation of adventitious root growth, which could be marked by ROS accumulations in the tip of adventitious roots, and their location was in the immediate vicinity of the meristematic area compared to their location in the differentiation zone in the optimal conditions. Morphological analyzes established differences in root architecture of plants exposed to heat or drought and optimal conditions according to the length of the main root, the number, length and location of secondary roots. Drought and heat stress applied at the germination stage drastically change the root parameters, which can have repercussions on further plant development.

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Keywords: genotype, tomato, reactive oxygen species, heat and drought stress, germination, root.

EXPANSION AND DIVERSITY OF THE BROOMRAPE RACES IN THE REPUBLIC OF MOLDOVA

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The recent increase in the world production of *Helianthus annuus* L. and the rapid evolution of its parasite - the broomrape (*Orobancha cumana* Wallr.) led to changes in the genetic structure of *O. cumana*. So far, eight races of broomrape (A, B, C, D, E, F, G and H) have been described, which have unfavorable consequences for sunflower cultivation. In the Republic of Moldova, the broomrape was first mentioned in 1863, only about 20 years after the introduction of sunflower in culture. Moreover, in the 1960s, a new biotype, was called the Moldovan race, or C race (Sharova, 1969). In the early 2000s, the results obtained on racial status allowed the reporting of race A-F. It should be noted that race A was predominant, being identified in about 30%. B race was highlighted in 22% of the locations, C race - 18%, D race - 15%, E race - 12%. Compared to the races \leq E, the presence of the race F was identified only in 3% of the localities (Petcovici et al., 2009). Thus, the proposed goal was to assess the racial status of *O. cumana* in the Republic of Moldova, in dynamics during the last 10 years (2010-2020). For this purpose, a series of expeditions were carried out in the country in which more than 130 sunflower fields were evaluated, and it was found that the infection is widespread. Subsequently, in 2014, the investigations covered almost the entire surface of the country (158 fields in 95 localities) (Duca et al., 2017). Broomrape was found in 63% of the southern localities, 47% of the central localities and only in 10% of the analyzed fields in the northern regions. At the same time, *O. cumana* seeds were collected from more than 60 populations, which were used to establish the strains of the parasite using differentiating host plants. As a result, the entire complex of known physiological races was detected. Thus, the less virulent races than E, were predominant, being identified in 37% of the investigated localities, the presence of the F race was noticed in 14% of the localities, especially from the south of the republic, and the G race in 27%. Research has shown that nine of the populations surveyed also infected resistant breeders resistant to the race G, which indicates the identification of the race H for the first time in the Republic of Moldova (22%). The appearance of this biotype of the parasite in our country was confirmed following a new assessment of the racial status of the broomrape, conducted in 2019, on agricultural fields in 15 localities in the Republic of Moldova. The results of this study reported the presence of races E-H, the new *O. cumana* race spread entirely throughout the country, the race H becoming the dominant physiological races of the parasite (56.2%) (Duca et al., 2022). The results obtained in the present study demonstrate an accelerated evolution of this sunflower parasite on the agricultural fields of the Republic of Moldova. Following the expeditions made in 2014, the presence of the race H of broomrape was reported, which was later shown to have spread over the entire surface of the country.

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Keywords: *O. cumana*, races, sunflower, broomrape

BIOMORPHOLOGICAL PECULIARITIES OF SOME SPECIES OF THE GENUS *CUPHEA* IN CONDITIONS OF INTRODUCTION

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Cuphea is a New World genus and the largest of the 32 genera of *Lythraceae* with about 260 species. Economic value of these plants is very diverse but of special interest is the ability of plants to synthesize and store oil (16 to 42%) in seeds containing capric, caprylic, lauric and myristic acids used in production of laundry powders, plasticizers, and also in perfumery, medicine and in production of biodiesel. The purpose of our investigations was to study the manifestation of biological parameters of the representatives of the genus *Cuphea* - *C. lanceolata* (1), *C. viscosissima* (2), *C. lutea* (3), in conditions of their introduction in the Central Area of Moldova. Thus, plants of all genotypes were characterized as herbaceous, annual plants with tap root system and upright stem. They have simple, opposite, and entire-kind leaves. Flowers are small, single, zygomorphic, located interaxillary. Corolla has six unequal petals, they are uniformly violet-cherry colored in plants of the 1st and 2nd species. Plants of the 3rd species have flowers with two violet petals and four white petals with red central vein. Androecium has eleven stamens located in two rings. Gynoecium is syncarpous. Stem, leaves and sepals have sticky hairy surface. Fruit represents a seed case where small oblate brown seeds develop. The investigated species are characterized by a fairly long flowering period (from the second decade of June until the first frosts). Representatives of the first 2 species begin to flower 8-10 days earlier than genotypes of the 3rd species. Fruit formation was noted to begin in the first decade of July and fructification lasted till October. To determine reproductive potential of plants, the analysis of pollen must be conducted because the changes in basic characteristics of pollen influence fertility and reproductive biology of plants. We performed morphological description of pollen grains using scanning electron microscopy method. For characteristics of species, the following parameters were used: diameter, shape, surface and aperture of pollen grain. Pollen grains of the first species have oblate shape, 20.65 to 23.04 μm in diameter. Aperture contains three pores, pore diameter varies within 4.5 to 6.0 μm . Pollen grains are sincolporate and have no apocolpium. The structure is wrinkled in mesocolpium. *Cuphea viscosissima* Jacq. Pollen grains are sincolporate, with three prominent pores and streaks located on the opposite poles. The shape of pollen grains is oblate, 21.0 to 22.86 μm in diameter. Pore diameter varies within 4.0 to 6.0 μm . Mesocolpium surface is smooth except for the area around the pore where it has wrinkled sculpture. *Cuphea lutea* Rose. The shape of pollen grains is oblate, 24.57 to 26.09 μm in diameter. Pore diameter varies within 7.62 to 8.0 μm . Mesocolpium surface is smooth except for the area around the pore where it has wrinkled sculpture. *Cuphea* species investigated show significant potential for productivity and resilience in introductory conditions.

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Keywords: genotype, reproductive potential, pollen grain, morphological traits.

**STUDY OF LAKE FUNGI BIODIVERSITY IN FROM THE
LA IZVOR LAKE (CHISINAU MUNICIPALITY)**

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Fungi (mushrooms) are indispensable components of biota in any ecosystem. In aquatic environments, fungi belong to important microbial communities for organic decomposition, nutrient cycle and energy flows and play important roles in the dynamics of the trophic network of freshwater ecosystems. Fungi are among the least studied groups of aquatic microorganisms, so the aim of the research was to study the biodiversity of fungi in Lake Izvor.

247 strains of fungi isolated from samples taken from La Izvor lake (water, silt, and biofilm) were studied. For the isolation of fungi, 5 agar media were used (Czapek: Sabourand, Malt - agar, Agar - nutritious, Raistrik), specific for the growth of filamentous fungi.

The highest fungal biodiversity was detected in the silt samples (87 strains), then in the water samples (85 strains), and the least in the biofilm samples (75 strains).

The cultural and morphological features of the isolated fungal strains were studied. As a result of the research, representatives of 18 genera of fungi were found, these being: *Penicillium*, *Aspergillus*, *Trichoderma*, *Alternaria*, *Fusarium*, *Botrytis*, *Monilinia*, *Mucor*, *Rhizopus*, *Acremonium*, *Cladosporium*, *Trichocladium*, *Phoma*, *Chaetomium*, *Stachybotrys*, *Talaromyces*. In all studied samples, the most common genera of filamentous fungi proved to be *Penicillium* and *Aspergillus*, followed by the genera *Trichoderma* and *Alternaria*. Together these genera represent about 90% of the total number of isolated strains. It was found that depending on the place of isolation, the representatives of the genus *Penicillium* predominate in the water, and in the silt, and biofilm samples – the representatives of the genus *Aspergillus*.

The obtained results showed that, from the genus *Penicillium*, the species predominates: *P. verrucosum* and *P. corylophilum*, from the genus *Aspergillus* predominates *A. niger* followed by *A. flavus* and *A. fumigatus*. *Trichoderma* strains belonging to the species were found: *viride*, *harzianum*, *atrobrunneum*, *simmonsii*, *longibrachiatum*, etc. *F. oxysporum* and *F. moniliforme* predominate among the species found in the genus *Fusarium*. Most of the isolated silt strains were *Aspergillus niger* and *Alternaria*.

The data obtained are in accordance with the data in the literature which states that freshwater predominates filamentous fungi of the genera *Penicillium*, *Aspergillus*, *Alternaria*, which contribute to the breakdown of organic matter, nutrient cycle, and energy flows.

Keywords: fungi, biodiversity, La Izvor lake.

NUTRITIONAL QUALITY AND DIGESTIBILITY OF MAIZE HYBRID PLANTS FOR SILAGE

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In order to assess the silage potential of maize hybrids, the nutritional quality and the enzymatic *in vitro* digestibility of five local maize hybrids (ZP 707, ZP 7357, ZP 7072, ZP 7777, and ZP 6263) was tested in the laboratory of the Group for Food Technology and Biochemistry of the Maize Research Institute "Zemun Polje". The hybrids were grown at a total of four locations, one in Srem (Autonomous Province of Vojvodina) and three in Central Serbia. The hybrid 7001 was used as a standard. The selection of hybrids for this research was made on the basis of the actuality of individual hybrids and the market orientation of the Maize Research Institute. The following properties were investigated: dry matter content, lignocellulose fiber content, and *in vitro* dry matter digestibility of the whole plant. According to the achieved results, it can be concluded that hybrids ZP 707, ZP 7357, followed by ZP 7777 proved to be the maize genotypes highly preferable for the production of silage. All tested hybrids achieved better results than the standard in most locations. Hybrid ZP 707 on average had the highest *in vitro* dry matter digestibility ($61.43 \pm 1.86\%$), as well as the lowest content of all lignocellulosic fibers (NDF-52.76%, ADF-24.40%, ADL-2.58, hemicellulose-28.36, and cellulose-21.82%), which all indicates its potential as a silage maize form suitable for cultivation in different agro-ecological conditions. In terms of digestibility and dry matter content, the ZP 707 hybrid can be singled out as the most stable, i.e. it is appropriate for growing both in lowland areas and at higher altitudes. ZP 6263 proved to be the most inferior hybrid at most locations, while based on *in vitro* digestibility and dry matter content, ZP 7072 hybrid varied the most. The digestibility of the whole plant was negatively affected by the higher content of primarily lignin (ADL), followed by ADF and cellulose fraction share. Although it is optimal to harvest silage maize in the waxy maturity stage of grain ripeness, when the dry matter content of the whole plant is in the range between 30 and 35%, the harvest time in some hybrids in some locations was significantly exceeded, which affected the results of dry matter digestibility. The findings obtained in this study can be highly useful for future breeding programs directed toward creating new and improved silage maize hybrids.

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Keywords: maize, hybrids, silage, animal feed, breeding

IMPORTANCE OF WALNUT (*Juglans regia* L.) WITHIN REPUBLIC OF MOLDOVA

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Among fruit growing species in the Republic of Moldova, the walnut is the most representative and the most appreciated from economic, social, ecological and aesthetical points of view. Always walnut has been a symbol of prodigality, virtue, resistance and wellbeing for Moldovan people. Walnut image is presented in the folk songs, romances, ballads, sayings, riddles, ceremonial customs mentioned in the writings of the local folklorists. It is a secular tradition to plant one walnut tree when a son is born into the family. Peasants always like to plant a walnut tree at the corner of their house - a pleasant place for recreation. Usually respected household have a tall and vigorous tree, which would be able to listen and understand you so well. Thus, it is the spirit of the people of Moldovan lands to grow walnuts.

The walnut has multiple uses due primarily to the food value of its fruits, leaves, and due to its quality wood, highly appreciated in the furniture industry, as well as the use of trees in some cases to fix kneaded and degraded land. Aesthetic and ecological role of walnuts is well known in the frame of different kind of social places (for example: parks, schools, sanatoriums, monasteries). Walnuts are the preferable tree species on secondary roadsides, alleys and usually serve as a basic species in the different kind of alleys and forest belts. Like in other regions dried nuts are gifted to children during religious celebrations. The dried kernel is used in preparing pies, cakes, candies and ice cream as well as simple foods like honey with kernel, etc. A special roly-poly called "*invirtita*" is present at Christmas, Easter and other religious celebrations and commemorations. Nuts kernel is an important ingredient of diverse culinary dishes such as salads, pastes, etc. Green nuts are used to make balsam, "sugariness". Alcoholic extracts from kernels mixed with different medicinal herbs, to vitaminize wines and spirits, which are used in medical scope too. The juvenile fruits are used for the preparation of different special extracts as an antidysentery or intestinal astringent function. Over centuries in our country shelled fresh kernel is eaten during the month of September (when lipids content is minimal), after special religious days, 29 August - Walnut Easter. Excellent edible and technical oil is extracted from walnut kernels, which is usually used also for technical purposes: in painting, to obtain technical ink and in the preparation of numerous cosmetic and pharmaceutical products. Among others fruit growing species in the Republic of Moldova, Green nuts are used to make balsam, "sugariness". Alcoholic extracts from kernels mixed with different medicinal herbs, to vitaminize wines and spirits, which are used in medical scope too. The juvenile fruits are used for the preparation of different special extracts as an antidysentery or intestinal astringent function.

Over centuries in our country shelled fresh kernel is eaten during the month of September (when lipids content is minimal), after special religious days, 29 August - Walnut Easter. Excellent edible and technical oil is extracted from walnut kernels, which is usually used also for technical purposes: in painting, to obtain technical ink and in the preparation of numerous cosmetic and pharmaceutical products. Walnut leaves are also used differently.

First of all, branches carrying fresh leaves are hung on doors during some summer religious days as a symbol of resistance, wellbeing, etc. A special tea for health fortification is prepared from its leaves. Dried leaves are very good for hair washing and against skin diseases. As a fruit growing species, the walnut provides raw material for many purposes. At all times walnut wood in Moldova is mainly used for the production of furniture (doors, windows), parquet, different confectionary like pipes, souvenir boxes and bails etc. Walnut leaves are also used differently. First of all, branches carrying fresh leaves are hung on doors during some summer religious days as a symbol of resistance, wellbeing, for tea, etc. Walnut (*Juglans regia* L.) is a species, which is permanently studied in the frame of horticultural researchers of Rep. Moldova.

The results of collaborative programs and implementation in different countries showed high ecological plasticity and adaptability of the main selected and created Moldovan varieties, considering changeable microclimatic and pedological local conditions. The evolution of walnut registration assortment also took into consideration consumers' preferences. Highlighted most of all, were the varieties with white walnut kernel, that were easy to shell and with a comparatively good taste and thin shell. Thus, the most popular varieties of walnuts in Moldova are: 'Kazacu', 'Kogalniceanu', 'Kostiujenskii', 'Kalarasskii', 'Schinoskii', 'Pescianski', 'Kojjeuti' Actually in the commercial walnut plantations, the following varieties were used for testing: 'Chandler', 'Franquette', 'Lara', 'Femor'. It is important to notice that in the last time Chandler is more adaptable for local condition and marketing requirements. In Republic of Moldova, with the serious problems of water for irrigations, as walnut rootstocks serve the biotypes and varieties of local origin of *Juglans regia* ('Kisinevskii', 'Kostiujenskii', 'Kazacu', etc.). This species ensures a good grafting affinity with all approved local and introduced varieties. Biological (including differentiation of floral and vegetative buds, flowering and pollination) and agronomical proprieties were detailed investigated for registered and introduced varieties, including comparative behavior within the vegetal rest periods of different local competitive micro cultural areas. Main of them they are characterized by their high adaptability to diverse local environmental (edaphical and microclimatical) conditions (Comanici, 1980; Tzurcanu, 2004; Pinte, 2004, 2021). The principle trials of nut culture there are: stable productivity and nut (fruit) quality. Studies of local walnut genetic resources (Pinte et al., 2021) demonstrate that Rep. of Moldova could be considered as a "pan-population" in which the plants are able to exchange genes by wind pollination. The trees with lateral fruit bearing, founded also in natural population, makes our country important for walnut breeding and implementing this trait is very important to enhance the productivity aimed by breeders. collection, characterization, propagation and sustainable use of walnut genetic resources, assessment of the adaptive potential and phenotypic plasticity, are therefore items of considerable importance both for the preservation in situ and ex situ biodiversity and basis for the improvement of new varieties and the prevention of the extinction of genetic sources.

Acknowledgements: The research was done in the frame of research project: 20.80009.5107.14 (Rep. Moldova, Research Institute for Horticulture and Alimentary Technologies).

Keywords: *Juglans regia* L., varieties, culinary, pharmaceutical properties, furniture and wood industry, adaptability, productivity.

LOCAL PLUM VARIETIES OF REPUBLIC OF MOLDOVA FOR BREEDING AND PRACTICAL PURPOSES

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Culture tradition of plum (*Prunus domestica* L.) between rivers Nistru and Prut (actual Republic of Moldova) dates back to almost 2000 years ago. Processed plumes (prunes) having important socio-economic and heritage value being intensively exported. Starting from the 18-19 centuries plums there are cultivated practically in all home garden (domestic orchards). According investigations of exploration main of them are named: Vinete de Tiraspol, Rotunda-1, Bardace, Bardace 3, Bardace 5, Goldani ciornaia, Prune moldoveneti, Vinete de Valcinets, Prune moldovenesti. During the 1800 years in the territory of actual RM there were intensively established commercial plums orchards on the basis of local, as well as introduced European varieties.

Factually in the RM more than 25000 private farmers there are involved in the production of fruits, especially plums. As consequence of dynamic expansion and reconversion of fruit trees plantation occurred. Of course, at the same time continue to be abandoned and lost old plum genotypes. Now practically plum (including 1-2 old varieties) grow in all local yards and orchards, dried plumes (prunes) continued to having significant socio-economic and heritage value. There are promoted significant change of the agriculture in general, that moved from the land-owner old style management to the modern intensive farming. Old catalogues of 7-10 main local nurseries listed a lot of varieties. As consequence a dynamic expansion and reconversion of fruit trees plantation occurred.

Therefore, evaluation and establishment of germplasm of old local plums genotypes/varieties are indispensable. All representative accessions were selected in north and central different pomological zones of the Republic of Moldova. Major characterization characteristics of the plants and fruits of old varieties were conducted using UPOV descriptor, field evaluations were based also on IBPGR plum descriptors. Chemical evaluation of created, selected, introduced varieties and hybrids, being performed on the laboratory of biochemistry as well as in the department for Food technology. The following parameters were examined: total sugars and organic acid content, vitamin C content; soluble dry material; non soluble solids substances, total phenol content. In this study was determine main fruits characteristics such as fruit weight, firmness, titratable acid content and soluble solid substances and important phenological periods such as harvest of these genotypes. Visual survey of the phytosanitary status (first of all tolerance to PPV on fruits and leaves) was also carried out.

Actually, in the Republic of Moldova plum continue to be one of the principal fruits of peasant domestic orchard, especially for vegetative propagated old varieties. In the same time there are promoted significant change of the agriculture in general, that moved from the land-owner old style management to the modern intensive farming. As consequence a dynamic expansion and reconversion of fruit trees plantation occurred. Of course, at the same time continue to be abandoned and lost old plum genotypes.

Therefore, evaluation and establishment of germplasm of old local plums genotypes/varieties are indispensable. It is necessary to conduct *in situ* and *on-farm* inventories of plum genfond. It is important also to notice that after characterization of main biological and fruits indicators we establish that differences between accessions are notable, especially concerning organoleptical and eating qualities.

Main representative local plum variety there is Vinete de Moldova with short description. **Synonym:** Vengherka moldavskaia. A lot of centuries was growth (multiplied) by shoots. Spreading: It is widespread all over in the rep. of Moldova and in the nearby regions of Romania and Ukraine. From 1998 year this cultivar is registered for large propagation in the Republic of Moldova. A d a p t e d to different soils and the moderate continental climate conditions. **Harvesting time:** fruits mature in late August and early September and in the north part of Moldova - in the middle of September. **Used value of the fruit:** of jam called 'povidla', jam, compote, sweet, brandy, but mainly for drying. **The tree:** Medium-vigorous. The crown is dense and wide-pyramidal. The scaffold branches are well-covered with fertile twigs. **Leaves:** Elliptic, medium to large size, dark green color. Younger trees have larger leaves. Leaf stem is medium thick, short to medium long, green or greenish-red color. **Flower:** Medium-large, white with elongated petals. Pistils and stamens are in the same level. Some **Physiological traits:** **Vigor:** Medium. **Blossoming:** on medium period and explosive. For one day 50 to 80% of the flowers can be opened. Self-fertile variety. **Productivity:** there are many biotypes that differ in size of the fruit and yield.

Fruit characteristics. Size and shape: standard Vinete de Moldova fruits are small to medium, weighing approximately 19-23 g, and there are types of fruit with a mass up to 25 g. The fruit is of prolonged ovoid form with oval shape. **Fruit powder:** well defined. **Fruit skin:** dark blue color with pronounced fruit powder, moderate thin, easy to separate of flesh. **Fruit flesh:** golden to green, moderate juicy, firm, sweet and sour, excellent quality. **Stone:** small, elongated shape. Stones are easily separated from fruit flesh (freestone).

Significantly to notice that created in Rep Moldova plum assortment for long time there was principal for orchard establishment. In the last time the most important for fresh consumption and 1-3 month preservation there are important local with late maturation varieties Udlinennaia and Super president, obtained from controlled hybridization. In the conditions of climate changes some old local as well as new created Moldovan plum varieties (especially **Udlinenaia** and **Super president**) represent a permanent source for important genetic traits, for instance: different desirable nutritional qualities, new type of processing, good capacities for transportation and relative long time of preservation within controlled conditions. Good resistance to extreme agro-ecological conditions, resistance to economically significant harmful biologic agents also there are present.

Acknowledgements: The research was done in the frame of research project: 184/23.10.19 A (Rep. Moldova, Research Institute for Horticulture and Alimentary Technologies partner of Institute of Genetic, Physiology and Plant Protection).

Keywords: *Prunus domestica* L., varieties, germplasm, biological indicators, organoleptical qualities, productivity.

EFFECTIVENESS OF CARBECOL IN PREVENTING AND COMBATING POWDERY MILDEW ON VINE

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In the Republic of Moldova the perennial plantations occupy an area of about 288.9 thousand ha, including orchards - 138.9 thousand ha and vines - 150.0 thousand ha. The intensive chemicalization of the republic's agriculture in the last 30-50 years has led to an increasing volume of used agrochemicals. This did not contribute to crop growth, but only to environmental pollution and to occurrence of new species/breeds of resistant pests. The implementation of different schemes for combining biorational pesticides in integrated protection allows to reduce the number of treatments with 2-3 times while lowering energy costs with 30% - 40%. This refers especially to the protection of multiannual crops and vegetables where the frequency and volume of pesticide application is quite high. The powdery mildew (oidium) of the vine (*Uncinula necator*) ranks second place after the damage caused to the vine crop. The pathogen attacks all the aerial organs of the plant: leaves, grass shoots, bunches and berries. Crop losses due to this disease can range from 10% to 80%.

The aim of the research was to evaluate the biological efficacy of Carbecol based on potassium bicarbonate (2KHCO₃) in preventing and combating powdery mildew (*Uncinula necator*) in vines. Given the low level of toxicity and risk to the environment, this group of preparations is of particular interest as biorational fungicides. The researches were carried out during the years 2020–2021 within the institutional project no. 20.80009.5107.19: “Strengthening the capacities for forecasting and combating harmful organisms and phytosanitary risk analysis in integrated plant protection”, in the central area of the Republic of Moldova, experimental group of the Institute of Genetics, Physiology and Plant Protection. The experiments were performed on Cardinal vine variety and consisted in 5 treatments applied with a portable sprinkler while the climatic conditions were favorable for the appearance and development of studied pathogen.

The experimental scheme included four variants: control, standard and two variants of Carbecol in different doses (4.0; 6.0 kg/ha). As a standard was the fungicide Kumulus DF (6.0 kg/ha), used in production. Each variant consisted of three repetitions, and each repetition with 10 hubs with the same crown parameters.

As a result of the statistical processing the highest biological efficacy was recorded at the chemical standard Kumulus, - 85.7%. In the Carbecol variant - 4.0 kg/ha, the biological efficacy constituted 72.8%, while in the Carbecol variant - 6.0 kg/ha, - 78.7%. Analyzing the obtained results, we can conclude that the biorational product Carbecol has demonstrated essential efficacy and can be used in the prevention and control of powdery mildew on vines, within the ecological system of plant protection.

Keywords: *Uncinula necator*, pathogen, carbecol, fungicide, biological efficacy.

DIFFERENTIAL GENES EXPRESSION UNDER ANTERO- AND RETROGRADE CONTROL IN SUNFLOWER MICROSPOROGENESIS

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Nowadays, sunflower is among the leading crops of major importance for global food security. The economic interest of sunflower breeding is mainly determined by the production of seeds, whose biological yield can be increased by exhaustive knowledge of reproductive development. The control of pollen fertility is essential for germplasm diversification, hybrids with a high degree of heterosis obtaining and, more recently, ensuring the biosecurity of the introduction of transgenic plants into culture, as well as for the numerous researches of male gametophyte in various physiological or changeable environmental conditions. Of particular interest are the peculiarities of male gametophyte development in plants with gibberellin-induced male sterility (GA₃-IMS), a phenomenon that is not based on dysfunctions generated by the different genomic interactions. Thus, fertile inbred lines with androsterile analogs, fertility restorer lines and progeny (F₁) with restored fertility form an excellent model for research of cell communication, and being associated with AG3-IMS may elucidate the antero- and retrograde signaling processes of male gametogenesis under the gibberellins action. Previously, was provided the first evidence that sunflower with GA₃ induced male sterility is associated with a similar transcript to mitochondrial orfH522 characteristic for PET1 cytotyp. Taking into account that the GA₃-IMS phenotype is not the result of nucleocytoplasmic incompatibility it was concluded that meiosis cell division, mtDNA replication and mitogenome maintenance are not reliable. Thus, candidate genes involved in the mentioned events during sunflower microsporogenesis were assessed by RT- qPCR. In these investigations were used near-isonuclear lines, fertile SW501 with the *H. annuus L.* cytoplasm and male sterile SW501CMS with *H. petiolaris* cytoplasm, Drofa F₁ hybrid and its parents. The plants were exogenously treated with GA₃ solution by spraying on developing inflorescence buds. The florets from inflorescence buds (R1-R3) were subjected to microscopical, physiological and molecular analyses. The model of additive/nonadditive genes expression was used to compare transcripts profiles between F₁ hybrid and its parents. Sterility of sunflower anthers as GA-induced response resulted from the perturbation of the endogen hormone concentration, as well as from the ability of cells to perceive this perturbation and activate the subsequent transduction pathways of cellular signals. It was ascertained various genes expression profiles related to sterility type, genetic background and stage of pollen development. All studied sunflower transcripts are gibberellin responsive and showed high degree level of co-expression. Several genes differentially expressed in sterile anthers (CMS and GA-IMS) were down-regulated, suggesting on DNA damage-responsive cell - cycle arrest, an important mechanism for the preservation of genomic stability. Also, the transcriptomic data showed more non-additive gene expression patterns in hybrids than additive revealing, often, microsporogenesis stage specificity. It is suggested that those genes whose expression changes in F₁ hybrids (non-additively) can be associated with heterosis, being involved in specific signaling pathways that could be targeted regulated. The fact that researches on the mechanism of heterosis continue to be essential as part of enhancing food production, there is considerable incentive to investigate the genomic consequences of heterotic gene pool development.

Acknowledgments: The study was supported by the project 20.80009.5107.01 (NARD).

Keywords: sunflower, pollen fertility, microsporogenesis, genes expression.

REVIEW OF PESTS OF THE FOREST PARK "RISHCANI", CHISINAU

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In the architecture of the city, the forest park is one of the main elements of green and suburban areas. Monitoring and control of the phytosanitary state of urban green spaces contributes both to the preservation of the appearance of the park, the species diversity of the flora and fauna of the park, and the safety of its visitors. Within the framework of the project 22.80015.7007.261T „Smart solutions and biotechnologies for the sustainable green spaces development under urban environment” we analyzed the species composition of plants in the Riscani park in Chisinau. Growing perennial plant species were identified on the designated experimental sites. The most widespread crops were selected for monitoring the phytosanitary condition, which are found not only in this park area, but also in other forest plantations in Moldova. Therefore, this work is of national importance. The main species in forest plantations, squares and parks of Moldovan cities are varieties of maple, oak, linden, elm, chestnut, etc. The studies were performed according to the traditional methods approved in the Republic of Moldova. This paper presents the results of observations for the period March-July 2022, observations are still ongoing. We have found that on the territory of the Riscani Park almost all deciduous species are affected by different types of aphids (*Aphidoidea* sp.). These are insects with piercing-sucking mouthparts and, in addition to direct negative effects, they are carriers of plant diseases. We also identified the pest of oak, the lace bug *Corythucha arcuata* (Say). In addition to oak, the bug infects chestnuts, elms, currants, hawthorn, plane trees, lilacs. The bug actively feeds on the leaf blade, polluting it with excrements. The rapid increase of the oak lace bug population leads to the complete destruction of the photosynthetic leaves tissue.

The most aggressive quercine pests are found mostly on the leaves: the insects from the fam. *Cynipidae*, the green oak moth (*Tortrix viridana*), the mottled umber (*Erannis defoliaria*), the winter moth (*Operophtera brumata*), the trumpet leaf miner (*Tischeria complanella*), and on fruits- the oak weevil (*Balaninus glandium* Marsh.).

The red pine sawfly (*Neodiprion sertifer*) was identified on conifers, which is the main pest that harms the common, Crimean and mountain pine, eating their needles. The beetle actively flies in the spring period. Its caterpillar feeds on needles. All damaged trees weaken, lose their decorative effect, reduce their growth, and begin to be populated by bark beetles. As a result, in the republic every year we observe a deterioration in the phytosanitary condition of green areas of forest park plantations in the urban environment. Poor phytosanitary condition of the crown can lead to the complete death of the tree and the reduction of green spaces. We consider it necessary to carry out phytosanitary monitoring of park plantings in order to develop a strategy and schemes for plant protection in the park areas of the republic.

Keywords: plant species, diversity, fauna, pests.

BEHAVIOR OF SOME SUNFLOWER HYBRIDS AT THE ATTACK OF THE PATHOGEN *BOTRYTIS CINEREA* Pers., IN THE SOUTH-EAST AREA OF ROMANIA

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Sunflower cultivation is an important component of the food industry. It is grown all over the world and therefore has a wide range of pathogens that can affect it. As phytosanitary treatments are not always effective and economical, the cultivation of disease-resistant hybrids is the main measure for both optimal yields and environmental protection.

Botrytis cinerea Pers. is a pathogen that causes quality depreciation of production to a wide range of plant species used in human nutrition. The pathogen attacks the sunflower seeds that lose their taste qualities, the latter being decisive in terms of using the product in the food industry.

The paper aimed to observe the behavior of some sunflower hybrids to the attack of the pathogen *Botrytis cinerea*, in the climatic conditions of the South-East area of Romania in 2021.

To make the observations, the biological material used was represented by nine hybrids created at the National Institute for Agricultural Research and Development-Fundulea, cultivated in climatic stress conditions. The experiment took place within the Agricultural Development Research Station-Braila, Chiscani Experimental Field, in 2021.

The hybrid H5 stood out through its low degree of attack of 1.41% compared to the average degree of attack of 2.27%. The weakest behavior regarding the pathogen attack was observed in the hybrid H4, with a degree of attack of 2.6%.

Keywords: sunflower, *Botrytis cinerea* Pers., production, degree of attack, climate stress.

ENERGETIC CRISIS AND GRAIN SORGHUM VARIETY MODEL

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The energetic crisis occupies practically all countries in the civilization world today. Energy prices have skyrocketed. It has great influence on all activities including agriculture.

However, energy prices have no effect on global processes like the climatic changes and creation of population which can do the food crisis. The solution for the problem of alimentary deficit maybe the introduction of drought resistant cultures as sorghum.

Is known, the successful promotion of culture in long time perspective can be regulated by just autochthonous varieties and hybrids adopted for local climatic and soil conditions. Also, the effective breeding is based first of all on clear vision of variety or hybrid model. Until recently, the main requirements for autochthonous grain sorghum variety and hybrid models were precocity and big seed. But in wet years in Republic of Moldova even the ultra-precocity grain sorghum hybrids have harvest high humidity. It increases dramatically energy consumption in the post-harvest period. The single solution of this problem is the using of desiccants. They provoke seed humidity reduction in short time for harvesting. But using of desiccants maybe realized on undersized sorghum plants only.

Thus, the main requirements for autochthonous grain sorghum variety and hybrid models are precocity, big seed and undersized.

Keywords: grain sorghum, hybrid model, desiccant, drought resistance.

PRELIMINARY DATA ON THE DEVELOPMENT OF THE INVASIVE SPECIES *HALYOMORPHA HALYS* (STAL) 1855, (HEMIPTERA; PENTATOMIDAE) IN THE REPUBLIC OF MOLDOVA

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The brown marmorated stink bug, *Halyomorpha halys* (Stal), 1855 (Hemiptera: Pentatomidae) (BMSB) – an invasive species with a natural range of China, Japan, the countries of the Korean peninsula. The bug is a dangerous quarantine pest included in the Common List of quarantine facilities of the Eurasian Economic Union. In 2019, an invasive species was first discovered on the territory of the Republic of Moldova. In recent years, BMSB has been rapidly accelerating population development, supported by favourable climatic conditions for this species, as well as abundant plant food. Studies of biological features of invasive pests always have a strict local binding, so their conduct is necessary and on the territory of the Republic of Moldova.

The research was carried out both in the laboratory and in the field, on IGPPP test fields of the Republic of Moldova.

To determine the pest's survival after wintering on November 1, 2021, imago BMSB in the amount of 50 individuals were distributed in two insulated gardens and placed on an open terrace. After 140 days (the third decade of March), the count of surviving bugs after wintering was carried out with a further calculation in percentage.

When analyzing the data, we noted that the survival rate of the population after wintering exceeds 50%. The survival rate of females is 12.9% higher than that of males, which we believe will lead to a faster rate of population density.

In native conditions, we marked the period of release of the bug from the diapause from the third decade of April - the second decade of May. In the aftermath, there is a period of intensive feeding lasting from 1 to 2 weeks, and only then comes the period of mating. Visual observations have shown that BMSB females produce eggs 3-4 times within 6-13 days. The number of eggs in one egg range from 10 to 32. The fertility of one female averages 96 eggs (68-124).

Thus, based on the preliminary data we proved that in the agroclimatic conditions of the Republic of Moldova, this species can develop favorably and can cause economically important crop losses of various crops.

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Keywords: *Halyomorpha halys*, invasive species, wintering, survival, fertility.

**SCREENING FOR HEAT-RESISTANCE OF POLLEN IN PROGENY OF
VIRUS-INFECTED TOMATO GENOTYPES**

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The reaction of the male gametophyte of virus-infected tomato plants is differentiated under conditions of high temperature. At the same time, information on the heat resistance of the progeny of these genotypes is limited. The aim of the research was to study the effect of high temperature on the variability of the male gametophyte of the progeny of plants infected with TMV (*Tobacco Mosaic Virus*) or TAV (*Tomato Aspermy Virus*).

An assessment of the thermal resistance of the male gametophyte of healthy plants and progeny of virus-infected plants revealed, on average, the highest level of this trait in control plants – 62.5%. The level of the thermal resistance in TMV/TAV progeny was lower, by 13.3% and 22.2 %, respectively. Differences in the length of pollen tubes were also established between the control and experimental variants. In TMV/TAV progeny, as in the case of pollen viability, a decrease in the length of pollen tubes was observed by 32.9% and 28.4%, respectively, compared to the control. This result can probably be determined by the lower growth rate of the pollen tubes in these genotypes.

It has been established that the male gametophyte of control plants and progeny of virus-infected plants of varieties Mihaela, Rufina and the wild species *S. pimpinellifolium* combines a high level of heat resistance of pollen viability and pollen tubes length, which suggests the prospect of their use in further research.

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Keywords: tomato, TMV, heat resistance, pollen.

PHAGE EFFICIENT AGAINST FIRE BLIGHT AND FRUIT TREES BACTERIAL CANKER PATHOGENS

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Fire blight and bacterial canker of apple, pear and quince, the economically most important diseases of fruit trees, are caused by the bacterial phyto-pathogens *Erwinia amylovora* and *Pseudomonas syringae* pv. *syringae*, respectively. *E. amylovora* and *Ps. syringae* pv. *syringae* are often present together in the aerial parts of plants and in the most cases both bacteria are detected in the same plant samples. Currently available bacteriosis control agents as, e.g., copper preparations or antibiotics act mostly unspecifically and are prone to resistance development. Alternative disease management strategies are, therefore, highly solicited.

Bacteriophages, i.e. highly specific bacterial viruses that infect and lyse bacteria, are under intensive evaluation as the biocontrol agents. The advantage of phages is that they are natural components of ecosystems, infect only bacteria sensitive to them and are non-toxic to plants, animals and humans.

Phages, infecting *E. amylovora* and *Ps. syringae* pv. *syringae* known up to now belong to the taxonomic families *Podoviridae*, *Myoviridae*, and *Siphoviridae*.

When studied the ability of phages isolated in the quince orchard to infect cells of fire blight and bacterial canker causative agents one phage was detected to be able to lyse both pathogens. A major capsid protein based multiplex PCR assay used for phage identification revealed that the isolated phage belongs to the Y2 group, which includes *Myoviridae* phages. Some of the representatives of the Y2 group are the broad host range phages. By pulsed-field gel electrophoresis analysis genome size of the phage was determined.

After a detailed study of biological effectiveness, the isolated bacteriophage could be used for biocontrol of the fire blight and bacterial canker pathogens.

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Keywords: phytopathogen, multiplex PCR assay, bacterial canker.

THE IMPACT OF THE PRUNING SYSTEM ON THE SWEET CHERRY TREE GROWTH AND FRUCTIFICATION

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The formation of crowns of sweet cherry trees, the pruning time and the existing growing technologies that allow the maximum use of the biological potential of modern gardens remain relevant. Along with the classical methods (rootstock, variety, fertilization, irrigation, etc.) of regulating the growth and fruiting of sweet cherry trees, the pruning during the growing season is a process of improving the physiological balance between the vegetative growth and the number of fruits. The pruning of trees in early autumn, namely in the first decade of September, in contrast with the conventional pruning carried out when the trees enter the dormant period, has a greater impact on yield and fruit size. As a general trend, the pruning during the growing season leads to an increase in the size and colour of the fruit and to a decrease in the incidence of brown rot. It is increasingly used in high density orchards.

The purpose of the research was to evaluate the impact of the time of the pruning of the sweet cherry trees of Kordia, Regina, Stella, Ferrovia and Skeena varieties, grafted on the Maxima 14 rootstock, on the yield and quality of sweet cherries (*Prunus avium* L.). The orchard was laid out with 3 m between trees in the row, and 5 m between rows. The canopy of the trees was small and of a naturally improved shape. The maintenance and thinning pruning of the sweet cherry trees were done during both the growing season and the dormant period. The pruning system consisted of four time periods: G1 – Pruning during the dormancy (control group), G2 – Pruning during the flowering period; G3 – Post-harvest pruning; G4 – Pruning in early autumn. Four groups of eight trees each were used in the experiment.

The orchard was irrigated using the drip irrigation system. Water was distributed by droppers fixed at a height of 40 cm from the ground in the direction of the row. The soil was kept artificially grassed. The weeds on the two-meter-wide strips of land between the rows were mowed down as needed and left as mulch. The grass of natural or artificial growth, which grew on the strips 2-2.5 m wide between the rows of trees, was mowed as needed and left as mulch. The weeds were killed with herbicides applied along the rows, or mechanically weeded twice or thrice times using sensitive weeding equipment.

The pruning of sweet cherry trees is a process, which has a great impact on the fruit quality and yield. The pruning time had a strong impact on the growth and fructification of the cherry tree varieties grown in an intensive farming system. The pruning done during the growing season (the post-harvest pruning and the pruning in early autumn) had a greater impact on the tree vegetative growth, i.e. the height of the tree, the average and total length of annual branches, and the section of the trunk. The sweet cherry trees of the Kordia, Regina, Stella, Ferrovia and Skeena varieties, grafted on the rootstock Maxima 14, in G 4 (the pruning was done in early autumn) formed fruit branches with a large number of flower buds, over against G1 in which the pruning was done during the dormant period (the control group).

Keywords: *Prunus avium* L., fruit quality, pruning time, growth

THE IMPACT OF THE CROWN SHAPE ON THE GROWTH AND FRUCTIFICATION OF CHERRY TREE VARIETIES GROWN IN AN INTENSIVE SYSTEM

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In the Republic of Moldova, new varieties of cherry trees grafted on semi dwarfing rootstocks (Gisela 5, Krymsk 6), moderate vigor rootstocks (MaxMa 14, Piku 1, Piku 4) and semi vigorous rootstocks (Gisela 6, PHL-C, Krymsk 6) are cultivated. Dwarfing rootstocks have made it possible to grow high density orchards of small trees, which are more productive and develop earlier. Fertile well-irrigated soils are used to grow trees grafted on the above-mentioned rootstocks. Medium-sized trees, which can be fully tended from the ground, provide early average yield of fruit and reduce fruit harvesting costs by increasing the harvesting productivity. Since the use of dwarfing (Gisela 5, Krymsk 6), moderate vigour (MaxMa Delbard 14, Piku 1, Piku 4) and semi vigorous (Gisela 6, P HL-C, Krymsk 6) rootstocks, the need has arisen to use small crown trees as well. As the result of this fact, the impact of the crown shape on the productivity of the Early Star, Samba and Black Star varieties, grafted on Gisela 6 was studied.

The aim of the research was to increase the productivity of sweet cherry orchards by identifying highly productive crown shapes for the Early Star, Samba and Black Star varieties, grafted on Gisela 6, suitable for the Republic of Moldova. The researches were carried out in the central orcharding area of the Republic of Moldova, at the StarAgroGroop Ltd., between the years 2018-2021. The impact of the crown shape on the growth and fructification of the Early Star, Samba and Black Star cherry tree varieties, grafted on the rootstock Gisela 6, was studied. The trees were established in 2015. The orchard had a planting density of 1250 trees/ha. The crowns of the trees were of improved thin spindle shape, and of Cup and Kym Green Bush shape. The orchard was irrigated using the drip irrigation system and fertilized using foliar applications of urea and microelements to fruit trees. The soil on the rows of trees was artificial grassed and treated with herbicides. The weeds on the two-meter-wide strips of land between the rows were mowed down as needed and left as mulch.

The fruit yield varied greatly depending on the late spring frosts during the opening of the buds and the flowering of the cherry trees, the variety and the shape of the crown. In 2019, the Samba variety cherry trees produced the best crop, namely 16.8 t/ha, by 56.4-139.9 % larger than the Early Star and Samba varieties. In 2020, the Black Star variety produced a rich harvest of 10.3 t/ha, which was 111.4-151.4% larger than the Early Star and Samba varieties. In 2021, the Samba variety stood out with the highest harvest (10.3-16.2 t/ha) followed by the Early Star variety (7.7-9.4 t/ha) and Black Star (3.7-5.5 t/ha). The highest yield on average over three years was obtained from the trees with improved thin spindle crowns as compared to the cupped and Kim Green Bush crowned trees. In the first four years of their fruiting, the fruit harvest in the Samba variety was significantly higher as opposed to the Early Star and Black Star varieties.

Keywords: variety, productivity, sweet cherry, orchard, growth.

EVALUATION OF *SOPHORA FLAVESCENS* EXTRACT FOR COMBAT THE RED MITE (*TETRANYCHUS URTICAE*) TO THE TOMATO CROP IN GREENHOUSE

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Intensive use of increased amounts of chemicals in the agricultural sector for the treatment of tomato crops in greenhouse, led to a demand for organic products on the market. The need to reduce crop losses raises the issue of developing effective plant protection measures that reduce the dynamics of the development of harmful organisms with a minimum number of chemical treatments. Tomato crops in greenhouse suffer huge losses due to damage caused by red mite. This pest can be present throughout the vegetation period if various preventive measures are not taken in time. The aim of this article is to demonstrate the biological efficacy of *Sophora flavescens* plant extract in controlling mites in tomato crops. For this purpose, three variants were installed. The first variant was witness, the second variant (standard) the insecticide Pelecol 0.8 l/ha, the third variant was used the extract from *Sophora flavescens*. 14 l/ha. No treatment was applied to the witness plants, so the development of mites was very rapid. Each of the variants had 3 repetitions. The degree of attack and the intensity of development were determined according to the method described by A. Hrapova (Roșca *et al.*, 1996). The first treatment was performed on 13.06.2022. Before testing the extracts on tomato plants, 12-26 mites were registered on the leaf. The application of the treatment contributed to the mortality of the pest both on the plants from the variant of the tested preparation and on the one from the standard variant.

Evaluating the data obtained after treatment, a decrease in the number of pests was observed in relation to the untreated plant. Thus, the biological efficacy of *Sophora flavescens* extract after the 3rd, 7th and 14th day on average was 80%, in the Etalon variant, it was 91%. From these data it results that the treatment of tomato plants with *Sophora flavescens* plant extract reduces the spread of mites, although it has demonstrated a slightly lower biological efficacy compared to the standard.

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Keywords: *Sophora flavescens*, plant extract, biological efficacy, tomato, red mite.

INFLUENCE OF MINOR COMPONENTS ON THE EFFICIENCY OF APPLE WORM SEX PHEROMONE

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The apple worm is the main pest of apple trees, leading to losses of up to 70-90% of production. Today, there is an intensive development of organic farming in the world. In this regard, the development of non-invasive methods and environmentally friendly products used in the biological protection of plants, including through the use of sex pheromone traps, is of major importance for agricultural producers worldwide. The use of approved pheromone-based preparations can minimize population density, which is extremely important for the production of a rich harvest of ecologically pure products. The role of minor components found in the literature is currently being investigated, for example, different authors list up to thirteen components for the apple pheromone sex pheromone, where their ratio varies in different compositions. In our work, we studied the effect of two minor components on the effectiveness of the codling moth sex pheromone.

In the laboratory "Integrated Plant Protection", within the IGPPP, the main component E8,E10-12:OH was synthesized and variants with different two-component pheromone compositions were prepared where the minor component dodecanol (C₁₂H₂₅OH) was added to the synthesized basic component in the amount of 5%, 10%, 30%, 60%, and the minor component tetradecanol (C₁₄H₂₉OH) in the amount of 3%, 6%, 12%, respectively, which were subsequently impregnated on rubber septas, according to the scheme of experiments, with the formation of pheromone sets on variants, composed of the delta-shaped trap body, sticky plate and rubber septa.

In 2020, experiments were performed in the apple orchard of the Scientific - Practical Institute of Horticulture and Food Technologies in Codru town. The observations were made with an interval of 5-7 days, the sticky plates were changed once in 15 days. From the obtained data it was found that the use of dodecanol as a minor component in the pheromone composition increased its biological efficacy. In variants where 60% and 30% of the minor component dodecanol were added, the average male catches increased by 56% and 62% respectively. An analogous trend in the use of the minor component tetradecanol was observed. In this case, the number of males caught in delta-traps with pheromone compositions where 12% and 6% of tetradecanol were added, increased by 29 - 35%, the statistical analysis did not confirm the differences observed due to the variability of the data obtained. In this way, a significant positive effect of the influence of minor components in two-component pheromone compositions was observed.

Acknowledgments: This work is a product derived from the State Program "Strengthening capacities of forecasting and control of harmful organisms and phytosanitary risk analysis in integrated plant protection" (Nr. 20.80009.5107.19).

Keywords: apple worm, pheromone, dodecanol, tetradecanol, biological efficacy.

RELATIONSHIP BETWEEN RELATIVE MATURITY AND GRAIN YIELD IN EXPERIMENTED NEW MAIZE HYBRIDS

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Very variable weather in the current changing climatic conditions, affects the establishment of the vegetation period of crops and exposes crops to failure, if farmers choose crops that are not adapted to relative maturity and that do not match the characteristics of the growing period. This study aimed to determine the relationship between the relative maturity of maize hybrids and grain yield and to determine the period of relative maturity that will support grain production in corn. In this experiment, a set of maize hybrids with different relative maturity were cultivated within the Institute of Crop Science „Porumbeni”, in the years 2020-2021. The total number of 200 maize hybrids were grouped into 4 maturity groups, entitled confirmed an approved native hybrid, depending on the relative maturity that ranged from 119 to 135 days. Maize hybrids were grown in optimal growing conditions. From the results obtained, it was observed that the control hybrids had a very low grain yield compared to the new hybrids experienced. In the first group, where 13 new hybrids were tested, the control hybrid Porumbeni 310, had 120 days until the relative maturation, and the grain yield was the lowest, namely 78.5%. In this group the new hybrids had from 119 to 124 days to full maturity, and the grain yield varied from 81.03% to 88.7%, which we can say that the grain yield of new hybrids was higher than the control, on average by 7.0%. The second group included 43 new hybrids and a control hybrid Porumbeni 374. The control hybrid had 122 days until full maturation and a grain yield of 80.7%, that is, 3% lower than the yield of newly experienced hybrids. The relative maturation of the hybrids in the second group varied between 124 and 128 days, 3 days longer than the control hybrid. The grain yield of new hybrids in this group was from 78.8% to 88.5%. The third group registered 45 new hybrids and the control hybrid Porumbeni 427. The number of days until full maturation was 127-132 days, depending on the year of experimentation, and the control hybrid recorded 125 days until maturation. The grain yield in new hybrids ranged from 78.5% to 87.6%, while the control hybrid had a yield of 80.8%, lower on average by 4%, compared to the grain yield in new hybrids. The newly tested hybrids, the fourth maturity group, were witnessed by the local hybrid Porumbeni 461, which recorded 130 days until full maturation and a grain yield of 77.9%, and which was the lowest grain yield, from all experienced hybrids. In contrast, the new hybrids tested in this group had full maturation from 129 days to 135 days, and the grain yield ranged from 78.6% to 87.5%, which is 6% higher than the grain yield recorded at the control hybrid.

On average, we recorded full maturation in the first group at 120 days, in the second group at 125 days, in the third group at 128 days and in the fourth group at 130 days. The average grain yield in each group was 84.6% in the first group, 83.6% for the second group, 83.9% in the third group and 83.6% in the fourth group, respectively. So, we can say that the obtained results showed an insignificant increase in grain yield in earlier maturing hybrids and an insignificant decrease in grain yield in later hybrids.

This experiment showed that relative maturity had an insignificant impact on grain yield in maize hybrids, however, medium and semilate maturing hybrids had low grain yields.

Keywords: hybrid, maize, maturation, grain yield.

BIOPESTICIDES - AN ALTERNATIVE AND ECO-FRIENDLY SOURCE FOR THE CONTROL OF PESTS

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Chemical pesticides are widely used worldwide but they are ecologically unacceptable. Use of chemical pesticides has resulted in the disturbance of our environment (Gulhane *et al.*, 2015). Due to the side effects of chemical pesticides, sustainable crop production through eco-friendly management is essentially required in the current scenario the biopesticides is ecofriendly pest control pesticides (Hubbard *et al.*, 2014). Presently, about 8.5% of EU's agricultural area is farmed organically, and the trends show that with the present growth rate, the EU will reach 15-18% by 2030.

The global biopesticides market is projected to grow at a CAGR of 14.7% from an estimated value of USD 4.3 billion in 2020 to reach USD 8.5 billion by 2025. Widely used biopesticides are living organisms which are mostly pathogenic for the target species only These biopesticides are good alternatives to conventional insecticides for sustainable management of insect pests (Prabha *et al.*, 2016).

The present paper gives information of *Bacillus thuringiensis* ssp. *kurstaki* preparats, we discuss how evolution, host range determination and pathogenesis have contributed to their inherent safety for non-target organisms including humans. The entomopathogenic microorganismsalso can accumulate themselves in the environment and the host population, and it can control the pest insects for a long term by forming epidemic disease in the pest insect population through the external environment stimulation. The quite stable food chain relation of plant-pest insect- natural enemy can be gradually established. Thus, it can reduce the risk of the pest insect continuous outbreak and realize the persistent control of pest insects.

Bacillus thuringiensis (Bt) are bacteria found naturally in the environments of every continent of the world. Some of the Bt strains are natural antagonists of some pests. A few Bt strains have been selected and commercialized as biological pest control products. These commercialized Bt strains are valuable to agriculture and public health because of their unique ability to naturally control certain destructive and disease-carrying insect pests while avoiding harm to non-target organisms (such as beneficial insects, people, other mammals, and fish). Biological insect control products based on Bt strains have been used safely and effectively in practical field conditions for more than 50 years.

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Keywords: *Bacillus thuringiensis* (Bt), biopesticide, pathogenesis, target specie, pest.

**BIO-ECOLOGICAL PECULIARITIES OF SOME NEW TAXA OF
BERBERIS THUNBERGII DC.**

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The implementation of the Global Biodiversity Strategy aims at a series of high-priority goals, one of them being the enrichment and diversification of the gene pool in green spaces. The representatives of the genus *Berberis* L. are high-value honey and ornamental plants. The genus *Berberis* L. belongs to the family *Berberidaceae* Juss. The genus *Berberis* L. includes about 175 species and a very large diversity of taxa. They occur in the temperate zone of Europe, Asia and North America. Thirty-one species and several varieties occur in the Republic of Moldova. In the last 4-5 years, in the "Alexandru Ciubotaru" National Botanical Garden (Institute) (hereinafter NBGI), 24 new taxa of Japanese barberry, bred in Poland, have been introduced and need to be researched, assessed bioecologically under the new growth conditions for further use. The goal of this research included the identification of the bioecological peculiarities of 4 *Berberis de thunbergii* DC. taxa – ‘Maria’, ‘Green Cloud’, ‘Red Torch’,

‘Dart’s Red Lady’ under new *ex-situ* conditions and the appreciation of the prospects of their use in landscaping.

The research was conducted in 2019-2022 in the plant nursery of the Dendrology Laboratory, within the project 20.80009.7007.19 "The introduction and development of technologies for propagation and cultivation of new species of woody plants by conventional techniques and tissue culture". The respective taxa, which grow in the collection of NBGI, are currently in the third and fourth growing season. The morphological parameters were determined for 10 plants of each taxon, as well as for 100 flowers and shoots. The frequency of flowers and fruits per 20-cm-long shoot and the fruit yield were determined in the fourth growing season. The researched taxa are tolerant to drought, frost, noxious substances; they do not require special care, only regular trimming to keep their shape and compliance with standard technology throughout the growing season. The studied shrubs are particularly showy in early spring, due to the beautiful yellow colour of the flowers, the abundance of flowering, the density of flowers and shoots per 20-cm-long shoot, as well as in early summer – due to the shade of the foliage, the shape and size of the plant and fruits and the various colors of the fruits (red, orange-red, purple-red), the abundance of fruiting, the long period of flowering and fruiting.

The studied taxa differ significantly in plant height, which varied from 36 cm in *Berberis thunbergii* ‘Green Cloud’ to 114 cm in *Berberis thunbergii* ‘Red Torch’. They also differ in crown diameter, which ranged from 50-52 cm (in the taxa *B. th.* ‘Maria’, *B. th.* ‘Green Cloud’) to 70-72 cm (*B. th.* ‘Red Torch’, ‘Dart’s Red Lady’). The density of fruits per 20-cm-long shoot varied from 2 pcs. (*B. m.* ‘Dart’s Red Lady’) to 14 pcs. (*B. th.* ‘Red Torch’). The taxon *B. th.* ‘Red Torch’ is characterized by a high percentage of fruit bearing (80.1%), *B. th.* ‘Dart’s Red Lady’ – 30.6%, and the other taxa have intermediate values of this index. The fruit bearing percentage correlates with both the biotype and the climatic conditions during the process of ontomorphogenesis.

Keywords: *Berberis thunbergii*, morphological parameters, biotype, growing season.

TRANSCRIPTOME ANALYSIS AND RESISTANCE GENE MINING OF SUNFLOWER RESISTANT *OROBANCHE CUMANA* RACE G

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Orobancha cumana (sunflower broomrape) is an obligatory, non-photosynthetic root parasitic plant that specifically infects the sunflower. Broomrape causes severe yield losses of sunflower. In order to control broomrape infestation, some resistance genes are being uncovered by our RNA-seq analysis. In this study, two sunflower varieties (resistant/sensitive sandaomei strain) were selected and inoculated with broomrape respectively. During the interaction between sunflower and broomrape, sunflower roots were sampled at germination stage, small nodule stage and large nodule stage, and transcriptome sequencing was performed. We finally deployed the Deseq2 assay to screen differentially expressed genes between resistant cultivars and sensitive cultivars. A total of 2,058 differentially expressed genes were obtained during germination stage, including 1093 up-regulated genes and 965 down-regulated genes. A total of 1541 differentially expressed genes were obtained at small nodule stage, including 867 up-regulated genes and 674 down-regulated genes. A total of 1231 differentially expressed genes were obtained at large nodule stage, including 327 up-regulated genes and 904 down-regulated genes.

The KEGG enrichment analysis showed that the up-regulated genes were mainly enriched in the Ras signaling pathway, Phenylpropanoid biosynthesis, Plant-pathogen interaction, Plant-pathogen interaction, MAPK signaling pathway, and these pathways related to resistance response in the germination stage and down-regulated genes mainly focus on Phenylalanine metabolism, Pentose and glucuronate interconversions. At the small nodule stage, the up-regulated genes mainly enriched in Ras signaling pathway, Aldosterone synthesis and secretion. The down-regulated genes mainly focus on Phenylpropanoid biosynthesis, Pentose and glucuronate interconversions, p53 signaling pathway. At the large nodule stage, the up-regulated genes mainly enriched in Metabolism of xenobiotics by cytochrome P450, beta - Alanine metabolism, beta-Alanine metabolism. The down-regulated genes mainly focus on Ras signaling pathway, Cutin, suberine and wax biosynthesis. Ras Signaling Pathway was up-regulated at germination stage and small nodule stage, and down-regulated at large nodule stage, indicating that this pathway was the key signaling pathway for sunflower resistance to broomrape.

Keywords: *Orobancha cumana*, transcriptome, sequencing, gene expression, germination stage, metabolism.

FIELD RESISTANCE OF SUNFLOWER VARIETIES RESOURCES TO SEED RUST SPOTS DISEASE

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Sunflower seed rust is an emerging disease found in the production in recent years. The disease is characterized by irregular rust spots on sunflower husk. Its occurrence had seriously affected sunflower appearance for seeds serving, and was becoming a bottleneck for sunflower industry. Field experiments were carried out in Wulateqian County and Wuyuan County in Bayannur, Inner Mongolia, China. In these areas, sunflower seeds suffered seriously. The resistance to seed rust spots was evaluated on 44 sunflower cultivars, including 39 confectionery sunflowers and 5 oil sunflowers. The diseased scales (0-4) were scored based on following criterion. level 0 = No rust spot of seeds on the whole capitulum, level 1 =Rust spots on the seed surface of all rust-spotted seeds account for an average of 1% to 25%, level 2 =The proportion is an average of $\geq 25\%$, level 3= The proportion is an average of $\geq 50\%$, level 4 = The proportion is an average of $\geq 75\%$. Seed rust spot rate (%) = number of rust spots on seeds / number of seeds $\times 100$. For each varieties, three replications with 15 plants were scored.

Results showed that, among the confectionery sunflowers in Wulateqian County, rate of seed rust spots of Sairui 1 was the lowest ($< 25\%$). 6 varieties had lower rates between 25% and 50% (such as JK 103, LJ 188, LD 5009, etc.), the others were more than 50%. Considering the severity levels, 20 varieties were in level 1, but with the disease rate ranging from 17.30% to 88.63%. In Wuyuan County, the disease rate was 61.02% to 99.63%, significantly higher than the same varieties in Wulateqian County, but with lower level. Only 10 varieties were in level 1 such as 3638C. For oil sunflower, 4 varieties were lower than 25%, except for XKS2029. In addition, rust occurrence of the same variety at different sowing times also showed significant differences. For example, for SH361, the rate was 34.49% on the sowing date of May 27th, but 98.47% on June 20th. It indicated that different sowing dates effected the occurrence of seed rust spots.

Keywords: sunflower, seed, disease, varieties, rust.

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Session B

PLANT, ANIMAL AND MICROBIAL BIOTECHNOLOGY



THE BIOMASS QUALITY OF CHIA *SALVIA HISPANICA* L. AND PROSPECTS FOR ITS USE IN MOLDOVA

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Chia *Salvia hispanica* Lamiaceae family, also called Mexican chia or salba chia, is an annual herbaceous plant, native to Mexico and Guatemala, where it was an important crop for pre-Columbian Aztecs and other Mesoamerican Indian cultures, characterized by its high nutritional and therapeutically potential. Chia is mainly cultivated as a seed crop. The culinary uses of chia seed have been as a whole seed, seed flour, seed mucilage, and seed oil. *Salvia hispanica* seeds are one of the most-concentrated sources of alpha- linolenic acid (ALA), a plant-based omega-3 fatty acid. Recently interest has been raised on biomass production as a potential forage source opening alleys toward the integration of chia in crop-livestock systems, also as feedstock for renewable energy production.

The objective of this research was to evaluate the quality of green mass from chia *Salvia hispanica* cultivated under the conditions of the Republic of Moldova, and the possibility to use them as fodder for animals and feedstock for biogas production, also the biomass quality of chia dry stalks after harvesting the seeds as feedstock for the production cellulosic ethanol.

We found that the dry matter of the harvested whole plants contained 107 g/kg CP, 80 g/kg ash, 347 g/kg CF, 348 g/kg ADF, 517 g/kg NDF, 65 g/kg ADL, 283 g/kg Cel,

199 g/kg HC, 123 g/kg TSS, with nutritive value 61.79% DMD, RFV= 111, 12.19 MJ/kg DE, 10.1 MJ/kg ME, 6.03 MJ/kg NEL. The nitrogen content in the green mass substrate was 1.71% and estimated content of carbon– 51.1%, the C/N = 29.9 respectively, which played an important role in degradation and biomethane production. The biochemical methane potential of *Salvia hispanica* green mass substrate reached 298 l/kg ODM.

The analysis of lignocellulose composition of chia dry stalks suggested that energy biomass contained 357 g/kg cellulose, 197 g/kg hemicellulose and 72 g/kg acid detergent lignin. The estimated theoretical ethanol yield from cell wall carbohydrates averaged 398 L/t.

Chia *Salvia hispanica* can serve as multi-purpose crops for seed production, alternative forage and feedstock for renewable energy production.

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Keywords: *Salvia hispanica* L., fodder, green mass, biomass quality.

THE INFLUENCE OF THE AGRICULTURAL MANAGEMENT SYSTEM ON THE SOIL MICROBIOME

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Soil is the fundamental resource of an agricultural ecosystem. Agricultural practices influence the composition of soil bacterial communities. The aim of our work was to study the structure of the soil microbiota in the case of different crop rotation and types of fertilization. The research was carried out in the long-term field experiment on the “*Biotron*” Experimental Station of the Academy of Sciences of Moldova in two crop rotations (with and without alfalfa). Characterization of the compositional diversity of the soil microbiome was achieved by sequencing amplicons targeting the 16s rDNA gene of prokaryotes (Scientific Center “Genomic Technologies, Proteomics and Cell Biology” of FSBSI ARRIAM, St. Petersburg, Russia).

Anthropogenic soil loading, agricultural management system influenced the formation of soil prokaryote communities. This influence is evident from the analysis of the abundance of *Actinobacteriota* phylum, which in our experiment was second in abundance after Proteobacteria. In the variant *Mineral background* (N45-90P30-60K60-90) was determined the highest abundance of *Actinobacteriota* phylum, being 19.0% in the alfalfa crop rotation and 20.1% in the rotation without alfalfa, respectively. The soil in the forest strip was characterized by the lowest abundance of *Actinobacteriota* filum - 16.0 %. Actinobacteria of the family *Sterptomycetaceae* had the highest relative abundance in the alfalfa rotation, variant *mineral background* (N45-90P30-60K60-90). The forest floor is exposed to less anthropogenic pressure and some oligotrophic-nutrient phyla are more abundant than in agricultural soils. The *Acidobacteriota* phylum had the highest abundance in the forest floor soil - 5.7%. These results can be explained by the fact that *Acidobacteriota* have mostly oligotrophic nutrition strategy with low growth rates and seem to be favoured in resource-limited conditions due to high substrate affinities. The group of microorganisms (clade) *Allorhizobium-Neorhizobium-Pararhizobium-Rhizobium*, which fixes atmospheric nitrogen and promotes plant growth, has the highest share - 1.1 % in the crop rotation without alfalfa, *Organic Fund* variant. The genus *Ensifer*, which includes bacteria capable of inducing root-knot formation in legumes, was detected only in the alfalfa soil with the mineral background variant (N45-90P30-60K60-90). The genus *Streptomyces* (biocontrol agents) had the highest relative abundance (1.3%) in the alfalfa soil with the organic backgroundvariant.

Agricultural practices alter the diversity and composition of soil microbial communities, and these altered communities impact the functioning of agricultural ecosystems. Organic fertilization leads to a significant increase in the abundance of some nutrition-related bacteria.

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Keywords: soil microbiome, crop rotation, fertilization, diversity.

DIRECTING THE GROWTH OF CARP LARVAE THROUGH THE APPLICATION OF THERMAL FACTOR

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The study aims to highlight the correlation between environmental parameters (temperature of the aquatic environment) and the consumption of nutrients, in particular, of the yolk sac, as well as the determination of the resistance and growth rate of fish larvae. At the same time, the study was carried out in order to determine the parameters that could be used as a method of increasing the adaptive capacities of animals to the unfavorable action of the environment. The biological material was represented by fish of the species *Cyprinus carpio* subjected to the application of low temperatures in different periods of postnatal ontogenesis. The experiment was performed on two batches of carp larvae aged 1 and 3 days. Each batch of larvae was divided into 3 experimental sublots, in which temperatures of 10 °C, 14 °C and 20 °C were applied. The experiment was carried out in glass vessels with a capacity of 3 liters of water and a density of 500 larvae per liter. The adaptation period to the tested temperatures was 1 hour. The period of application of low temperatures was 1, 3, 5, 7, and 10 days. After the end of the low temperature application period, the carp larvae were transferred to vessels with a water temperature of 20 °C and received abundant food. The studied parameters were monitored at the end of the application periods of low temperatures and additionally at the end after 10 days of feeding the larvae.

The analysis of the obtained data proves that the number of carp larvae aged 1 day, survived after the application of low temperatures is higher than the number of carp larvae aged 3 days and, correspondingly, constitutes 92,83% compared to 80,14%. At a temperature of 20 °C the number of larvae in experimental groups I and II constituted 87,82% and 66,33%, respectively. After 10 days of feeding the larvae, their number in experimental group I is - 83,6%, and in experimental group II - 73,1%.

Further research has focused on determining the influence of environmental factors on the resorption of the yolk sac of carp larvae. Therefore, in the experimental group I the presence of the yolk sac was registered throughout the experimental period (10 days) at application of the low temperature of 10 °C and 14 °C. At the same time, at the temperature of 10 °C the size of the yolk sac at the age of 5 days did not differ significantly from its initial dimensions. In experimental group II, the presence of the yolk sac was registered only at the temperature of 10 °C until the age of 7 days of ontogenesis. At a temperature of 14 °C the yolk sac was practically no longer present at the age of 5 days of the larvae. At the temperature of 20 °C the presence of the yolk sac was registered only up to 3-4 days of application of the thermal factor. Thus, it can be mentioned that the application of low temperature on carp larvae leads to retention of their development with the keeping of yolk sac for a period of up to 10-12 days after birth. These results are registered at a temperature of 10 °C in the group in which the larvae at the beginning of the experiment were one-day old. Therefore, by applying the thermal factor, it is possible to manage the duration of the development period of carp larvae and to maintain them in case of need for a longer time in the breeding fish farms.

Keywords: fish larvae, adaptive capacitie, thermal factor.

THE ROLE AND IMPORTANCE OF REPRODUCTIVE BIOTECHNOLOGIES IN ANIMAL BIODIVERSITY

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The role and importance of recent biotechnology is constantly growing due to the need to maintain and conservation of biodiversity, as well as increase animal productivity. One of the main biotechnologies of interventions in increasing the improvement of livestock is the continuous improvement of the reproductive properties of the most valuable males, selected from existing livestock based on scientific criteria argued biologically and technologically. Also, the increase of the rhythm of improvement of the herds, foresees the wide application of the reproductive biotechnologies by methods, procedures and operations necessary for the obtaining in optimal conditions, of new generations of animals. The performance of efficient reproductive biotechnologies and their monitoring are imperative for sustainability in any system of breeding and obtaining products of animal origin. Therefore, the existence of obvious challenges for increasing animal productivity in changing environmental conditions can be achieved through conventional breeding biotechnologies. The main effects of the application of reproductive biotechnologies are both in the accelerated improvement of livestock and their health, and in the conditions of reducing the cost price per unit of product. The emergence and use of modern reproductive biotechnologies have opened many avenues for scientific research into the reproductive phenomenon both *in vitro* and *in vivo*. For example, the technology of sex sorting of sperm, which is an advantage in raising animals, to obtain offspring of the desired sex, either male or female. Some biotechnologies also include certain biotechnologies, among which is the induction of estrus and ovulation in physiological off-season, the induction of polioovulation, the synchronization of estrus, the programming of calvings, etc. Animal breeding biotechnologies are currently an area of major importance and performance, a new and promising field of contemporary biology. At the fundamental scientific level with experimental applications, the effects of sperm encapsulation are currently being investigated for a longer preservation of sperm *in vivo*. This biotechnology has been designed to extend the life of sperm at body temperature and to allow the progressive release of viable sperm over several days in various species, including humans. Moreover, biotechnology research is currently being applied on the transcriptomics of the mRNA study at various stages of development, including spermatogenesis; identifying different biomarkers in semen and predicting fertility; biotechnology of intracytoplasmic sperm injection used to treat male infertility, as well as the application of other biotechnologies in various allied fields such as genomics, proteomics, bioinformatics, etc., which are already used in various fields of reproduction, including various animal species. Therefore, there is a need for a clear policy for the correct application and handling of biotechnologies with a multi-institutional approach to solving animal reproduction problems.

Keywords: biotechnologie, biodiversity, animal breeding, reproduction problems.

**RESEARCH ON THE INVASIVE IMPACT OF HARMFUL INSECT
COMPLEXES ASSOCIATED WITH PARASITIC NEMATODES
AND PATHOGENIC VIRUS VECTORS IN PRODUCTIVE PLUM
ORCHARDS**

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The cultivation of plum is a centuries-old tradition, surpassed only by apple, and the production of plum fruits obtained in the Republic of Moldova is over 35%, with an important share in exports, about 30%. Plum is such an appreciated crop due to the high productivity of the trees, the special qualities of the fruits and the numerous valuable varieties with diverse fruiting time. The current plum orchards are capital investments, which can be exploited for 20-25 years, and the efficiency of their use depends on the correctness of the establishment of orchards and care techniques in the first 4-5 years after planting.

One of the priorities in the cultivation of plums is the study of the evolution of the parasitic vectorial impact of microbial infections of complexes of harmful organisms, such as associations of harmful insects and invasive nematode complexes in the soil, which also act as vectors of pathogenic viruses, triggering viral infections, which usually occur in pathosystems, triggered by the development of perennial monoculture of plum plantations in time and space. According to the research programs developed based on the project – State Program 2020-2023, we started addressing these strategic objectives in the development of the horticultural sector in the Republic of Moldova, where research on the phytosanitary biological control of invasive and vector insect and nematode communities is conducted annually, to establish the parasitic, functional impact and the structure of associated populations in young and productive plum orchards, breeding nurseries, including other related species such as peach, apricot, cherry etc.

The intensive plum orchards, nurseries for reproducing the planting material from 6 administrative districts, on areas of over 500 hectares, in the North zone (Briceni, Soroca), the Center zone (Criuleni, Nisporeni), the South-East zone (Căușeni, Ștefan-Vodă) of R. Moldova were investigated. More than 300 samples of soil, young trees, young shoots, young seedlings attacked by viruses were taken from various systems of orchards and breeding nurseries. The soil samples were taken at a depth of 30-60 cm, at intervals of 15-20 days, covering the main phenological stages throughout the growing season. To establish the entomo-helminthiotoxic and virotic parasitic impact, we performed visual and optical observations to identify local and extensive damage, with symptoms of specific viral infections on trees. The results of the analyzes are interpreted by the values of abundance and frequency of the species (insects on average per 100 leaves, 100 fruits analyzed; nematodes – individuals / 100 cm³ soil), classified by ecological-trophic specialization depending on the age of plantations, environmental conditions, variety, soil, maintenance systems.

As a result of the phytosanitary records carried out during the growing season, in 2020-2022, on the entomo-parasitic impact of the range of invasive insects, a very strong attack was periodically detected, which caused significant losses in fruit production and quality. Their presence in plum orchards is quite difficult to detect, they are often detected too late and spraying insecticides is no longer effective. The damage is usually very extensive; in severe cases the entire plum production is compromised. The most dangerous species detected on plum trees in new type productive orchards are: *Cydia funebrana* – plum fruit moth, *Cydia molesta* – oriental fruit moth, *Hyalopterus pruni* – mealy plum aphid, *Hoplocampa minuta* – black plum sawfly, *Eurytoma schreineri* – plum seed wasp, species of ectoparasitic aphids and vectors of pathogenic viruses of the genus *Aphis sp.*, *Myzus sp.*, some species of cicadas, mites etc. For this reason, farmers should pay particular attention to spraying insecticides.

The concomitant nematological surveys are also very important, because they also occur as invasive forms and vectors of pathogenic viruses, detected in plantations of various fruit species, including plum. This includes nematode species established as specialized complexes on plum trees, where the most common species that form associations and belong to the orders *Tylenchida* and *Dorylaimida*, the genera: *Pratylenchus*, *Pratylenchus*, *Tylenchus*, *Helicotylenchus*, *Ditylenchus*, *Xiphinema spp.*, *Longidorus*, *Trichodorus*, with endo-ectoparasitic specialization and specialized vectors of various forms of pathogenic viruses, which can trigger specific viral infections in fruit plantations. According to the bibliographic review and the results obtained by us, currently the presence of 15 species of nematodes with invasive impact and vectors of pathogenic viruses specific to plums have been identified. They are represented by associations of the order *Dorylaimida*, the genera *Xiphinema* and *Longidorus*, the species: *Xiphinema index*, *X. vuittenzei*, *X. riversi*, *X. brevicolle*, *X. diversicaudatum*, *Longidorus elongatus*, responsible for the transmission of 20 species of pathogenic viruses and viral infections in plum orchards, detected in practically all areas and sectors investigated.

The investigations carried out in the intensive plum orchards have also highlighted the more advanced etiological composition of 3 viral diseases (ring pattern virus, fan-leaf virus, plum pox virus), detected immediately after the formation of vegetative organs (first growing season), infested and through the numerous insect species with sting-sucking buccal apparatus, such as: aphids, cicadas, mites, wasps, which are associated with complexes of vector nematodes in the soil, especially in the sectors that were previously planted with classic orchards, frequently detected in all zones and growing seasons.

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Keywords: plum orchard, harmful insect, parasitic vector, nematode, viral diseases.

VIABILITY OF STREPTOMYCES STRAINS AND ITS VARIANTS AFTER FREEZE-DRYING IN CNMN

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There are currently more than 20 types of strain preservation methods, which can be divided into four categories: subculturing, drying, freeze-drying, and cryopreservation.

All modern methods of preservation and long-term storage of microorganism collection cultures are based on the transfer of cells to an anabiotic state with an appropriate partial state (storage on medium with minimal necessity of nutrients, in sterile vessels, under a layer of mineral oil, in distilled water, at low temperatures, etc.) or complete (drying, freeze-drying, cryopreservation, etc.) cessation of metabolism. Each of the methods has its own advantages and disadvantages and can have a different effect not only on the viability, but also on the preservation of the characteristic properties and physiological, biochemical, and genetic features of the culture. In connection with the need to develop integrated approaches to the conservation of microorganisms, it is important to note that the practice of conservation, which has developed over many decades, has empirically developed a number of examples which, to one degree or another, correspond to those mechanisms. Immersion of cells of microorganisms in an anabiotic state which have been identified (and continue to be detected) in the study of the formation, properties and germination of specialized resting cells of microorganisms.

Thus, freeze-drying of microbial strains is one of the most sustainable methods, but it requires the greatest manpower, special equipment and trained personnel. Thus, the purpose of the research was to evaluate the viability of *Streptomyces* strains maintained in the National Collection of Non-Pathogenic Microorganisms (CNMN) after freeze-drying to determine the effectiveness of the method.

The basic strains and their variants served as objects of study: *Streptomyces levoris* (CNMN-Ac-01, CNMN-Ac-15, CNMN-Ac-16); *S. mauvecolor* (CNMN-Ac-12, CNMN-Ac-17, CNMN-Ac-18); *S. gougerotii* (CNMN-Ac-14, CNMN-Ac-19, CNMN-Ac-20).

For the long-term preservation of lyophilized *Streptomyces* strains, protective medium gelatin 2.5 % + glucose 7.5 % was used. The contents of the vials with sporulating material was frozen at -50°C. Freeze-drying taken place for 8 hours, at the Labconco 6 plus freeze-drying machine at a pressure of 6-7 Pa and a temperature of -94°C. The determination of the viable germ load by inoculation on solid culture media, the cells that cause colony formation are called colony forming units and their number is approximately equal to the number of microbial cells in the sample. According to results the viability of all 9 strains after freeze-drying varying between 88.7 – 97.8 %. Freeze-drying was successful, because the viability of the strains was higher than 85.0 %.

Acknowledgments: The research was funded out within the project 20.80009.7007.09 (ANCD).

Keywords: microbial strains, viability, conservation method, freeze-drying method.

SPECIFIC STREPTOCOCCI OF THE DIGESTIVE TRACT AND THEIR SUITABILITY FOR INCLUSION IN PROBIOTIC PREPARATIONS

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It is now known that normal intestinal bacteriocenosis consists of bacilli and cocci. The elaboration of probiotic biopreparations, intended to reconstitute its deregulated composition, is mainly carried out with the use of bacteria in the form of bacilli, and of cocci - very limited (only *Enterococcus faecium* species).

The aim of the present work was to study the background of intestinal streptococci in healthy children of various ages and to argue their use in the development of microbial composition of new probiotic biopreparations.

The investigations were carried out in several steps. The first was devoted to the isolation of streptococci currently existing in the healthy intestinal contents; the second - to their identification and the third - to the calculation of the percentage share of various genera of intestinal streptococci specific to children of various ages. As a result, 834 streptococcal monotypic isolates were isolated and assigned to three genera of the *Streptococcaceae* family. The percentage share was higher in streptococci specific to children aged 1-3 years (genus *Lactococcus*), constituting 46.31-46.98%. In second place were those of the genus *Streptococcus* (31.57-37.73 %) and third - *Enterococcus* (18.66-22.10 %). In children aged 5-16 years, there was a predominance of enterococci (38.18-46.77%), a sharp decrease in lactococci (28.18-13.70 %) and a relative maintenance of those of the genus *Streptococcus* (33.92-39.51 %).

Thus, in healthy children of various ages it was found:

- the incidence of some intestinal representatives of the family *Streptococcaceae*;
- their genus membership and the percentage share of streptococci of the genera useful to the body.

It was demonstrated the opportunity of recommendation and differentiated use of streptococci from the studied genera in case of elaboration of new composition of probiotic biopreparations, intended for children of 1-3 years (genus *Lactococcus*), 5-16 years - (*Enterococcus*) and 1-16 years - (*Streptococcus*).

Keywords: intestinal streptococci, children, biopreparat, probiotic effect.

**NEW DATA ON LADYBUGS (INSECTA) FROM THE REPUBLIC OF
MOLDOVA**

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Ladybugs (Insecta: *Coccinellidae*) are beneficial, predatory insects that enjoy increased attention from researchers due to their high potential as biological control agents in organic agriculture. Despite its small size, ladybugs consume a large number of plant pests such as aphids, thrips, mites, scale, whitefly populations etc.

The first study of ladybugs species diversity from the Republic of Moldova was performed, after which 48 species from 28 genera and 7 tribes belonging to the family Coccinellidae were identified. Among the most common species native for the Republic of Moldova are *Adalia bipunctata*, *Coccinella septempunctata*, *C. undecimpunctata*, *Coccinula quatuordecimpustulata*, *Hippodamia variegata*, *Tythaspis sedecimpunctata* and invasive *Harmonia axyridis*.

Acknowledgments: Research was supported by the project Nr. 20.80009.7007.02.

Keywords: insect, biological control agent, diversity, Republic of Moldova.

**INITIATION OF THE *IN VITRO* CULTURE OF THE
MACROCARPON VACCINIUM AITON VARIETY 'EARLY BLACK'**

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The research was carried out in the Laboratory of Embryology and Biotechnology of the National Botanical Garden (Institute) "Alexandru Ciubotaru". Cranberry (*Vaccinium macrocarpon* Aiton), is a shrub about 20-30 cm tall, with red fruits (1-2 cm in diameter), evergreen leaves and white or pale pink flowers. Fruits have multiple health benefits, an increased content of protein, vitamins A, B1, B2, C, minerals, tannins, fiber, flavonoids, saponins, phenols and essential fatty acids Omega 3, 6 and 9, oils and sugars, are energizing, helps eliminate stress, anxiety and depression. Based on the nutritional and therapeutic importance of cranberries, research has been initiated on obtaining seedlings by in vitro multiplication. During the research, several sterilization procedures were developed. According to the literature for the inoculation of *Vaccinium* species is used plant material consisting of shoots fragments containing apical buds or nodal fragments, harvested from mature plants.

The explants were taken from the donor plants in June, during the active vegetation period of the plants. Fragments of shoots with apical buds were taken for inoculation. The biological material was brought to the laboratory, where it was fragmented and washed under cold water. After washing with detergent solution, the fragments were placed in KMnO₄ solution (about 3%) to which 2.3 drops of Tween were added for 10 min. The biological material was then transferred to the box, rinsed 4 times with sterile distilled water (autoclaved) and sterilized. For this species we used 3 sterilization regimens: 1) the plant material was sterilized with a solution of mercuric chloride (ethanol mercury chloride) 0.1% for 5 minutes; 2) the plant material was aseptitized with 0.05% chlorhexidine solution for 5 minutes; 3) the plant material was sterilized with 0.25% lysoformin solution for 10 minutes. This was followed by rinsing with sterile distilled water, then placing the biological material in 3% H₂O₂ solution (for 1 min.) And repeated rinsing with sterile distilled water. After this process, the biological material was fragmented under the laminar and inoculated into test tubes (one inoculum in the test tube) on agarized nutrient medium: WPM - 100%, zeatin - 0.5 mg / l, sucrose - 30 g / l, agar - 5 g / l, pH- 5.0.

Following the research, we established that exposure to these sterilization reagents did not cause necrotization of the explants, however the most beneficial method of asepsis for the given culture is with mercury chloride of 0.1% (5 minutes), the process of infestation of explants being less than 50%. After sterilization with a chlorhexidine solution of 0.05%, about 30% of viable inoculums were obtained, with a solution of lysoformin of 0.25% - about 10%.

Acknowledgments: The research was carried out within the project: 20.80009.19 "Introduction and elaboration of technologies for multiplication and cultivation by conventional techniques and in vitro cultures of new woody plant species".

Keywords: *Vaccinium macrocarpon*, variety, in vitro multiplication.

**INFLUENCE OF SOME COORDINATION COMPOUNDS WITH
POLYDENTATE LIGANDS ON THE PROTEOLYTIC ACTIVITY OF
FUSARIUM GIBBOSUM CNMN FD 12**

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In order to increase the enzymatic activity of mycelial fungi the influence of some new coordination compounds of Ba, Sr and Ca, including as a ligand dimethyl ester of 2,6-pyridinedicarboxylic acid, on the activity of acid (pH-3.6), neutral (pH-7.4) and alkaline (pH-9.0) proteases synthesized by the mycelial fungal strain *Fusarium gibbosum* CNMN FD 12 was studied. The compounds were added to the nutritive medium in concentrations of 5, 10 and 15 mg/L. The enzymatic activity was determined in dynamics on the 4-6th days of cultivation, period of maximum accumulation of proteases. The effect of the tested compounds on the enzymatic activity varied significantly depending on the composition and concentration of the compound, as well as the duration of cultivation and the type of enzymes. It was found that the compound of barium in concentrations of 5 and 10 mg/L intensified the biosynthesis of enzymes, ensuring on the 4th day of cultivation high levels of alkaline proteases, comparable to the maximum value presented by the control (without coordination compounds) on 5th day of cultivation. Thus, the activity of alkaline proteases on the 4th day of cultivation was by 25.7 and 35.8% higher compared to the control from the same day and by 6.2 and 14.7% - compared to the maximum value of control (5th day). The metalocomplex of Sr showed a pronounced inhibitory effect on the accumulation of acid proteases in all experimental variants and an increase in alkaline protease activity by 44.2-83.7%. Calcium containing compound exerted a moderate positive effect on acid proteases, in all tested concentrations, ensuring the increase of activity by 13.7-15.8%. In the same time, in concentrations of 15 mg/L, it increased the activity of alkaline proteases by 11.6% and extended the period of active synthesis of neutral proteases, the activity significantly exceeding (by 307.3%) the level of the control of the same day and being practically similar with the maximal value of control.

The evaluated compounds exerted a significant distinct influence, from negative to neutral and positive, on the biosynthesis of the enzymatic components of the proteolytic complex synthesized by the micromycete *Fusarium gibbosum* CNMN FD 12 and can be used for obtaining of enzymatic preparations with different, programmed composition, depending on the field of application.

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Keywords: *Fusarium gibbosum*, ligand, coordination compounds, proteases synthesis.

MEANS OF ENHANCING THE ROLE OF BIOLOGICAL NITROGEN IN PHYTOTECHNY

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According to several sources, biological nitrogen plays an important role in the growth, development and productivity of agricultural plants. The share of biological nitrogen in their harvest is about 20%. The rest of the nitrogen comes from mineral fertilizers (30%), organic (8%), seeds (7%) and soil (35%). It is also known that with increasing doses of mineral fertilizers their effectiveness decreases, in agricultural production increases the accumulation of nitrates, intensifies their migration into the environment (water, soil, air, etc.). One of the alternative sources of supplemental plant in nutrition with nitrogen is atmospheric nitrogen fixed microbially. The intensification of the use of molecular nitrogen can be performed in several ways such as: increasing the areas of the crops of leguminous plants, bringing them in the structure of crops up to 20-25%. For this it is necessary to ensure the optimal conditions for the symbiotic fixation process (optimization of aeration and water regime, introduction of the required amount of phosphorus fertilizers and necessary micro-fertilizers).

The use of microbial preparations based on nitrogen-fixing symbiotrophic bacteria, usually significantly increases the symbiotic potential, positively influences the harvest and accumulation of crude protein. The effectiveness of inoculants can be considerably increased by creating "super-effective" *Rhizobium* strains (using the methods of molecular biology and genetics), which would combine all the qualities (activity, ecological plasticity, technology, competitive ability, etc.).

When developing new varieties and hybrids of leguminous plants, it is necessary to assess the "symbiosis" in order to select plant forms that have an increased potential for fixing biological nitrogen. If in the years 1960-1990 soybean varieties and hybrids were created which in symbiosis with the nodule bacteria after its harvest in the soil accumulate from 60 to 90 kg / ha of biological nitrogen, then after the 90s of last century, with the development molecular biology varieties and hybrids with an increased potential for molecular nitrogen accumulation in the atmosphere of 300-500 kg / ha were developed.

In the Republic of Moldova, strains of soybean nodule bacteria with high biological nitrogen fixation capacities were selected, based on which the *Rizolic* biopreparation was obtained, which is currently used for seed treatment before sowing. The research carried out on the lots of the Experimental Base of the ASM, of the Field Crops Research Institute "Selectia" and on the testing field of agricultural crops belonging to the Ministry of Agriculture and Food Industry (s. Bacioi) showed that bacterialization of seeds is an important element in soybean plant cultivation.

Following several years of research, the following conclusion was reached: the use of nodular bacteria of *Rhizobium japonicum* RD2 in soybean culture promotes plant growth, development and productivity while stimulating the nitrogen-fixing activity of the rhizobio-root system.

Keywords: microbial preparation, biological nitrogen, agricultural production.

THE USE OF MICROBIAL BIOPREPARATIONS IN MODERN BIOTECHNOLOGIES

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The creation of new local biopreparations that are the basis of local bacterial, capable of presenwing that are of perspective, than tran their use with imported properties, which are of interest and practical for the country. Mixed microbial biopreparations are created from strains of agronomie microorganisms and their improvement aims to direct the processes that occur in the soil with scientific and practical applications.

Preparations obtained with microorganisms and biologically active substances that differ especially when applied on microorganisms and biologically active substances under conditions of combination with different ecological-physiological bacteria that differ especially in their application on different agro-climatic zones in the composition of preparations with several components can be created from symbiotic associative and rhizosphere microorganisms. Biopreparations create with a complex action, at the same time complex action, at the same time taking into account the properties to decide many problems of biological protection of plants and increase the amount of production (vegetable, fruit, herbs, animal feed), but also improvement.

Most often the bacterial preparations are created to increase the productivity of agricultural plant, that are used microorganisms to the following families, species classes, *Rheziobiaceae*, as well as the genera *Azotobacter*, *Bacillus*, *Pseudomonas*, *Agrobacterium*, *Azospirillum*, Such, non-hazardous organic preparations that are created on the basis of microorganisms isolated from natural objects (agricultural plants).

Concurrent microorganisms are of major importance, as the spermosphere (phyloplane) of plants, contribute to the correlations of plants, which move up the complicated ecological pattern for plants of plants from the necessary microorganisms, harmful and neutral. In the rhizosphere area with bacteria, actinomycetes, fungi, algae, nematodes. In the rhizosphere we find many negative bacteriogram of these bacteria) that protect plants from phytopathogens, which stimulate growth and increase plant productivity. Except with free azotrophs and in associations, species (*Azotobacter*, *Bacillus*, *Klebsiellas*, *Azospirillum*) play an important role in the association and in nitrogen-fixing symbiotic communities, partly in the formation of nodules of berry plants with the combined use of several *Rhizobium* and *Bradrrhizobium* strains. The second, third and fourth components of microbial preparations, which contain bacteria, rhizobacteria, mycorrhizal fungi, as well as biologically active substances have a practical importance have a practical importance in their use in modern biotechnologies.

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Keywords: biopreparat, microorganism, active substance.

BIODETERIATION OF PLASTIC MATERIALS BY PHYTOREMEDIATING MICROORGANISMS

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In the Republic of Moldova and in the EU countries we have a dense pollution of the environment with plastics, including polyethylene, include low density polyethylene (LDPE). Plastic under the action of microorganisms biodegrade into smaller particles, with hit the soil, water and air, reflecting a negative impact on the soil environment plant growth and productivity. The lower the biodegradation of plastic by plants leads to negative consequences on food security and the development of sustainable agriculture. Biodegradation refers more to the impact of microorganisms on the safety of plastic properties, without the chemical transformation of carbon-containing compounds into plastic and biodegradation for these processes must take time. Microbial communities resistant to various adverse conditions can have many unique characteristics. Among a number of properties of soil microorganisms in different climatic zones, with different capacities to decompose plastic, it is worth mentioning more and more. Adaptation to new carbon sources can create new characteristics of microorganisms, especially those that produce active enzymes. Adaptive enzymes to adverse conditions of microorganisms that offer you insights into a new wide range of applied problems, such as non-recyclable plastic pollution. Synthetic plastics present in everyday materials are the main anthropogenic residues, which enter with polluting environment. The reverse, changes in the ecosystem caused by anthropogenic influences such as plastic pollution can have an aromatic impact on a global scale.

Although the issue of plastics remains unresolved, different podules are considered to reduce their impact on the environment. One of them is to use microorganisms capable of biodegrading plastic. Thus, the potential of microorganisms from various unfavorable conditions can be used in outdoor air landfills. Among the prominent microbial agents used for biodegradation, belonging to the following species *Pseudomonas*, *Streptomyces*, *Corynebacterium*, *Asthrobacter*, *Micrococcus*, *Adreia*, *Leifsonia*, *Cryobacterium* and *Flavobacterium*, *Colvella*, *Marimonas* and *Shewwanella*. In laboratory conditions vegetative experiments were carried out with the aim of applying photostimulating microorganisms with non-recyclable polyethylene biodegradation capabilities to the development and growth of *P. sativum* plants. The results were carried out experimentally and can be used for the purpose of stimulating, growing peas and forming the rhizobioradicular system in soil conditions contaminated with LDPE. The strain was applied as a study object. *Rhizobium leguminosarum sp1*. In some variants in the present study process (LDPE) inhibition of growth and development was observed. Phenomenon saturation of the plant during growth and development, being due to the presence of small polyethylene particles leading to sudden major toxicity. Bacteria seeding with *Rizobium.leguminasatum sp 1*. was manifested with respect to the matrix with all the characteristics of the plant increased significantly by 16.8% in the dry mass of the plant, and in the variants with film compared to the control, being 11.8%. The presence in the soil of plastic in the form of polyethylene (LDPE) by way of biodegradation suddenly leads to inhibition of the growth of *P. sativum* plants and the formation of the rhizobio radicular system until their loss.

Acknowledgments: The results were obtained within research project 20.80009.7007.03 supported financially by NARD.

Keywords: pollution, biodegradation capabilities, microbial agents.

STUDY OF THE SYNERGISM BETWEEN MICROBIOLOGICAL AGENTS IN CONTROL OF APPLE SCAR

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The apple during the vegetation is attacked by a wide complex of pathogens that lead to enormous losses of fruit, both in quantity and quality. Among the most dangerous, widespread and common diseases of the apple are: apple scar (*Venturia inaequalis*), powdery mildew (*Podosphaera leucotricha*), brown rot (*Monilinia fructigena*), fire blight (*Erwinia amylovora*), and bacterial canker (*Pseudomonas syringae* pv. *syringae*).

The purpose of this study – evaluation of the synergism between microbiological agents *Pseudomonas fluorescens* AP-33 (trade name Rizoplan) and *Trichoderma lignorum* M-10 (trade name Trichodermin-SC) in control of the apple scar (*Venturia inaequalis* Wint). The experiments were mounted in the field in apple in 8 variants /3 repetitions/3 trees per repetition, according to approved methods. In the variant Standard was used chemical fungicide Jeck –Pot, EC (Difenoconazole+Penconazole 200+100 g/l). The biological efficacy in the control of apple scar in the result of the foliar application have been evaluated as followed: Rizoplan 2,0 l/ha - 72.3%; Trichodermin SC 2,0 l/ha - 76.6%; Rizoplan + Trichodermin SC 2.0 l/ha- 74.5%; Rizoplan 3.0 l/ha - 78.7%; Trichodermin SC 3,0 l / ha - 80.9% and combine application of the Rizoplan + Trichodermin SC 3,0 l / ha - 81.9%. The intensity of the disease development was maximum in the Control variant (4.7%) and minimum in the Standard variant 0.7%. In the variant Standard (Jeck –Pot, EC) with an application rate of 0.4 l/ha the biological efficiency reached 85.1%. The biological efficacy in fruits also depended on the application rate: variant Rizoplan 2.0 l/ha - 77.2%; Trichodermin SC 2,0 l / ha -78.6%; in the variant Rizoplan + Trichodermin SC 2.0 l/ha - 82.8%; Rizoplan 3.0 l/ha - 77.9% and Trichodermin SC 3.0 l/ha -79.3%; in the variant Rizoplan + Trichodermin SC 3,0 l / ha-84.1%, the best efficacy. The biological efficacy against apple scar in the Chemical standard (Jeck –Pot, EC) with an application rate of 0.4 l/ha was 80.7%. In can be concluded that the application rate of 3.0 l/ha of the mixture Rizoplan + Trichodermin SC have demonstrated the highest efficacy in the control of apple scar: 81.9% - on leaves and 84.1% - on fruits.

Acknowledgements: Study was carried out within the State Program project nr. 20.80009.7007.16., funded by the National Agency for Research and Development of Republic of Moldova (ancd.gov.md).

Keywords: apple scar, synergism, Rizoplan, Trichodermin-SC, diseases of the apple.

ANTIOXIDATIVE PROTECTION IN THE BIOTECHNOLOGY OF AGRICULTURAL BREEDING

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Negative external and internal biotechnological and biological factors cause decreased welfare and stress of birds, and their adaptation is due to the integrity of exogenous and endogenous antioxidant mechanisms under stress. For example, temperature, noise, light, transport and chemical effects, consumption of stimulants, food change, technological, biotechnological, experimental and hierarchical factors in the herd. In the conditions of industrial poultry farming, the adaptation of birds is possible through the use of antioxidants and other components of the ration. Therefore, a balanced diet with a set of important nutrients can be considered as one of the main elements for the successful control of stressors in birds. The main factor in combating stress is the antioxidant system of the bird's body. The latter is diverse, responsible for protecting cells from the action of free radicals and includes multiple components - natural fat-soluble antioxidants, water-soluble antioxidants, antioxidant enzymes, the thiolredox system and other. The structural components of protective antioxidants are located in cellular organs, in subcellular or extracellular space. The functionality of these structures of the living cell antioxidant system provides the three main protection mechanisms. First, the formation of free radicals is prevented by removing their precursors with antioxidant enzymes, SOD, glutathione, GSH-Px and metal binding proteins.

The second mechanism - is achieved by the properties of vitamin E, ubiquinol, carotenoids, vitamin A, ascorbic acid, uric acid and other antioxidants. The third mechanism of protection occurs through the functionality of lipases, peptidases or proteases and other enzymes, which systemically eliminate and repair damaged molecules. Therefore, the scientific community proposes the concept of antioxidant protection of cells, which consists in the fact that the protection activated by the received antioxidants is based on preventing the release of free electrons in cellular mitochondria by purifying the original radicals. Therefore, in the final stage, metal ions are bound to their binding proteins and transformed into non-toxic substances. In parallel with exogenous antioxidants, endogenous mechanisms take place in the body of birds, mainly of enzymatic origin of antioxidant protection.

These enzymatic complexes metabolize free radicals in the presence of important micronutrient cofactors, including selenium, iron, manganese, copper and zinc. Simultaneously with maintaining the metabolism in the body of birds during the growing period, antioxidants also influence the oxidation of lipids in meat after slaughter during storage and in eggs, thus helping to maintain the health of products and human health. At the same time, the use of antioxidants remains of scientific importance for further agro-ecological research in the field of agricultural production, which provides for the welfare of the environment during the further development of animal biotechnologies in the agro-industrial complex.

Keywords: adaptation of birds, protective antioxidants, agricultural breeding, biological factors, biotechnology.

**TAXUS BACCATA L.-MORPHO-STRUCTURAL CHARACTERS IN
SPONTANEOUS AND CULTIVATED TAXA**

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Taxus baccata L. is a conifer species with a special economic status. This study aims to present the micro-morphological and histo-anatomical changes of the leaf apparatus in the genus *Taxus* L. taxa, spontaneous and cultivated in Romania.

The biological material analyzed was represented by leaves collected from yew individuals belonging to the spontaneous species *Taxus baccata* L. (Yew Reserve - Tudora Forest, Botoșani County) and from two taxa: *T. baccata* `Robusta` and *T. baccata*, cultivated (S.C. Doropad S.R.L. Suceava). The cross-sections and double coloration (iodine green and ruthenium red) were made according to the methodology of the Vegetal Anatomy Laboratory of the “Al. I. Cuza” University.

All species show the following characteristics: in the vicinity of the stomatal band can be observed a band of regular radially elongated epidermal cells, arranged in 10-11 rows, approximately rectangular, narrow outline, with numerous papillary protuberances, in contrast to the remaining approximately 10-12 bands towards the edge of the leaf blade, with rectangular cells, covered by a fine, smooth cuticle, covering the straight outer walls, devoid of papillary protuberances. With the following difference: in the taxa

T. baccata spontaneous and *T. baccata* `Robusta` this band of papilliform epidermal cells is absent, its characteristics being very similar to those of the midrib.

The histo-anatomical analysis of the leaf revealed the following aspects: the leaf is hypostomatous, with stomata arranged in bands in variable numbers per band, on either side of the vein, and, as a peculiarity, it does not present lignified sclerenchyma hypodermis; the resin canals are absent; the tracheid parenchyma is poorly represented and is mainly seen on the lateral side of the conducting fascicle, in the form of small expansions which exceed the limits imposed by the perfectly superimposed phloem tissue over the woody tissue; the crenulated appearance of the cuticle is because it molds onto the protuberances of the outer walls of the lower epidermal cells (papillary cells).

The micro-morphological and histo-anatomical investigations carried out on the leaves of the studied taxa are in general agreement with the literature for the spontaneous *T. baccata* (Goodman et al., 2001).

Keywords: *Taxus baccata* L., *T. baccata* `Robusta`, micro-morphological analysis, histo-anatomical investigations.

THE ROLE OF *PELOPHYLAX RIDIBUNDUS* (PALLAS, 1771) IN THE FORMATION AND MAINTENANCE OF PARASITIC ZOONOSES

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Amphibians as definitive, intermediate, complimentary and reservoir hosts, for various species of parasites, are "vector" organisms which, being obligatory in the development of parasites, constitute the favorable environment for the penetration, development and conservation of evolutionary forms of parasitic agents.

Amphibians are a category of biological vectors that, moving from the aquatic environment to the terrestrial one, ensure development and multiplication, at least for one biological stage, which gives them the role of source of a pathogen and, at the same time, transmits a variety of parasitic forms. They have a special role in the contamination of areas favorable to certain parasites and participate directly in the formation of parasitic zoonoses.

The role of amphibians in formation and maintenance the zoonosis, it started when it was amplified the direct human contact with animals and has expanded with the introduction of domestication of different vertebrate animals.

According to the helminthological investigations performed on *Pelophylax ridibundus* species, amphibians from Ranidae family, in the center and southern area of the Republic of Moldova, the presence of 19 helminths species was established: *Haematoleechus variegatus* Rudolphi, 1819; *Codonocephalus urniger* Rudolphi, 1819; *Opisthioglyphe ranae* Froelich, 1791; *Paralepoderma brumpti* Buttner, 1951; *Prostotocus confusus* Looss, 1894; *Tylodelphys excavata* Rudolphi, 1803; *Diplodiscus subclavatus* Pallas, 1760; *Parastrigea robusta* Szidat, 1928, *Strigea falconis* Szidat, 1928; *Gorgoderia varsoviensis* Sinitzin, 1905; *Haplometra cylindracea* Zeder, 1800; *Pleurogenoides medians* Olsson, 1876, *Cosmocerca ornata* Dujardin, 1845; *Oswaldocruzia filiformis* Goeze, 1782; *Icosiella neglecta* Diesing, 1851; *Spirocerca lupi* Rudolphi, 1809; *Toxocara canis* Werner, 1782; *T. leonina* Linstow, 1902 and *Acanthocephalus ranae* Schrank, 1788, which from a taxonomic point of view fall into 3 classes, 7 orders, 17 families and 18 genera.

According our helminthological investigations performed on amphibians from the families *Ranidae* and *Bufo* *nidae*, we are established their role as hosts for two species of helminths *Toxocara canis* and *T. leonina* with an impact on the health, in the foreground, of humans and animals (cats, dogs).

Infestation of amphibians with the nematode species *Spirocerca lupi* Rudolphi, 1809 is another proof of their role as vectors, but also their participation in the formation and maintenance of parasitic zoonoses. The species *Spirocerca lupi* causes spirocercosis - one of the parasitic diseases of both domestic and wild vertebrates, it was spread all over the world. This species of nematode forms spirocercosis in carnivores (dog, fox, wolf), and accidentally in goats, horses, cattle, pigs, etc., it is located in the esophagus, clinically characterized by digestive, cardiovascular and general disorders. By detecting the trematode species *Codonocephalus urniger* Rudolphi, 1819 - a trematode with trixene life cycle, the role of amphibians as "biological vectors" is also asserted, because

various species of birds such as: *Botaurus stellaris* Linnaeus, 1758, *Ixobrychus minutus* Linnaeus, 1766, *Ardea purpurea* Linnaeus, 1766, *Egretta garzetta* Linnaeus and other, are the definitive hosts. *Parastrigea robusta* Szidat, 1928, from the Strigeidae family is another species of trematode, that was detected in amphibians in the muscles and less often on the mesentery. The larval form of this species (metacercarie) is also found in fish: *Abramis brama*, *Atherina mochon pontica*, *Alburnus alburnus* and other. The adult forms are parasitic in the intestines of herons and of day predators, especially of the Ardeiformes order - *Ardea cinerea*, *A. purpurea*. The infection of birds with trematode of *Parastrigea robusta* species, this is causes parastrigeosis.

The *Strigea falconis* Szidat, 1928 is a trematode species similar to family Strigeidae that was found in amphibians under the muscular fascias around the neck, chest, legs, under the serosa of the esophagus and goiter, in the connective tissue between the trachea and esophagus, under the skin of the neck, chest and legs. Adult forms parasitize in the bird intestines of different orders: Falconiformes, less often Strigiformes and accidentally Passeriformes (*Oriolus melanocephalus*), Galliformes (*Meleagris gallopavo*), Gharadriiformes (*Charadrius dubius*) and Columbiformes (*Streptopelia chinensis*) causing Strigeosis. The larval forms of meso- and metacercariae are found in birds of the Ardeiformes, Columbiformes, Ralliformes, Steganopodes, Anseriformes, Charadriiformes, Lariformes, Falconiformes, Coraciiformes, Galliformes, Strigiformes, Piciformes, Passeriformes orders, as well in amphibians *R. lessonae* species.

Based on the above, the *Pelophylax ridibundus* species, represent of vertebrates species with an amphibiont lifestyle that directly participates in the formation and maintenance of outbreaks of parasitic agents specific to fish, birds, mammals and humans. Therefore, out of the 19 helminths species detected as a result of laboratory helminthological investigations, all species have faunal importance, bioindicators and contribute to solving the problems regarding the zoogeographic reasoning, of which, 6 species are of medical-veterinary importance, through which is affirm the role of amphibians as vectors of various groups of parasites specific too other groups of animals, both domestic, wild, pets and humans

From the above, in the Republic of Moldova, these results for the first time to demonstrate that the amphibian host may be a vector of parasitic agents and suggests that we should begin to consider the role of amphibians in the dissemination and control of some parasitic agents. This aspect be particularly important in coming years, as warmer temperatures are known to increase feeding of amphibians rates, which means that expected warming from climate change may exacerbate the potential role of amphibians as vectors.

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Keywords: amphibians, biological vectors, parasitic zoonoses, helminthological investigations.

**ASEPTICIZATION OF PLANT MATERIAL OF SOME SPECIES
OF FAM. AMARYLLIDACEAE L.**

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The family *Amaryllidaceae* L. is represented by herbaceous perennials, bulbous or rhizomatous plants, belonging to the Order *Asparagales*, monocotyledons.

Snowdrops are plants that naturally occur in temperate areas of Europe, growing in shady areas, on neutral or slightly acidic soils. In the wild flora, snowdrops grow in deciduous forests, in thickets, in glades and wet and shady grasslands, on plains, on hills and mountains.

In vitro plant propagation by tissue culture is an alternative method of regeneration of rare and endangered species, because in such a case the plant material is usually limited. Sampling small amounts of tissue (explant) from plants does not harm *in situ* populations. In initiating tissue culture of plant species of the Fam. *Amaryllidaceae* L., an important step is the asepticization of the plant material. The techniques differ from one crop to another, and are carried out depending on the characteristics of the plant. Usually, the plants collected from open ground are more infested as compared with those grown in protected environment. The preliminary care of stored plants can reduce the amount of pathogens in explants. The treatment of plants with fungicidal or bactericidal agents is a good option, but it is sometimes not effective when taking bulb tissue under natural conditions.

The research was conducted according to the basic protocol with some optimizations for rare plant species of Fam. *Amaryllidaceae* L. (*Sternbergia colchiciflora* Waldst. & Kit., *Galanthus nivalis* L., *Galanthus plicatus* Bieb., *Leucojum aestivum* L.), concerning the sterilization regime and obtaining the plant material in order to introduce and micropropagate them by tissue culture.

Thus, four sterilizing agents were tested: calcium hypochlorite, ethyl alcohol, mercury chloride and sodium hypochlorite. The best results were obtained after the use of mercury chloride, with a seedling viability rate of 60% in *Galanthus nivalis* L. and *Galanthus plicatus* Bieb., 70% in *Leucojum aestivum* L. and 85% in *Sternbergia colchiciflora* Waldst. & Kit.

The other sterilizing agents proved to be ineffective, since fungal and bacterial infections were detected in most inoculated explants.

Acknowledgments: The research was carried out within the project 20.80009.7007.19 "The introduction and development of technologies for propagation and cultivation of new species of woody plants by conventional techniques and tissue culture".

Keywords: family *Amaryllidaceae* L., asepticization of the plant material, rare plant species, sterilizing agents, pathogens in explants.

CONDITIONALLY PATHOGENIC AGENTS OF THE FAMILY ENTEROBACTERIACEAE CAUSING ACUTE DIARRHEA DISEASES

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Acute diarrheal diseases are caused by microorganisms from the family *Enterobacteriaceae*, which are very numerous and diverse according to their biological characteristics. The family *Enterobacteriaceae* brings together about 30 genera with over 100 species, which inhabit the intestines of humans and animals, being spread with feces everywhere in the environment. According to the World Health Organization (WHO) and UNICEF, there are approximately 2 billion cases of diarrhea each year worldwide, and 1.9 million children under the age of 5 die from BDA, mostly in developing countries.

The studies were conducted during the years 2017-2021. Clinical samples (fecal masses) were collected from patients and investigated in the microbiological laboratory of the MTA Buiucani Public Health Medical Institution of Chisinau. The bacteriological analysis was based on obtaining the pure culture of the pathogen and its subsequent study. A total of 3752 samples were investigated. The identification was made according to the types of colonies. The *Enterobacteriaceae* family forms S, M or R type colonies, developed on complex culture media of general utility (usual, with native proteins) and special (selective, biochemical tests). *Klebsiella* forms large colonies (up to 4-5 mm), mucoid, with a tendency to converge. *Proteus* forms invasive cultures that spread in the form of concentric waves. *Serratia marcescens* produces red pigment. *Escherichia coli* produces burgundy pink colonies, indole positive on UTI Agar chromogenic medium.

Following the investigations carried out within the group of intestinal bacteria, pathogenic and conditionally pathogenic *Enterobacteriaceae* species were identified. Conditionally pathogenic *enterobacteriaceae* are *Klebsiella*, *Proteus*, *Citrobacter*, *Edwardsiella*, *Enterobacter*, *Escherichia*, *Hafnia*, *Morganella*, *Serratia*, *Proteus*, *Providencia* etc. and can cause food poisoning, diarrheal disease, suppurative or septicemic nosocomial (hospital) infections with endotoxin shock. Analyzing the quantitative indices for conditionally pathogenic *Enterobacteriaceae* during the study period, we find that acute diarrheal diseases were more frequently caused by *Klebsiella* (6.0% - 12.9%), *Enterobacter* (1.2% - 5.8%), *Citrobacter* (0.8% - 2.7%), and *E. coli hemolytica* (1.5% - 3.4%). Quantitative indices of conditionally pathogenic representatives of the family *Enterobacteriaceae* varies from year to year, the highest incidence being registered in 2020 in *Klebsiella* and *E. coli hemolytica*, in 2018 in *Enterobacter* and in 2021 in *Citrobacter*. The variation in morbidity caused by conditionally pathogenic *Enterobacteriaceae* is due to living conditions, food and water quality and sanitary regime.

Acknowledgments: The research was conducted within the doctoral project "Pathogens of acute diarrheal diseases - morpho-cultural features, methods of identification, antibiotic resistance and the dynamics of spread in Chisinau city."

Keywords: acute diarrheal diseases, conditionally pathogenic *enterobacteriaceae*, types of colonies, morbidity.

INFLUENCE OF EXTERNAL FACTORS ON THE DEVELOPMENT OF *S. SPINOSA* ON THE LIQUID MEDIUM

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Spinosad is a natural insecticide derived from a species of actinomycete bacterium, *Saccharopolyspora spinosa* (Mertz and Yao 1990), which shows the effectiveness of a synthetic insecticide. It consists of the two most active metabolites, called spinosyn A and D. spinosyns, new macrolides, are natural metabolites produced under aerial fermentation by the actinomycete *Saccharopolyspora spinosa*. These compounds contain a unique system of tetracyclic rings to which two different sugars are attached. These features make spinosad a tool for integrated pest management. The discovery and characterization of *S. spinosa* is a new opportunity a progressive insect management tools in natural products. To obtain these metabolites it is necessary to cultivate in large quantities on liquid media. After the research was initiated, the optimization of the solid nutrient medium was performed in order to maintain the culture, when a uniform development of *S. spinosa* colonies was obtained and a sporulation at 7-10 days, the study of in-depth cultivation on liquid media was studied. Then followed the research of the literature to determine the starting points for the development of the liquid culture medium, several key components were determined. According to literature data, most media had an initial pH of 6.8 when inoculated, the growth was very good and even *S. spinosa* has a good production of spinosad. At the first tests, a very weak growth was determined and the mycelium was not even formed. Likewise, when inoculating samples of cultural liquid on solid media to determine CFU, growth was almost absent and solitary colonies were rarely reported. That is why we went on the path of pH adjustment to weak basic, which led to a rapid and very good growth of *S. spinosa*. After the first negative attempts the cultivation time was also optimized from four days was increased to seven days. Little progress was made in cultivation, but they were insignificant. The cultivation temperature was maintained at 28-30 °C, which allows a good development of the culture on agarized media. After several failed attempts, the idea appeared that maybe the initial amount of inoculum is insufficient and the culture simply fails to develop, in order to verify this hypothesis, the use of the mother culture was used. To obtain this, a 750 ml flask was taken and inoculated with spore suspension, cultured for four days, then the cultural liquid was used as culture. When using this process, a uniform growth was obtained in all repetitions of the same compositions of the environment and also the growth begins very quickly. For an industrial use of *S. spinosa* it is also necessary to study each factor separately but with a higher affinity and to experiment with the composition of the environment to obtain a high productivity and a low price of both the environment and those generated by the process. cultivation.

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Keywords: *Saccharopolyspora spinosa*, active metabolites, pest management, actinomycete, spinosad.

PROTEOMICS ANALYSIS OF NICOTINE METABOLISM REGULATION IN PAENARTHROBACTER NICOTINOVORANS

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Paenarthrobacter nicotinovorans is a soil Gram-positive nicotine-degrading microorganism (NDM) that harbors a 165 kb pAO1 catabolic megaplasmid. The nicotine catabolic genes on pAO1 have been sequenced, but not all the details on the regulation and interplay of this pathway with the general metabolism of the cell are available. Also, little is known on how the cells cope with the accumulation and toxicity of the resulting nicotine metabolic by-products.

Here, we used nanoLC-MS/MS and performed a time-based proteomic study of *P. nicotinovorans* grown in the presence or absence of nicotine. *P. nicotinovorans* cultures were grown in the presence and in the absence of nicotine. Samples were taken every hour for the determination of bacterial growth (OD at 660 nm), accumulation of the characteristic nicotine-blue pigment (OD at 585 nm) and nicotine levels (by HPLC). Cells were harvested at 3 different time intervals: 7-, 10- and 24-hours after inoculation and washed twice for the removal of nicotine-related metabolites. The cells were lysed, separated on 9–16% SDS-PAGE maxi gels, in-gel digested using trypsin, and the resulting peptides mixture was analyzed by nanoLC-MS/MS using a NanoAcquity UPLC (Waters, Milford, MA, USA) coupled to a Q-TOF Xevo G2 MS (Waters). Data analysis was performed using Mascot v.2.5.1 and Scaffold v.4.8.2).

The approach allowed us to identify a total of 915 proteins grouped in 528 non-redundant protein clusters. We found an extensive number of proteins that are both plasmidal- and chromosomal-encoded which allows us to relate the observed differences in protein abundance to the accumulation of known nicotine intermediates and metabolites. A number of five new chromosome encoded enzymes related to the general metabolism of the cell have been found to be regulated by nicotine and provide an anaplerotic pathway that links the Krebs cycle to the nicotine catabolism in this bacterium. Our time-course proteomics experiments also allow us to determine when the Krebs cycle is active and when the nicotine pathway becomes active and when the two of them work together for an efficient energetic metabolism via expression of various proteins through chromosomal-plasmidial gene regulation. The mass spectrometry-based proteomics data has been deposited to PRIDE with the dataset identifier PXD012577.

Data provides insights into cells adaptation to nicotine metabolic intermediates and is useful for future attempts to genetically-engineer the strain.

Keywords: *Paenarthrobacter nicotinovorans*, proteomic study, nicotine metabolic by-products, chromosome encoded enzymes.

ATHERINA (*ATHERINA BOYERI* RISSO, 1810) FROM THE KUCHURGAN RESERVOIR

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The Kuchurgan reservoir is a reservoir subject to high anthropogenic pressure. This is due to the construction and commissioning of a thermal power plant - Moldavskaya GRES. Before the construction of the power plant and the transformation of the natural estuary into a reservoir - the cooler of the MGRES, the same species of fish were found in it as in the reservoirs of the Lower Dniester. On the territory of Moldova, this is the only body of water that is fundamentally different from other aquatic ecosystems in the region, mainly by a moderate degree of thermofication and increased mineralization - about 2500 mg/l.

Atherina appeared in the Kuchurgan reservoir-cooler of the Moldavskaya GRES in the early 1990s. Atherina got into the reservoir, most likely together with the pumped water from the river. Turunchuk. Due to the high potential of reproduction, as well as eurybiontism, sinter in a short time took a dominant position in terms of numbers in the ichthyocenosis of the Kuchurgan reservoir. Due to the high mineralization and increased thermofication of the reservoir, atherina has found a favorable ecological niche for itself here. Due to the fact that the mineralization of the water in the reservoir is lower than that of the sea, as well as competitive trophic relations with native species, the smelt has formed a local herd with slowed down, almost 2 times compared to the sea, growth, earlier puberty and shortened, almost two times, life cycle. According to the results of control catches in 2019-2022. atherine is the absolute dominant in abundance in the ichthyocenosis of the reservoir.

Currently, it is the most massive species of fish in the reservoir. According to the dominance index, it belongs to the D5 category, according to the constancy index - C2 and C3, according to the environmental significance index - to the W5 category. The maximum dimensions and weight of males are 9.4 cm and 3.9 g, and females are 9.8 cm and 4.3 g, which is lower in comparison with the marine form.

In the Kuchurgan Reservoir, the atherina migrates en masse twice to the coastal part of the reservoir - in spring from early March to early April and in autumn from early October to early November. Being heat-loving, in the autumn-winter periods it migrates to the warm channels of the Moldavskaya GRES, forming large accumulations, which is associated with a higher temperature in comparison with the open water area of the reservoir by about 5 degrees or more. Due to the high concentration of atherine in the warm channels in the autumn-winter period, the number of predators increases here, in particular, asp, which actively feeds on atherine. In addition to predators, carp also feeds on smelt, so in early spring, anglers use smelt as bait and catch carps of impressive size (about 10 kg).

Acknowledgments: Research was carried out within the framework of the project №.20.80009.7007.06 AQUABIO

Keywords: atherina, aquatic ecosystems, ichthyocenosis, predators, potential of reproduction, eurybiontism.

**PRESENCE OF THE SPECIES *GLAREOLA PRATINCOLA*
(CHARADRIIFORMES, GLAREOLIDAE) IN THE LOWER PRUT ARIA**

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The wetland of Lower Prut is an attractive sector for an impressive 228 species of wild birds. On May 22, 2022 in the arranged fish ponds in the northern part of Lake Manta (Crihana Veche village, Cahul district) 6 specimens belonging to the species were reported *Glareola Pratincola* (Linnaeus, 1766). Collared Pratincole is a rare species in the avifauna of our country, which prefers arid areas, exposed to the sun, sandy, with little vegetation, near the waters. In Romania it is a summer guest, a nesting species. In the Republic of Moldova, it was encountered during migration, the last report being mentioned by ornithologists on September 9, 2000 near Lake Cuciurgan. Some 19th century authors claimed that in the autumn months huge flocks landed in the lower meadows of the Dniester and Prut. Continuous habitat degradation has significantly reduced the species' numbers.

The first 4 specimens encountered this year were observed on the dry strip in the vicinity of the water, where the grassy carpet was made up of small plants of *Rumex acetosa* and fragments of dried stems of *Oenanthe aquatica*. At our appearance two specimens flew, two more remained on the ground, fluttering, spreading and swelling its wings. Near the birds were 5 specimens of *Charadrius dubius* who were looking for food at the water's edge. Our attention was drawn by the behavior of the specimens of *Recurvirostra avosetta* and *Himantopus himantopus*, who were flying and making shrill sounds in flight, as if wanting to attack. The specimens left on the ground mimicked that they were injured. They defended the colony located in the poor vegetation where nests could be seen consisting of 3-4 eggs. We also met 2 pairs of *Piet avocet* that accompanied their chicks through the small water. At about 50-60 m from the colony, through the grassy carpet about 8 cm high, we came across 2 mature specimens of *Collared Pratincole*. And in this case the birds were agitated, attracting attention by the movements made on the ground or by the sounds made in flight. This kind of behavior is practiced by birds during the breeding season, when adult birds want to take the predator away from the nesting site. Most likely the nests could be hidden in the higher grass zone in the middle of the field. Presence during the summer of the species *Glareola pratincola* and birds behavior involves nesting the species in the Manta Lake ecosystem.

Acknowledgments: The studies were performed within the State Program project 20.80009.7007.02 and doctoral project Ornithofauna of Ramsar wetland "Lower Prut Lakes".

Keywords: *Glareola pratincola*, avifauna, colony of wild birds, nesting the species, rare species.

THE INFLUENCE OF CLASSICAL BIOTECHNOLOGIES ON THE WELFARE OF AGRICULTURAL BIRDS

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Modern biotechnology, representing the interference of biological and technological performance, currently practically occupies the most privileged position in the practice of the poultry sector. At the same time, biotechnology is the use of the concepts of engineering and biological sciences to produce goods from biologically derived materials, for example food in industrial poultry farming. By applying biotechnologies in current poultry farming, real, technologically feasible performances are obtained and the prospects of an increased yield in industrial poultry farming are created, with decisive economic and social effects for the near future. The main advantages of obtaining meat and eggs in biotechnological conditions are their properties to improve animal and human health, maintaining biodiversity and balancing the environment. Poultry biotechnology aims to change the biological properties of the body of birds and increase their productivity. Moreover, current biotechnology evaluates through scientific research according to market development trends, market size perspective, potential opportunities, market share by types of poultry biotechnology and practical production applications. In addition, increasing the health and productivity of birds and increasing research and development in poultry biotechnology are the main drivers of this agricultural sector. Raising awareness of basic and applied research on bird welfare and the implementation of advanced biotechnologies is one of the factors in the future development of the poultry sector. Recent advances in molecular biology, immunology and genetic engineering have multilaterally expanded the scope of study and use of biotechnology in agricultural poultry in industrial growth conditions. Poultry meat and eggs are sources of high quality protein and amino acids, minerals, lipids and fatty acids, vitamins, carbohydrates and other bioactive components. There is also a special specialized field of scientific research on the use of whole products obtained through biotechnologies, especially in poultry feed. The future of biotechnology is to use the principles of increasing the development of birds and their health, increasing reproduction and improving nutritional quality, as well as the safety of poultry products. Currently poultry farming is continuously improved through a variety of biotechnological technologies.

Advances in biotechnology are evolving rapidly, scientific achievements in poultry genome sequencing, technological advances in molecular markers and other applications of biotechnology will provide new research opportunities to improve and modernize the poultry breeding and exploitation industry. In the future, it is expected that the genetic composition of birds will be available, allowing the selection of the most suitable agricultural birds for a given production framework. If the genetic information of a bird population is sufficiently diverse, it is more likely to adapt successfully to environmental changes.

Keywords: modern biotechnology, poultry farming, health and productivity of birds, market development trends.

**REFERINȚE ASUPRA CONDIȚIILOR GEOGRAFICE ȘI A CERINȚELOR
DE PROIECT PENTRU REPARAȚIA PODULUI PESTE RĂUL RĂUT
ÎNTRE LOCALITĂȚILE USTIA ȘI ZOLONCENI DIN RAIONUL
CRIULENI**

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Podurile sunt obiecte din cadrul infrastructurii rutiere, construite prin lucrări tehnice pentru a susține o cale de comunicație și de a asigura continuitatea ei peste multiple obstacole, întâlnite în condițiile geografice ale traseului, obstacole pe care calea de comunicație le traversează modificând și unele elemente ale mediului. Obstacolele pot fi cursuri de apă, văi accidentate sau intersecții cu alte căi de comunicație, cum ar fi căile ferate sau chiar alte trasee rutiere. Prin urmare, podurile asigură continuitatea drumului peste obstacolul traversat, atât pe pod, iar în unele cazuri și sub acesta, datorită spațiului liber amenajat corespunzător cerințelor tehnice și rutiere.

În timpul lucrărilor de reparație a unor podurilor, care fac parte din procesul de reabilitare în zonele inundabile cum sunt văile râurilor, sunt utilizate echipamente grele și o varietate de utilaje, care produc modificări în albia minoră și cea majoră. La fel are loc degradarea vegetației locale, deteriorează structura solului, respectiv influențează negativ asupra ihtiofaunei din râu și a ecosistemelor adiacente. Ploile care cad în timpul acestor lucrări de construcție sau reparație, pot cauza formarea de jgheaburi de eroziune, pot deforma și afecta stabilitatea malurilor din văile râurilor, pot deplasa anumite cantități de materiale în albia râului care vor spori procesul de colmatare etc. Efectele lucrărilor menționate asupra zonelor inundabile sunt considerate semnificative și necesită implementarea unor măsuri de atenuare nemijlocit în timpul de realizare a proiectelor de reabilitare a drumurilor și a podurilor.

Rețeaua de drumuri din Republica Moldova depășește prin obiective de infrastructură precum podurile, multiple obstacole, în special râuri de diferite dimensiuni, iar rețeaua de râuri din bazinul Răutului nu este o excepție. Acest articol reflectă rezultatele cercetării condițiilor geografice în care a fost construit, apoi reabilitat podul peste râul Răut, amplasat între localitățile Ustia și Zolonceni din raionul Criuleni. Mărimea segmentului de râu în conformitate cu sistemului de ierarhizare a rețelelor de râuri după Horton-Strahler, peste care traversează acest pod este de ordinul VII, fiind cel mai mare în acest caz. Prin urmare, la realizarea proiectului s-a ținut prioritar cont de caracteristicile climatice, ale reliefului, cele geotehnice și hidrogeologice. În același timp, reparația podului și a traseului de drum din apropiere a vizat și un șir de măsuri de protecție a mediului, precum: minimizarea impactului asupra reliefului din valea râului, realizarea unor lucrări antierozionale, care să diminueze posibilitatea scurgerii materialului solid în râu, protecția vegetației și semănatul de vegetație ierboasă pentru consolidarea malurilor și a terenului adiacent carosabilului etc.

Keywords: infrastructura rutieră, condițiile geografice ale traseului, rețeaua de râuri, cerințele de proiect pentru reparația podului.

ADVANTAGES OF CRYOCONSERVATION OF SPERM IN REPRODUCTIVE BIOTECHNOLOGY

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Cryopreservation of sperm is one of the most important procedures in the development of biotechnologies for assisted reproduction. In some farm animals, the use of cryopreserved semen has multiple benefits. Cryopreservation of sperm allows long-term storage, dispersal of genes of genetically superior animals from one generation to another and long-distance transport of semen. Another advantage of using cryopreserved semen is that it facilitates the easy sowing of females at the optimal time of reproduction. Moreover, cryopreservation of sperm is an essential biotechnology for the management of sperm banks, thus contributing to the conservation of animal biodiversity and the protection of rare and endangered species. In scientific research, the main attention in this field has been paid to the improvement of biotechnologies and freezing-thawing regimes of semen in animal species of agricultural interest. The scientific interest consists in the use of antioxidants, cryoprotectants in optimal concentrations and minimizing the freezing-thawing time, which are some of the essential requirements in maintaining the structure and function of sperm during cryopreservation. The latter are very vulnerable in the freezing and thawing process, because the reproductive cells are very sensitive to thermal changes and their viability is compromised after thawing. Currently, neither the major achievements of scientific research on the synthesis of synthetic media and freezing protocols, the properties of the researched antioxidants, elucidating how low temperatures cause damage to sperm and a number of other factors do not fully solve the problem of sperm quality. The main damage that occurs during cryopreservation results from the exposure of reproductive cells to temperature variations, which leads to the formation of ice crystals, which structurally damage the inside of the cell and the environment. In parallel, osmotic changes occur. In addition, during freezing and thawing, irreversible lipid-protein changes occur in the plasma membrane as the primary structure of sperm. Thermal variations also change the configuration of phospholipids that increase the rigidity and fragility of the plasmalemma, as well as its permeability, causing a decrease in sperm metabolism. The cryopreservation process also causes chromatin damage that results in DNA fragmentation, the integrity of which varies with protamine, histone, and the intact disulfide bridges between cysteine residues. The effects of molecule damage during cryopreservation are directly reflected on the fertilizing capacity of frozen-thawed sperm. Thus, cryopreservation of sperm is an essential biotechnology of assisted reproduction in virtually all species of mammals, including humans.

Keywords: cryopreservation of sperm, reproductive biotechnology, sperm banks, freezing-thawing regimes, sperm quality.

VIABILITY AND STABILITY OF AQUATIC FUNGI OF BIOTECHNOLOGICAL INTEREST AFTER LYOPHILIZATION

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Microorganisms are inexhaustible and advantageous sources of bioactive substances (antibiotics, vitamins, proteins, food additives, enzymes, organic acids, biopreparations for agricultural use, etc.), for which they are widely used in biotechnology. The sustainable functioning of national collections of microorganisms and their permanent completion is a key necessity for the further expansion of biotechnologies in all industrially developed countries. Fungi are a permanent component of aquatic ecosystems and belong to important microbial communities for organic decomposition, nutrient cycle and energy flows. Of particular interest are the obligatory aquatic species, i.e. those that not only appear and develop actively in water, but, most importantly, cannot reproduce outside the aquatic environment, but are also migratory fungi. Ecological groups found in the seas and freshwater include terrestrial fungi. According to data from various scientific publications, in freshwater environments, the most commonly detected are fungi representing the species: *Aspergillus*, *Penicillium*, *Trichoderma*, *Talaromyces*, *Acremonium*, *Alternaria*, *Fusarium*, *Mucor*, *Rhizopus*. Aquatic fungi produce hydrolytic enzymes that degrade many compounds, thus contributing to the purification of aquatic environments. In addition, they have a high metabolic capacity for carbon sequestration and are believed to be key elements in the carbon cycle and regulators of the global climate. The method of storage of microorganisms of industrial interest is important for any microbiological investigation, and their long-term conservation, without obvious modification of morphological-cultural and biochemical characters is a task of major importance for any collection. The aim of the research was to evaluate the viability and stability of aquatic fungi of biotechnological interest after lyophilization. The object of study was served 20 strains of aquatic fungi, isolated from the lake La izvor, which possesses significant enzymatic and antimicrobial activity. The strains were lyophilized in the defatted milk + 7% glucose protection medium. After lyophilization, the viability, enzymatic activity (catalase), and antifungal activity against phytopathogens were evaluated: *A. niger*, *Alt. Alternata*, *B. cinerea*,

F. solani, *F. oxysporum*. The results obtained in the researches showed that the viability of aquatic fungal strains after lyophilization varies within the limits of 92.8-99%, compared to the initial titer, up to lyophilization. The antifungal activity of the strains, compared to phytopathogenic fungi, after lyophilization was approximately at the level up to lyophilization. Enzymatic activity (catalase) did not change significantly after lyophilization. Also, after lyophilization no changes of morphological and cultural peculiarities were detected. Thus, we can conclude that, after the lyophilization process, the studied fungal strains have kept their high viability and morphological, cultural and biosynthetic stability.

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Keywords: collections of microorganisms, obligatory aquatic species, viability and stability of aquatic fungi, lyophilization, biosynthetic stability.

THE IMPACT OF ZnO AND Cu NANOPARTICLES SUPPLEMENTED IN THE REHYDRATION MEDIUM ON LYOPHILIZED MICROMYCETES

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Rehydration of lyophilized microorganisms is an important step in the recovery of strains from the state of anabiosis. There is a wide range of lyophilized strain regeneration media, which demonstrate various results. It has been shown experimentally that viability increases if the rehydration solution with the protective medium is selected correctly. When recovering cells after lyophilization, the duration and temperature of rehydration are important. Complex rehydration media play an important role in repairing damaged cells by providing additional nutrients and essential cell components. The rehydration process includes three simultaneous processes: absorbing water into the dry material, swelling and restoring the soluble materials in the cell. The extent of rehydration depends on the degree of damage to the cell structure and the chemical changes caused by dehydration.

At present, nanoparticles (NPs) are widely used in biotechnology and microbiology, with the help of which the biosynthetic processes of microorganisms used as producers of bioactive substances can be regulated. NPs, due to their very small size, are able to penetrate the body's microstructure, accumulate on the cell surface or penetrate the cell wall, thus altering the functioning of biological systems. Publications in recent years demonstrate the effect of NPs on the viability, development and biosynthetic processes in microorganisms. With the help of NPs introduced into the culture medium of microorganisms, their morphological features can be modified and biosynthetic processes can be stimulated, thus obtaining the expected microbial product of a higher quantity and quality.

The aim of the research was to study the impact of Cu and ZnO NPs supplemented in the rehydration medium on lyophilized micromycetes.

Twenty micromycete strains, representatives of the genera: *Aspergillus*, *Penicillium*, and *Trichoderma*. The strains were lyophilized in the defatted milk + 7% glucose protection medium. Rehydration of strains was performed with distilled H₂O - control variant and 2 NP variants (NP): H₂O + NP Cu; H₂O + NP ZnO. At the first stage, the optimal concentration of NP in the rehydration medium was selected. 3 concentrations (mg / l) were tested: 0.005; 0.001; 0.01.

The viability and stability of micromycete strains, after lyophilization, rehydrated with media supplemented with NP of Cu or ZnO in a concentration of 0.001 mg / l were evaluated. It was shown that in the variant with NP, the viability of the strains exceeds the control variant by 1-4%, and the antifungal activity of the strains of the genus *Penicillium* and *Trichoderma* significantly exceeds the control. In the variant with NP ZnO there was a decrease by 2-11% compared to the control.

Changes in the morpho-cultural properties of micromycete strains in the NP variants compared to the control variant were not detected.

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Keywords: lyophilized micromycetes, rehydration medium, state of anabiosis, nanoparticles (NPs), culture medium of microorganisms.

DENSITY DYNAMICS OF RODENT SPECIES IN AGROCENOSES IN THE REPUBLIC OF MOLDOVA IN 2021

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Research carried out in the autumn wheat field at the "Horasti" station showed that at the beginning of the second decade of April the relative density was 5%. The dominant species is *A. sylvaticus* (100%). The females are lactating, they reproduce in the third decade of March. In the willow plantation along the dry canal, the density was 2.5%, the dominant species is *A. uralensis* (100%), males being developed. From now on, into the third decade of May, an upward dynamic in relative rodent density has been determined. The values of this parameter are as follows: grassy cover near the acacia grove – 11.5%; fallow bordered by the wheat field – 10.9% and the edge of the wheat field bordered by the willow plantation – 43.4%.

As a result of favourable conditions, the wheat crop developed intensively and the spike was formed. In the fallow at the border with the wheat field the dominant species is *A. sylvaticus* (66.7%), which forms a community with *A. uralensis* (33.3%). The sex ratio for *A. sylvaticus* is 1:1, 50% of individuals being juveniles. This indicates a relatively strong reproductive potential. The highest relative density was recorded in the wheat field ecotone with the willow plantation (43.4%). An 8-fold increase in the relative density of rodent populations was recorded in the wheat field in the spring period concomitant with the upward trend in rainfall. In July, a relative rodent density of 26.7% was recorded in the wheat field ecotone with fallow. This crop is at the final ripening stage, but not yet harvested due to unfavourable weather conditions (heavy rainfall). Wheat dominates in the wheat field ecotone with fallow, with 33.3% of *M. spicilegus* species. Its presence was recorded only in July. It is followed by *A. sylvaticus*, *A. flavicollis* and *A. uralensis* with 16.7% each. The dominance of *M. arvalis* and *A. agrarius* is 8.3%. Following the harvest of the autumn wheat crop at the end of July, the relative density of small rodents decreased compared to the beginning of the month (15.8%).

In September, the following relative density values were recorded: in the forest belt bordered by the plowland – 17.3%, in the forest belt bordered by fallow – 11.8% and in the ecotone of acacia forest bordered by the sunflower field – 21.4%. The decrease in relative density values compared to the previous months is explained by the reduced amount of precipitation during this period, which is reflected by the lower aridity index (25). The small rodent community in the forest fallow biotope consists of three species, the dominant being *A. flavicollis* (50%). *A. agrarius* and *M. arvalis* are dominant with 25% each. At the end of October, the relative density of rodents in the forest strip was 6.7%, the ecotone of the forest strip with fallow and the ecotone of the unharvested maize field bordered by the forest strip along the dry canal – 20% each. In the poplar field the dominance of *M. arvalis* was 66.7%, followed by *A. agrarius*, *A. flavicollis*, *A. sylvaticus* with 11.1% each.

Keywords: rodent species, density dynamics, reproductive potential, relative density, dominant species.

COMPLEX APPLICATION OF *BACILLUS SPP.* AND BIOREGULATORS FOR THE CONTROL OF PESTS

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The losses to the world agriculture, various crops and agricultural products caused by diseases, weeds and pests, account for 35% of the annual production. An increase in yield per hectare can be obtained through the use of scientifically based protection systems. On a global scale, microbial pesticides only account for approximately 1–2% of all pesticides sold; however, they have shown long term growth over the past decade in contrast to chemical pesticides, which have consistently declined in the global market (Bailey et al., 2010). Some sources have recently estimated that the growth in microbial pesticides could reach 3% of the pesticide market in 2014 (Glare et al., 2012). The development and use of bacterial biopesticides as classical, conservation and augmentative biological control agents have included a number of successes and some setbacks in the past 15 years.

The aim of the work was to establish the possibility of entomopathogenic strains *Bacillus thuringiensis* ssp. *kurstaki* (Bt) and *Bacillus thuringiensis* ssp. *thuringiensis* (BT) application in a tank mixture for spraying. For this purpose, the effect of the recommended and half concentrations of para-aminobenzoic acid (PABA) on the above-mentioned bacteria colonies was examined *in vitro*. Bacteria were cultivated in liquid mineral nutrient medium for 48 hours at 29°C to the titer of 10⁹ CFU/ml. The suspension was inoculated on the agarized CGA nutrient media in Petri dishes. After bacterial cultures had grown for 24 hours, sterile disks (five disks per three Petri dishes) soaked in the substance biologic active emulsions were placed on their surfaces. After a week of incubation, the interaction of the studied concentrations of para-aminobenzoic acid (PABA) with bacterial culture was recorded. Bacterial growth inhibition zones were not found. This allows to assume that it is possible to combine working solutions of bioregulators with bacterial strains suspensions and at the same time to reduce the para-aminobenzoic acid (PABA) after effect. The similar results have been reported for *B. thuringiensis* mixtures with the pesticides Sumi-Alpha, Regent, Decis and *Pseudomonas* sp. *Bacillus* sp. with the pesticides Ridomil, Quadris, Raxil and Colfo-Super (Адрианов Ф.Д., 2011; Попов Ю.Б., 2008; Войтка Д.В., 2018).

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Keywords: *Bacillus* spp., bioregulators, bacterial biopesticides, para-aminobenzoic acid.

IMPACT OF NANOPARTICLES IN THE CULTIVATION MEDIUM ON THE VIABILITY AND STABILITY OF MICROMYCETES AFTER LYOPHILIZATION

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One of the factors influencing the viability of micromycetes after lyophilization and prolonged storage is the cultivation medium used before. Cultivation media are substrates that provide the nutrients and physico-chemical conditions necessary for the growth and multiplication of microorganisms. The nutritional needs of microorganisms for optimal growth and development are individual and this must be taken into account when preparing cultivation media. The main factors that ensure the vital activity, growth, and reproduction of microorganisms are nutrition, respiration and living conditions. Currently, nanoparticles (NP) are a means of stimulating the growth and biosynthetic processes of micromycetes of biotechnological interest. Research on the effect of NP on biological objects is necessary and current, because NP are obtained through nanotechnologies from modified materials at the atomic or molecular level. They have unique properties, with a different behavior from conventional materials. The action of NP on microorganisms, according to data from the literature, is different and depends on the microorganism studied, the quantity, size, and duration of the applied NP. NP can stimulate the growth and development of microorganisms, but also decrease or inhibit their growth. The aim of the research: to study the impact of supplemented NP in the culture medium on the viability and stability of micromycetes after lyophilization. In the study were taken 20 strains of micromycetes from the genera *Aspergillus* (5), *Penicillium* (10), *Trichoderma* (5). The strains were grown on Czapek medium - the control variant, and variants in which Czapek medium was supplemented with NP: Cu, Co, ZnO, Fe₂O₃, Fe₂ZnO₄, Fe₂CuO₄, in different concentrations. It was found that the action of NP studied, on the morpho-cultural peculiarities is individual, the best cultures grow and develop on Czapek medium supplemented with NP of Fe₂ZnO₄, Fe₂CuO₄, ZnO, in a concentration of 5 mg / l. NP Fe₂CuO₄, Fe₂ZnO₄, ZnO, supplemented in the micromycete cultivation medium before lyophilization, acted differently on their viability. In some cases, they stimulated, and in other cases they decreased the viability of the strains after lyophilization. Thus, the evaluation of the viability of the strains after 1 year of lyophilized storage, showed that the decrease of the viability of the strains in the variants with NP during storage is slower compared to the control variant. In most cultures, after 1 year of lyophilized storage, the viability in NP variants exceeded the control by 2-10%. It has also been established that NP Fe₂ZnO₄, Fe₂CuO₄, ZnO substituted in the culture medium before lyophilization has a positive effect on the biosynthetic properties of micromycetes. NP Fe₂ZnO₄ and ZnO act more beneficial on the biosynthetic properties, stimulating the antifungal activity against the tested phytopathogens from 2.2% to 21.4%, compared to the control.

Acknowledgments: The research was funded out within the project 20.80009.7007.09 (ANCD).

Keywords: nanoparticles, viability, stability, micromycetes, lyophilization.

THE QUALITY OF FRESH AND ENSEILED BIOMASS FROM NEW CULTIVAR „MARIA” OF *HELIANTHUS TUBEROSUS*

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Topinambur *Helianthus tuberosus* L. belonging to the botanical family Asteraceae, native to the central region of North America, is a crop with great potential for human consumption, generating tubers that store carbohydrates such as inulin and fructooligosaccharides have beneficial effects on nutrition and human health. The aerial part is used as fodder for animals in the summer season and the tubers in the winter season. *Helianthus tuberosus* is food resource for pollinators, since flowering occurs at a time of low food supply for bees and wasps.

The main objective of this research has been to evaluate quality of fresh and ensiled biomass from new cultivar „Maria” of topinambur, *Helianthus tuberosus* which has been cultivated in the experimental field of the National Botanical Garden (Institute), Chișinău, and prospects of its use as fodder for farm animals or as substrates for biomethane production. The results of our research revealed that the dry matter of the harvested whole plants contained 117 g/kg CP, 77 g/kg ash, 329 g/kg CF, 360 g/kg ADF, 586 g/kg NDF, 64 g/kg ADL, 296 g/kg Cel, 226 g/kg HC, 214 g/kg TSS, with nutritive value 60.9% DMD, RFV= 97, 12.03 MJ/kg DE, 9.88 MJ/kg ME and 5.89 MJ/kg NEL. The ensiled mass was characterized by agreeable colour with specific smell and pH 3.85, the silage dry matter contained 116 g/kg CP, 99 g/kg ash, 345 g/kg ADF, 522 g/kg NDF, 46 g/kg ADL, 299 g/kg Cel, 177 g/kg HC, 214 g/kg TSS, with nutritive value 62.0% DMD, RFV= 111, 12.24 MJ/kg DE, 9.91 MJ/kg ME and 6.06 MJ/kg NEL. The green and ensiled mass substrates for anaerobic digestion, have optimal C/N ratio, amount of lignin and hemicellulose, biomethane potential varied from 272 to 329 l/kg ODM.

The green mass and silage obtained from the new cultivar „Maria” of *Helianthus tuberosus* largely meets the standards and can be used as alternative feed for animals and substrate for anaerobic digestion in biogas plants.

Acknowledgments: Research was carried out within the projects of the State Program no. 20.80009.5107.02 “Mobilization of plant genetic resources, plant breeding and use as forage, melliferous and energy crops in bioeconomy” financed by the National Agency for Research and Development.

Keywords: *Helianthus tuberosus*, quality of biomass, cultivar „Maria”, alternative feed, anaerobic digestion.

VALORIZATION OF WINE YEAST SEDIMENTS AS A SOURCE OF LIPID PREPARATIONS

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Currently, various types of medicinal, food and animal feed preparations are developed on the basis of biologically active compounds of microbial origin. It has been demonstrated that yeasts could be used as biotechnological object, due to the high quality of biomass, are an excellent source for valuable biological compounds, such as vitamins, amino acids, unsaturated fatty acids, polysaccharides, antioxidant enzymes and other metabolites.

According to the specialized literature, bioadditives obtained from microbial biomass, due to the safety and effectiveness, significantly increase productivity and fertility of farm animals. Particular attention is paid to the problem of using yeast sediments, rich in valuable biocomponents, which accumulated in significant quantities on wineries and can be used in agricultural and industrial activities.

Determination of the biochemical composition of yeast sediments such as a secondary product derived from the wine production process, is of great practical importance for development of the new preparations based on the biologically active compounds, in particular lipids.

Thus, the purpose of this research was to evaluate the biochemical composition of the yeast biomass obtained from wine waste sediments for obtaining of lipid preparations with the perspective for use in animal husbandry.

The yeast biomass after production of the *Merlot*, *Cabernet* and *Rkatsiteli* wines, offered by the Cricova winery, was used as research object. Lipid content in the biomass was determined gravimetrically by extraction with the mixture of ethanol:chloroform:acetic acid. Determination of the fractional composition of lipids was carried out by the method of thin layer chromatography.

In the study of total lipid content, it was established that the yeast biomass contained from 2.5 ± 0.03 by $11.1 \pm 1.0\%$ d. w. of lipids, the maximal lipids accumulation has been identified in sediments from the *Rkatsiteli* white wine. As a result of the lipid fractionation procedure, it was demonstrated that the yeast sediments is characterized by the content of phospholipids - $17.43 \pm 1.05 \dots 20.61 \pm 0.77\%$, sterols - $13.77 \pm 0.54 \dots 20.01 \pm 0.19\%$, diglycerides - $13.61 \pm 0.13 \dots 14.85 \pm 1.06\%$, triglycerides - $15.96 \pm 1.0 \dots 22.32 \pm 0.84\%$, sterol ethers - $26.35 \pm 0.77 \dots 40.23 \pm 2.39\%$ of the total amount of lipids.

Thus, according to the obtained data, we can conclude that the yeast biomass such as by-products of the wine industry can be used as an excellent source for the production of lipid preparations. The utilization of wastes from the wine industry contributes to the protection of environment.

Acknowledgments: The results were obtained within Project 20.80009.5107.16 New biologically active microbial preparations for increasing the reproductive and productive potential of animals of zootechnical interest, funded by the NARD.

Keywords: lipid preparations, biochemical composition, yeast biomass, wine industry.

CAPACITY OF MICROBIAL MULTIPLICATION DEPENDING ON THE FORM OF PREPARATION OF SOME ONCOPROTECTIVE PLANTS

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The constant increase of oncological morbidity affects the condition of the normal intestinal microbial flora, presenting an aggravating element in the case of chemo- and radiotherapy. The quantitative decrease of bifido and lactobacteria leads to the onset of dysbiosis, which in turn causes various physiological disorders. Globally, there is a growing interest in the use of natural herbal remedies against many diseases of various etiologies. Some native medicinal plants were used in the study, which contain anticancer, antitumor and antiproliferative agents: celandine (*Chelidonium majus*); mistletoe (*Viscum album*); wormwood (*Artemisia absinthium*); thorns (*Xanthium spinosum*) and calamus (*Acorus calamus*). Selective culture medias based on decoction, infusion, alcoholic tincture and cold maceration, prepared from the medicinal plants mentioned above, on the degree of multiplication of some representatives of normal microbial flora were tested. Subsequently, two forms of preparation were selected for each medicinal plant, which proved to be more effective. The data obtained show a different multiplication of bifido- and lactobacteria depending on the pharmaceutical form of preparation: celandine in the form of decoction led to an increase of 5.21 and 9.06%, compared to the tincture; cold macerated mistletoe - by 8.04 and 4.12%, compared to the infusion of this plant; wormwood infusion showed a better performance by 7.20 and 12.35% compared to tincture; thorns in the form of cold maceration - by 2.42 and 1.59%, compared to the infusion; and regarding calamus, no substantial difference was found between the form of cold maceration and infusion (powder preparations), compared to the decoction, for the preparation of which the dried plant was used. Cold maceration of mistletoe and thorns is more effective compared to other forms of preparation, probably due to mucilage, which contains both common plants with other substances – free amino acids, viscous acid, polysaccharides, glycosides, evercetin, minerals, vitamin C, choline, acetylcholine.

Thus, the cold macerated form proved to be the most effective in the case of two medicinal plants – mistletoe and thorn; the form of alcohol-based tincture had less influence on the multiplication of lactobacteria compared to bifidobacteria (wormwood and celandine). The infusion form can be used in subsequent research for wormwood, and calamus – in any form of preparation. When developing microbial phytopreparations with oncoprotective action, it is necessary to consider the potential anticancer, the dose and the optimal pharmaceutical form of preparation of medicinal plants, their impact on the intestinal microbial flora, as well as the risks. Being adjuvant preparations and less toxic than traditional therapies, they can still interact with some drugs (celandine may increase or decrease the effect of some drugs, wormwood may reduce the effectiveness of anticonvulsants, calamus may interact with anticoagulants, sedatives), so care is needed.

Keywords: *Chelidonium majus*, *Viscum album*, *Artemisia absinthium*, *Xanthium spinosum*, *Acorus calamus* adjuvant preparations, oncoprotective plants.

**NEW FAUNISTIC RECORDS OF THE ENDANGERED SPECIES
PROSERPINUS PROSERPINA (LEPIDOPTERA: SPHINGIDAE) FROM
THE REPUBLIC OF MOLDOVA**

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The Hawk-moths (Lepidoptera: Sphingidae) are among the largest and most easily recognized Lepidoptera. Hawk-moths are medium-sized to large moths with strong and robust bodies and relatively short wings. Most species are nocturnal, but some genera (e.g. *Hemaris*) are active during the day. In the fauna of the Republic of Moldova the family Sphingidae is represented by 20 species, about 53% of the European fauna. Four species from this family require protection and conservation: *Acherontia atropos*, *Dolbina elegans*, *Marumba quercus* and *Proserpinus proserpina*. The species *Proserpinus proserpina* are mentioned in the Red Book of the Republic of Moldova (2015) with critically endangered statut (CR). The species is also included in the Bern Convention (Annex II), IUCN Red List of Threatened Animals in Category VU and Nature 2000. *Proserpinus proserpina*, are cited in the Red Book of Ukraine with CR statut, and the Red List of Romania with VU statut.

The species are mentioned for the first time in the fauna of the Republic of Moldova in 1929 by Miller E., Zubovschi N. and Ruscinschi A. in the “*Bulletin of the National Museum of Natural History*”, being so far the only reporting on the territory of the country. The authors mentioned that found a larvae of this species near the village of Bularda (Călărăși district).

Following research conducted in 2012-2021, the species has been reported only in Horăști village (Ialoveni district), on 02.05.22, leg. Galina Bușmachiu.



Fig. The species *Proserpinus proserpina*, Horăști (Ialoveni district), 02.05.22
(Photo: Galina Bușmachiu)

The species develops one generation. Adults take flight in May-June. Butterflies are active at dusk and night, feed on the nectar of flowers attracted by their strong smell, such as *Jasminus* and *Echium*. Larve develop from July to August-September. The caterpillars generally feed on *Epilobium*, but rarely on the eponymous *Oenothera*. Hibernates in pupal stage in the soil. *Proserpinus proserpina* inhabits moist, warm habitats such as wetlands, gravel pits, meadow ditches, stream banks and clearcuts. The limitation factors are the distruction of natural habitats (drainage and intensification).

Acknowledgments: The researches were carried in the project 20.80009.7007.02. from State program of the Institute of Zoology.

Keywords: *Proserpinus proserpina*, endangered species, red book, fauna, conservation.

DYNAMICS OF ACCUMULATION OF EXOPOLYSACCHARIDES IN CULTURAL LIQUID AT THE CULTIVATION OF *SPIRULINA PLATENSIS* SUPPLEMENTED WITH COORDINATIVE COMPOUNDS OF CU (II)

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Spirulina platensis is considered an excellent source of protein, vitamins, lipids, minerals, polysaccharides, nucleic acids, enzymes and pigments. In recent years, many polysaccharides isolated from seaweed have found applications in food, pharmaceuticals and cosmetics. Polysaccharides are a type of biomacromolecules that exist as structural components of the cell wall of seaweed and possess such activities as anti-tumour, immunomodulatory, antimicrobial, antioxidant, anticoagulant, antiviral etc. Usually, the bioactivities of polysaccharides are closely correlated with their chemical properties, such as molecular dimensions, types and ratios of constitutive monosaccharides and the characteristics of glycosidic bonds.

In this context, the aim of the present research was to evaluate the dynamics of accumulation of exopolysaccharides in the cultural liquid of the cyanobacterium *Sp. platensis* at the cultivation on the mineral nutrient *SP-1* supplemented with coordinative compounds of Cu (II), depending some parameters and cultivation conditions (temperature, pH and irradiation regime). Productivity of *Sp. platensis* was determined daily using photocolometric methods. As a stimulator and regulator of EPS synthesis, was selected the coordinative compound $[\text{CuL}(\text{NO}_3)_2]$ in a concentration of 2 mg / l which had a maximum effect on their synthesis, when spirulina was cultured for 7 days. Cultivation was performed for 26 days, during which time the acidic and sulfated exopolysaccharides content was recorded on each cultivation day. Thus, it was attested that there is a gradual accumulation of acidic exopolysaccharides, the maximum accumulation of was detected on the 18th day, reaching the value of 55.08 mg / l, then there is a gradual decrease in their content. In the case of the reference sample, the content of exopolysaccharides accumulated up to day 18 shows lower values than in the case of the compound $\text{CuL}(\text{NO}_3)_2$, but their content continues to increase, reaching a maximum of 63.83 mg/l on 22nd day, in the following days the exopolysaccharide content decreases insignificantly. The dynamics of the accumulation of sulfated exopolysaccharides in the cultural liquid of spirulina shows like in the case of acidic exopolysaccharides, the maximum value on the 18th day being about 46.00 mg/l approximately 85% of total accumulated EPS. After the 18th day of cultivation, the sulfated EPS content gradually decreases, so on the 26th day the values stabilize at 33.91 mg / l. As a result of this research we can deduce the following conclusion that under the action of the coordinative compound $\text{CuL}(\text{NO}_3)_2$ the maximum accumulation of total acidic and sulfated exopolysaccharides is recorded earlier (day 18) than in the case of the reference sample (day 22) and the presence of Cu^{2+} ions, influenced the synthesis and accumulation of acidic and sulfated exopolysaccharides in the cultural liquid of the cyanobacterium *Spirulina platensis*.

Keywords: *Spirulina platensis*, exopolysaccharides, cultural liquid, coordinative compound, dynamics of the accumulation.

METHODS OF CONSERVATION OF MICROALGAE AND CYANOBACTERIA

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Microorganisms are one of the most essential components of the biosphere. The creation and maintenance of a collection of microorganisms plays an extremely important role in the study of microbial diversity, as well as in the conservation of genetically stable producers used in biotechnological production, medicine and environmental biotechnology. Of the 50,000 species of microalgae, it is estimated that about 2.5% are found in culture collections. The development of methods for the conservation of cyanobacteria and microalgae is less intensive than for other groups of microorganisms. Conservation methods can be divided into two groups: periodic cultivation and long-term conservation methods. Periodic cultivation of cyanobacterial and microalgae cultures is used to maintain healthy, physiologically, morphologically and genetically representative specimens. It is attractive to researchers due to its simplicity, the constant availability of crops for work and the ability to control their purity and properties. The needs to maintain the genetic stability of the species, the high costs and the laborious re-sowing of algae have led to the development of methods for their long-term conservation, which have different efficiencies.

The freeze-drying method is a method of storing dry cells, which allows them to be stored for a long time at low temperatures, usually at 4 ° C, without access to oxygen, humidity and light. Lyophilization does not ensure 100% preservation of the viability of microorganism cells and the highest quality of a dry product. It leads to the selection of the most resistant cells in culture, which may not have the desired properties. This method is used successfully to preserve cyanobacteria that reproduce by akinetes (spores), as well as for those species that are able to synthesize exopolysaccharides. Freeze-drying for many algae ensures extremely low levels of crop viability (0-1%) during storage and is not suitable for long-term storage. Therefore, lyophilization is an effective preservation method only for some algae strains. Suspension of cells with protective agents before lyophilization (skim milk, sucrose) increases the number of surviving cultures. Cryopreservation methods are performed by direct immersion of ampoules with microorganisms and cryoprotective solution in liquid nitrogen at -196 °C. Cryoprotectants increase the viability of cells at cryogenic temperatures. The most commonly used cryoprotective compounds are dimethylsulfoxide (DMSO), glycerol and methanol.

Thus, the issue of long-term conservation of microalgae is relevant, current and attractive to researchers and is one of the basic objectives of the national collection of non-pathogenic microorganisms that constantly seeks to improve conservation methods for long-term storage of microalgae and other microbial resources valuable.

Acknowledgments: The research was funded out within the project 20.80009.7007.09 (ANCD).

Keywords: conservation, microalgae, cyanobacteria, national collection, long-term storage, microbial resources.

ACTINOBACTERIA AS BIOCONTROL AGENTS FOR COMBATING PEST INSECTS

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In the pursuit of profit and food security, in the condition of climate change, world agriculture, applying excessive various synthetic inputs, provoke many negative phenomena, which seriously affect human health and the state of the environment. Since the application of pesticide against pests and different hazardous to environment and human health, many efforts have been oriented in order to elaboration and application of ecologic friendly means.

To ensure the sustainable development of agriculture, many novel products have been discovered from the research and development on biological plant protection, which are superior in safety for humans and environmental. Among the microbiological means of plant protection, a special place belongs to actinobacteria, which are prevalent in soil, water, crops and environment. Thus, actinobacteria became as a good alternative for the management of crop pests. These, as well as the metabolites found in actinobacteria, serve to develop a significant range of effective and harmless preparations.

The avermectins, represents a group of macrocyclic lactones natural means homologues produced by the soil *Streptomyces avermitilis* and act as a complex of eight closely related avermectins, which demonstrate the highly pathogenic to many arthropods. Abamectin is considered as a selective pesticide, which has several advantageous traits include safe to humans and environment and low pathogeny to nontarget pests. Emamectin benzoate is a synthetic version of abamectin having broader insecticidal activity than abamectin. Milbemectin is an insecticidal and acaricidal product isolated from the fermentation broth of *Streptomyces hygroscopicus*, which are the secondary metabolites of actinobacteria. The polynactins are very effective against mites under high moisture conditions and have been utilized for the management of thus pests.

The spinosyns are a distinctive family of fermentation-derived insecticides, from *Saccharopolyspora spinosa* having potent activity against a large spectrum of insects and have lower environmental impact.

Our research is aimed at determining the morphological and cultural characteristics, the possibility of growing on various culture media, as well as the activity in the control of harmful insects, which cannot be effectively combated with other environmentally harmless means of protection.

Acknowledgments: The work was carried out with the support of the State Program project (2020-2023) „The synergism between natural factors and microbiological, environmentally friendly, means of regulating of the population’s density of harmful organisms for the crops protection in conventional and organic farming” no. 20.80009.7007.16.

Keywords: actinobacteria, biocontrol agents, pest insects, biological plant protection, avermectins.

**HELMINTOFAUNA IN PHEASANT (*PHASIANUS COLCHICUS L.*)
MAINTAINED IN CAPTIVITY IN MOLDOVA**

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The common pheasant (*Phasianus colchicus L.*) is the most important bird for the avian fauna of the Republic of Moldova, both in terms of numbers and distribution, as well as hunting perspectives.

The efficient and continuous exploitation of species for hunting purposes requires the most detailed knowledge of their way of life, the correlations between the populations of this species, as well as their level of parasite infestation (Zamornea M. et al. 2017).

The investigations regarding the determination of the pheasant parasite species have been performed in the Parasitology and Helminthology Laboratory of the Institute of Zoology. Biological, samples were collected from the Central area of the Republic of Moldova in the period 2020-2022.

The following methods were used: the coproovoscopic methods (Fulleborn, Darling), the coprolarvoscopic methods (Popov, Baermann), partial parasitological investigations (after K. I. Skriabin) and successive washing were used. The collected material was further examined using the MBC-9 magnifier (ob.14x2) and the Novex Holland B ob microscope. 20-40 WF 10x Din / 20mm.

In pheasants from the 127 samples collected, 14 parasite species were recorded: *Capillaria annulata* (Molin, 1858), *Syngamus tracheia* (Montagu, 1811), *Heterakis isolonche* (Linstow, 1906), *Ascaridia galli* (Schränk, 1788), *Heterakis gallinarum* (Schränk, 1788), *Trichostrongylus tenuis* (Mehlis, 1846), *Capillaria caudinflata* (Zeder, 1800), *Eimeria colchici* (Norton, 1967), *Eimeria duodenalis* (Norton, 1967), *Eimeria phasiani* (Tyzzer, 1929), *Choanotaenia infundibulum* (Bloch, 1779), *Raillietina tetragona* (Molin, 1858), *Prosthogonimus ovatus* (Rud., 1803), *Raillietina echinobotrida* (Megnin, 1880), which were distributed in the 4 classes (Trematoda, Cestoda, Nematoda, Conoidasida), 7 families (*Prosthogonimidae*, *Davaineidae*, *Capillariidae*, *Syngamidae*, *Heterakidae*, *Trichostrongylidae*, *Emeriidae*) and 8 genera (*Prosthogonimus*, *Raillietina*, *Capillaria*, *Syngamus*, *Heterakis*, *Ascaridia*, *Trichostrongylus*, *Eimeria*).

Acknowledgments: The research was carried out with the support of the institutional project - state program: *Diversity of hematophagous arthropods, zoo- and phyto-helminths, their vulnerability and tolerance strategies to climatic factors and elaboration of innovative procedures for integrated control of species with socio-economic value*: 20.80009.7007.12 F, 2020-2022.

Keywords: *Phasianus colchicus L.*, helminthofauna, avian fauna, parasite infestation.

**PRODUCTIVITY, CAROTENOID AND GLYCEROL CONTENT OF
DUNALIELLA SALINA CULTIVATED IN THE PRESENCE OF GeO_2
WITH VARYING LIGHTING REGIME**

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Due to the increased content of bioactive substances, especially carotenoids and glycerol, microalga *Dunaliella salina* is of interest and practical advantage in industrial cultivation, being a microalga of biotechnological interest. And due to the metabolisation by *D. salina* of microelements from the structure of chemical compounds, microalgal biomass acquires high nutritional values and new qualitative properties (immunostimulatory, regenerative, cytoprotective etc.), a valuable fact in obtaining ecological multifunctional products with unique properties.

The trends of modern biotechnology require obtaining products in a closed cycle by reusing industrial waste. Green microalga *D. salina* is also an attractive object for the cultivation on the liquid waste especially obtained from the production of the biomass of other microalgae. Thus, the aim of this research was to evaluate the productivity, the content of carotenoids and glycerol in the biomass of *D. salina* cultivated on organo-mineral medium, elaborated from the cultural liquid, resulting from the cultivation of *Spirulina platensis* (production waste), in the presence of GeO_2 compound with the modification of lighting. It was established that light intensity is an important factor in the accumulation of *D. salina* biomass, cultivated both in the absence and in the presence of the GeO_2 compound. The productivity of the control sample grown under intense illumination was 54.2% higher than the control sample grown under regular illumination. The addition of the GeO_2 inorganic compound showed an increase in the *D. salina* productivity by 12.4% at the concentration of 10 mg/L compound and regular lighting, and by 11.2% at the concentration of 20 mg/L, but intense lighting, compared to corresponding controlsamples.

In the samples grown under regular illumination (3500 lx), the most representative stimulating effect on the synthesis of carotenoids was observed for 5 mg/L GeO_2 compound. The content of carotenoids increased with 22.95%. The further increase of the GeO_2 concentration led to the decrease of the carotenoid content. In the samples grown under intense illumination (5000 lx), the highest content of carotenoids were obtained at the addition of the maximal concentration of GeO_2 (20 mg/L). The increase of the carotenoid content was 27%.

The amount of glycerol increased with the increase of the GeO_2 concentration in the samples cultivated under intense illumination (5000 lx) and showed the highest experimental values at the maximal added concentration of 20 mg/L. The increase of glycerol was 33.2%, compared to the control sample. At the regular illumination (3500 lx), only the addition of the minimal concentration of the GeO_2 compound (5 mg/L) stimulated the synthesis of the glycerol. The increase of glycerol content was 24.2%, compared to the control sample. As the concentration of GeO_2 increased in the culture medium, the accumulation of glycerol in the *D. salina* biomass decreased.

Keywords: *Dunaliella salina*, productivity, content of carotenoids, glycerol, biotechnology.

ANTIMICROBIAL ACTIVITY OF POLYSACCHARIDE-CONTAINING SPIRULINA EXTRACTS

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In recent years, the research on cyanobacteria and algae has demonstrated that many extracts obtained from their biomass have been shown to have antibacterials, antivirals, antioxidants, antitumoral effects.

Cyanobacterium *Spirulina platensis* is widely used as food bioadditives due to its valuable biochemical composition: high protein content (60–70%), including phycobiliproteins, acid polysaccharides, polyunsaturated fatty acids, β -carotene, chlorophyll a, vitamins, minerals, and the presence of secondary metabolites such as polyphenols, sterols, et.al

For the qualitative screening of the antimicrobial activity of the investigated polysaccharides extracts, the well method was used, standardized for the control of the antimicrobial activity proposed by the CLSI standard.

The extract with polysaccharides content obtained from the biomass of spirulina cultivated in the presence of the zinc acetate showed bactericidal action on the strains of *Staphylococcus aureus* ATCC25923 and *Bacillus cereus* ATCC 11778 in 1: 1 dilution and inhibitory action in 1: 2 dillution.

The extract with polysaccharide content obtained from standard biomass showed bactericidal action in 1:1 dilution and inhibitory action in dilution of 1:2 on *S. aureus* ATCC25923 strains and only inhibitory action in 1: 2 dillution on *B. cereus* ATCC 11778 strains.

In the future, the action of polysaccharide-containing spirulina extracts on some gram-negative bacteria will be researched, as well as their antifungal action.

Keywords: *Spirulina platensis*, polysaccharides, bactericidal action, antimicrobial activity.

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Session C

MOLECULAR BIOLOGY AND BIOMEDICINE

NUTRIGENETICS-THE FUTURE OF PERSONALIZED NUTRITION

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Obesity is a multifactorial disease, with genetic polymorphisms accounting for around 70% of the population variation. Interactions between genes and nutrients can greatly influence an individual's response to diet, making it necessary for a more personalized approach in nutrition to be implemented. Associations between single nucleotide polymorphisms (SNPs) and nutrients have been analyzed. Carriers of FADS, APOE4, ALOX5, FFAR4 gene polymorphisms have shown different patterns of lipid metabolism, each responding differently to omega-3 fatty acid supplementation.

MTHFR SNPs carriers have also shown a varying response to folate intake, which helps regulate homocysteine levels that have been associated with a higher risk of cardiovascular disease. SNPs located in the MC4R, FTO, PPAR genes have been found to influence the adherence to the Mediterranean diet. A personalized intake of macro- and micronutrients that takes into account a person's genetic background can improve the lipid profile as well as reduce inflammation, which in turn helps prevent cardiovascular disease in patients with obesity. Further research is needed to implement these findings into clinical practice.

Keywords: nutrigenetics, personalized nutrition, obesity, multifactorial disease, lipid metabolism, person's genetic background.

THE INFLUENCE OF NUTRIENTS ON THE METABOLISM DEPENDING ON THE TYPE OF STRESS REACTIVITY

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The experiment was performed on white rats Vistar line selected based on the reactive type of the organism (analogous to the asthenic type). For the selective choice of experimental animals with general non-specific activity of the body and with the level of stress-reactivity, the methods of "forced swimming" and "high cross-shaped labyrinth" were used. From the animals with the reactive type of the organism were formed according to the analogous principle (body mass, age, sex) 4 experimental groups of animals, of 4-5 individuals in each, which were maintained in the same conditions with an analogous diet. The difference was found in the fact that each group of animals will receive their food ration which will differ according to the caloric structure. The caloric structure of the food rations is represented in table 1. The duration of the experiment was 2 months.

Table 1. Caloric structure of food rations according to experimental groups (%)

	I group (control)	II group	III group	IV group
Proteins	8	11	12	14
Fats	35	28	27	25
Carbohydrates	57	61	61	61

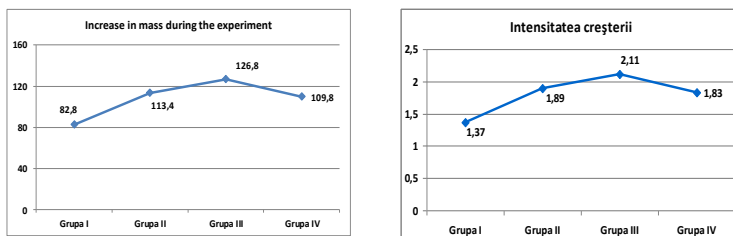


Figure 1. Changing body mass depending on the caloric structure of the ration (g/animal)

Thus, according to the results obtained and presented in figure 1, allows us to make a preliminary conclusion that the best variant of the ration with caloric structure for the asthenic type of constitution is the structure of the 3rd variant: proteins - 12%, fats - 27%, carbohydrates - 61%. The biological value of food rations must be determined according to the formula of the caloric structure of the ration that reflects its balance, the type of exchange of substances of the individual and the fermentative systems that underlie it. The formula of the caloric structure of rations is one of the most appropriate indicators. It allows to identify the optimal ratio of proteins, fats and carbohydrates - the basic energy carriers that define the nature and quality of metabolism. The health of the individual, but also possible dysfunctions and pathology depends on the ratio of these ingredients in the ration.

Keywords: stress reactivity, nutrients, metabolism, ration, caloric structure, health.

IMPACT OF THE PHYTOPREPARATUS APUSET-6 ON THE ENDOCRINE PANCREAS-THYROID AXIS

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Thyroid hormones influence multiple metabolic processes through changes in the concentration and activity of many enzymes, the metabolism of substrates, vitamins and minerals, the rate of secretion and inactivation of other hormones and the response of target organs. Thus, the prophylaxis of the thyroid gland prevents a series of severe autoimmune diseases. In addition to drug treatment, naturist medicine is becoming more and more widespread, in order to maintain the positive effects after treatment or as a prophylactic method at an early stage. Scientific research confirms positive actions in the treatment of the endocrine-thyroid pancreatic axis. Thus, new horizons are opened in the practice of therapy and treatment of endocrine pathological conditions.

Purpose was to highlight changes in physiological indices in endocrine-thyroid pancreatic disorders in white laboratory rats against the background of APUSET-6 biopreparation.

Research object - the white laboratory rat. The model of the experimental hypothyroidism - by administering the solution of potassium thiocyanate, in a ratio of 20 mg per 100 g of body mass. The biopreparation consists of the following plants: *Anethum graveolens*, *Plantago major*, *Urtica dioica*, *Salvia officinalis*, *Equisetum arvense*, *Thymus serpyllum*.

This study highlighted the pathological effect of hypothyroidism on the body, which is manifested by disorders in all organ systems, and in late states - coma and death. Research has shown that interdependence between hypothyroidism and diabetes mellitus results in the determination of insulin hyposecretion (2.07 ± 0.17 pmol/l after administration of K⁺ thiocyanate compared to the initial concentration of 2.58 ± 0.21 pmol/l), a fact that conditions the increase in blood glucose levels (6.4 ± 0.34 mmol/l in pathology and 4.0 ± 0.22 mmol/l in the control group). The analysis of the leukocyte formula showed an increase in all indices against the background of hypothyroidism in the white laboratory rats. These changes indicate that changes in cellular immunity are closely related to the pathological conditions of the thyroid gland.

Experimentally, it has been shown that the administration of APUSET-6 biopreparation has a high efficacy in the prophylaxis of thyroid dysfunction, thus protecting the affected body from hypothyroidism, diabetes and acute infections, by maintaining hematological indices within the norm.

Keywords: endocrine pancreas-thyroid axis, Apuset-6 biopreparation, physiological indices, pancreatic disorders, hypothyroidism, diabetes mellitus.

**PROPHYLAXIS OF RESPIRATORY SYNDROMES BY
NEUROIMMUNOMODULATORY ACTION OF NATURAL
ENVIRONMENTAL FACTORS**

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Review demonstrated particular interest on neuromodulation of inflammation, based on immunomodulatory action of parasympathetic nervous system and an indirect inhibition on the cytokine storm triggered by acute respiratory distress syndrome (ARDS). ARDS can be induced by direct lung injury due to bacterial or viral infection, inhalation of smoke, toxic chemicals, or aspiration of gastric contents, or indirect lung injury. Vagal neuromodulation may be a promising alternative immunomodulatory prophylaxis for respiratory syndromes due to its potent systemic anti-inflammatory effects. As a result, the release of pro-inflammatory cytokines is weakened, coagulation is modulated, and circulatory failure is prevented. The cholinergic anti-inflammatory pathway (CAIP) is a potent anti-inflammatory indirect mechanism of action on the spleen. Vagus immunomodulation is realized through the $\alpha 7$ nAChR receptor, which is found not only on neurons and macrophages, but also on other non-neuronal cells, including immune cells such as monocytes, T- and B-lymphocytes, and dendritic cells. The impact on the body of natural environmental factors is favorable due to its neuromodulatory influence, which produces an immunomodulatory effect of the parasympathetic nervous system on adaptive immunity. Urbanization and rising living standards are associated with unhealthy diet, lack of physical activity and air pollution, hypoxia, hypercapnia, noise, leading to an imbalance in the sympathetic and parasympathetic components of the autonomic nervous system.

The organization of green recreational areas, environmentally friendly urban planning, transport traffic, the exclusion of desynchronization and central fatigue during occupational activities, somatosensory stimulation are the basic components of a complex neuroimmunomodulatory preventive effect.

Keywords: acute respiratory distress syndrome, autonomic nervous system, neuromodulatory influence, alternative immunomodulatory prophylaxis.

DIFFERENTIAL DIAGNOSIS OF CDG THROUGH MOLECULAR ANALYSIS OF GALT AND ALDOB GENES

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Errors in the synthesis, assembly and/or processing of glycans provoke a group of metabolic genetic pathologies known as Congenital Disorders of Glycosylations (CDG) that have been described at the moment around 150 types. Galactosemia and fructosemia can cause false-positive results in diagnosis of CDG due to mutation in *GALT* and *ALDOB* genes leading to changes in carbohydrate metabolism.

The aim of this study was to establish and implement the methods for molecular diagnosis of galactosemia and fructosemia necessary for differential diagnosis of CDG.

As a material for the study have been used samples of urine, DBS and DNA collected from 8 patients. Biochemical diagnosis was based on assessing of blood Galactose level, *GALT* enzyme activity and NMR spectroscopy of urine (Galactose, Galactitol and Fructose-1- phosphate). For molecular genetic diagnosis have been used PCR-RFLP and Sanger sequencing methods necessary for identification of genetic variants in *GALT* and *ALDOB* genes.

The design of primers characteristic to all exons of *GALT* and *ALDOB* genes was created with Primer3 software and checked by NCBI Primer-BLAST. Implementation of the molecular-genetic methods have been done on DNA samples of 8 patients. 7 children was suspected for galactosemia, having liver affection and sepsis (*E. coli*) after breastfeeding during the first week of life, null or reduced *GALT* activity and high urine Galactose [28.7-50.7 mol/molCrea] and Galactitol [~10 mol/molCrea]. The last one had clinical features characteristic for fructosemia, but have normal level of fructose-1-phosphate in urine, caused by his refusal to eat fruits. During molecular genetic analysis in patients with galactosemia has been identified *p. Q188R* mutation in 50% of alleles, followed by *p.E203L* (25%) and *p.K285N* (17%). It is noteworthy that 8% of alleles have not been identified yet. In the patient with fructoseemia has been detected a pathological deletion (c.113-1_115del) of 4 nucleotides located in intron-exon junction between intron 2 and exon 3. This mutation could cause skipping of exon sequence during splicing of *ALDOB* gene, leading to a null activity of the residual enzyme.

The obtained results confirmed the efficiency of implemented of molecular-genetic methods in the diagnosis of galactosemia and fructosemia, that can be used successfully in the differential diagnosis of CDG.

Keywords: *GALT* and *ALDOB* genes, differential diagnosis of CDG, galactosemia, fructosemia, mutation.

**BAICALEIN PREVENT SCOPOLAMINE-INDUCED MEMORY
IMPAIRMENT IN ZEBRA FISH BY INCREASING THE CREB PROTEIN
LEVEL AND THE mRNA EXPRESSION OF *BDNF*
AND *CREB* GENE**

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Baicalein (Baic), is a natural flavonoid, which was isolated for the first time from the roots of the plant *Scutellaria baicalensis*, variety, *Georgi*. Numerous studies have shown that Baic has many pharmacological properties and a variety of important biological activities such as its anti-inflammatory, anticancer and neuroprotective activity. However, its effects on the neuropathology of Alzheimer's disease (AD) have not been well studied.

Brain-derived neurotrophic factor (BDNF) is a neurotrophin with vital functions in the survival of neuronal cells. The cAMP response element binding protein (CREB) is an essential neuroprotein for long-term changes in synaptic plasticity that mediate the conversion of short-term memory to long-term memory.

This study aimed to investigate the effects of Baic pretreatment on cognitive and cholinergic deficiency, oxidative and molecular status, and neural protection against neuroinflammation in the animal model of dementia represented by zebrafish (*Danio rerio*) which was induced by immersion administration of a muscarinic cholinergic receptor blocker represented by scopolamine (Sco 100 µM).

Baic (1, 3 and 5 µg / L) was administered by immersion to zebrafish once a day for 16 days. To assess the level of anxiety of zebrafish we used the new aquarium diving test (NTT), to assess the spatial memory we used the Y maze test, and to assess the recognition memory we used the new object recognition test (NOR). To evaluate the effects of Baic on the oxidative and cholinergic status in the zebrafishes brain, we evaluated the specific activity of Superoxide dismutase (SOD), Catalase (CAT), Malondialdehyde (MDA) and Acetylcholinesterase (AChE) activity. Also, in this study we investigated the effects of Baic on *bdnf* and *creb* mRNA expression.

Our results show that Baic can effectively restore the antioxidant defense mechanism by increasing the level of antioxidant activity in the brain. Baic also can improve cognitive dysfunction of amnesic fish by increasing the absolute gene expression of *bdnf* and *creb* and by inhibiting AChE, which is also correlated with improved memory parameters, as shown in behavioral approaches (NTT, Y maze and NOR).

The authors state that they have no conflicts of interest.

Keywords: memory impairment, Baicalein, *Danio rerio*, cognitive and cholinergic deficiency, neuroinflammation, dementia.

THE LINK BETWEEN CLINICAL MANIFESTATIONS OF SMA AND UNBALANCED GENOMIC CHANGES

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Spinal muscular atrophy (SMA) is an autosomal recessively inherited progressive neuromuscular disease caused in 95% of deletions in the SMN1 gene. SMA can also be suspected along with other clinical manifestations such as mental retardation, global developmental delay, epilepsy, autism, neurological syndromes and birth defects.

We report 2 cases of 2 month and respectively 16 month boys born at term in a no-consanguineous families. Both children presented with severe hypotonia with lack of reflexes and dynamics, suggesting SMA and other clinical manifestations. The molecular-genetic examination of SMN1 gene through PCR-RFLP and qPCR methods showed deletions of exons 7 and 8 for one of them but not for the other patient. At the same time, considering other clinical manifestations associated with chromosomal genetic abnormalities, the constitutional karyotype with subsequent molecular karyotype investigation was indicated. Subsequent the result of the constitutional karyotype was normal (46 XY) for both. Molecular karyotype, through Array-CGH method, identified the following unbalanced haploinsufficient genetic changes: a 1398 Kb microdeletion in the region of chromosome 5q13.2 (of which OMIM registered genes such as SMN1, NAIP, GTF2H2, SERF associated with SMA) and a microdeletion of 4832 Kb in the region of chromosome 10q11.22-q11.23 (of which 6 morbid genes, registered OMIM) for patient without SMA diagnosed and for patient of 2 mo, with SMA diagnosed, there have been identified unbalanced genomic abnormalities (461kb duplication on chr1q and 1.78Mb deletion on chr5q, among them are the SMN1 and SMN2 genes) and regions with LOH (3.6Mb region on chr1p and 4.8Mb region on chr14q including TMEM260 gene).

In this cases, a detailed phenotypic and genotypic approach revealed that the diagnosis or suspicion of SMA was mimicked by much more complex genomic abnormalities such as microdeletion and microduplication of chromosomal sectors that are too small to be detected by conventional cytogenetic methods. Molecular karyotype is extremely important in clinical utility for patients with such genomic changes, both in diagnosis and in long-term management and the determination of the risk of recurrence. Its scientific utility is also significant, with new microdeletion or microduplication syndromes being recognized and clinically delineated.

Keywords: spinal muscular atrophy, deletions of exons, chromosomal genetic abnormalities, molecular karyotype, diagnosis, long-term management, determination of the recurrence risk.

DEVELOPMENT OF METHOD FOR SHORT REPEATS EXPANSION CAUSED ATAXIAS DIAGNOSTIC

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There are known many hereditary ataxias caused by short repeats expansions, most prominent of them being Huntington disease, Friedreich ataxia and a number of spinocerebellar ataxias (SCA`s).

These ataxias are very similar with each other being sometimes almost phenocopies, so a molecular-genetic diagnostic method is strictly necessary for differential diagnostics. There is also a well-known effect called genetic anticipation, when some alleles, usually ones with a quite large, but still normal number of repeats may further expand during gametogenesis or first zygote divisions. It puts potential their children of people harboring these alleles at risk of disease development. That`s why a screening of affected child`s parents and their siblings is necessary.

A number of previously-published papers shows that number of repeats is most frequently directly correlated with age of disease onset and speed of its progression. So, counting repeats number may give not only diagnostic, but prognostic information for affected persons.

In this work we developed a diagnostic approach based on a combination of agarose gel AFLP and capillary electrophoresis-based fragment analysis to detect short repeats expansion and calculate total repeats number for a number of diseases, including different forms of SCA, Huntington disease and Friedreich ataxia. As a test group a selection of patient`s DNA with presupposed diagnosis of SCA or Friedreich ataxia stored in LGMU biobank was used. As a control group were used DNA of presumptive healthy individuals from the same biobank.

Keywords: SCA, repeats, Friedreich, Huntington, ataxia.

EVALUATION OF POLYMORPHISM INFORMATION OF GENETIC DIVERSITY IN BROOMRAPE FROM BULGARIA

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In this study, genomic polymorphism in 4 broomrape populations from different regions of Bulgaria was investigated through amplification by polymerase chain reaction of intersimple sequence repeat (ISSR) regions.

Out of 137 amplicons produced by 13 ISSR primers, 118 were found polymorphic (86.13%). The average was 10.54 fragments per primer. Moreover, ISSRs yielded 5 common and 14 specific bands. The fragment sizes ranged from 0.23 to 4 kb. This study demonstrated that 3'-anchored primers based on dinucleotide repeats with GA and AC motifs have higher polymorphism (96.43 and 100%, respectively) than primers with other di-, tri- or tetranucleotide repeats. In general, the highest number of polymorphic DNA fragments and high percentage of polymorphism were produced using primers (AG)₈YA, BC841, BC857, BC810, BC807, (CTC)₄RC and (CA)₆RG, thus demonstrating the potential of the primer system. The performance of ISSR markers used was evaluated by means of different parameters such as polymorphic information content (PIC), effective multiplex ratio (EMR), marker index (MI), Simpson's coefficient (h_i) and resolving power (RP). The PIC values for all primers varied from 0.16 to 0.35 with an average of 0.26. The highest PIC value was detected for primers (GACA)₄, BC857, (AG)₈YA (0.35, 0.33, 0.31, respectively) and lowest PIC index of 0.16 for two primers, namely, (CA)₆AC and BC835. The EMR values may be influenced by the fraction of polymorphic loci. The highest EMR was detected with the primers (AG)₈YA (17.05), BC841 (12.07), BC857 (12.00), (CAG)₅ (10.56) and the lowest was shown by the primer (CA)₆AC (0.67) with a mean EMR of 8.03 per primer. The usefulness of the system markers used was also determined by the calculation of MI for each primer. The highest MI was shown by the primers (AG)₈YA (5.35), BC857 (3.90), BC841 (3.40), (CAG)₅ (3.14) and the lowest in the primer (CA)₆AC (0.11) with a mean MI of 2.23 per primer. The discrimination potential of each primer was expressed by h_i, where the highest value was detected in BC810 (0.58) and the lowest value in (CT)₈TC (0.02). The average value of h_i for all primers (0.18) indicates that all samples are diverse and primers used are able to discriminate between all samples. The RP is the ability of a primer to differentiate between genotypes. The average RP was 8.48 per primer. The highest RP value was detected with the primer (AG)₈YA (15.04), BC841 (14.92), BC810 (14.63), (CAG)₅ (11.58), BC807 (10.04) and the lowest with the primer (CA)₆RG (1.33).

The moderate values of PIC, EMR, MI, h_i, RP parameters for the most ISSR primers could be attributed to the large genetic variability among the broomrape genotypes and/or highly informative ISSR markers used in this study.

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Keywords: broomrape populations, genomic polymorphism, ISSR analysis, informative markers.

GENOME OF THE NICOTINE-DEGRADING *PAENARTHROBACTER NICOTINOVORANS* ATCC 49919

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Paenarthrobacter nicotinovorans ATCC 49919 is a nicotine degrading microorganism isolated from soil on which tobacco was cultivated. Because the bacterium uses the pyridine pathway for nicotine degradation, it could be used for the conversion of nicotine containing waste into valuable green chemicals such as 3-succinoylpyridine or 6hydroxynicotine. Here, two technologies were employed to sequence the strain's genomic DNA. Illumina NovaSeq 6000 produces short reads and, in contrast, the Oxford Nanopore Technology MinION yields long read sequences. The complete genome was obtained using the hybrid assembler Unicycler. The genome is organized in two circular replicons, the largest replicon representing the chromosome of 4 316 184 bp, with an overall GC content of 63.2%.

The second replicon measures 165 141 bp and is the pAO1 megaplasmid, with an overall GC content of 59.7%. The chromosome presented a total of 3953 coding sequences, 54 tRNAs, 2 ncRNAs, 1 tmRNA and 6 identical ribosomal operons. The plasmid shows 99.99% identity with the previously described pAO1 megaplasmid sequence (GenBank entry AJ507836.131). Low identity at the nucleotide level was found when comparing the complete genome of *P. nicotinovorans* ATCC 49919 with genomes of nicotine degrading microorganisms that also use the pyridine pathway for nicotine degradation. Hence, the complete genome sequence of *P. nicotinovorans* ATCC 49919 provides a necessary reference genome for microorganisms using the pyridine pathway for nicotine degradation and, more importantly, is an essential starting point for exploring the regulation of nicotine catabolism.

Keywords: *Paenarthrobacter nicotinovorans* ATCC 49919, sequence the genomic DNA, genome organization, nicotine catabolism, pyridine pathway.

MOLECULAR ANALYSIS OF PROKARYOTIC MICROBIAL COMMUNITIES IN A TYPICAL CHERNOZEM

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The works were carried out in 2021 in the Biotron long-term stationary experiment. 4 variants of chernozem of two land use systems were studied: arable land of fodder crop rotation with alfalfa: 1 - control, without fertilizers, 2 - mineral background, 3 - organic background (cattle manure) and a forest belt located nearby. Molecular genetic analysis was performed using the equipment of the Central Collective Use Center "Genomic Technologies, Proteomics and Cell Biology" of the All-Russian Research Institute of Microbiology, St. Petersburg, Russia.

High-throughput pyrosequencing of amplified DNA (sequencing of the M4 variable region of the 16S rDNA gene) revealed 12 types of *Bacteria* (*Proteobacteria*, *Actinobacteriota*, *Bacteroidota*, *Firmicutes*, *Acidobacteriota*, *Verrucomicrobiota*, *Planctomycetota*, *Myxococcota*, *Nitrospirota*, *Fibrobacteriota*, *Gemmatimonadota*, *Chloroflexi*) and 1 type *Archaea* (*Crenarchaeota*). The largest number of phyla (13) was noted in the soil of the control and in the soil of the mineral background. There were 12 of them in the soil of the organic background, and 11 in the soil of the forest belt. The dominant position (abundance over 10%) was occupied by 3 phyla, including 2 phyla of *Bacteria*: *Proteobacteria* and *Actinobacteriota* and archaeotes: *Crenarchaeota*. In all cases, the strongest competition between bacteria and archaea is observed, and despite the fact that archaeota are qualitatively smaller, they are in the lead everywhere, except for the soil of the forest belt. The contribution of commonly found phyla (1-5%) was 9.2-14.1% with the largest values in the soil of the organic background (14.1%) and the smallest in the soil of the forest belt (9.2%). These are usually 5 phyla: *Firmicutes*, *Acidobacteriota*, *Planctomycetota*, *Myxococcota*, and *Bacteroidota*. The total participation of rare phyla ranged from 0.98 to 1.6%. The lowest abundance was recorded by phyla of the unfertilized background, then organic and mineral backgrounds. They were 4-5 phyla: *Myxococcota*, *Nitrospirota*, *Gemmatimonadota*, *Fifrobacteriota*, and in some cases *Chloroflexi*.

Acknowledgments: Research was carried out within the framework of research project 20.80009.5107 "Efficient use of soil resources and microbial diversity through the use of elements of biological (organic) farming."

Keywords: chernozem, prokaryotic microbial communities, pyrosequencing of DNA, bacteria, archaea.

CONDITIONED REFLEX LEARNING AND MEMORY OF WHITE RATS OF DIFFERENT AGES UNDER THE INFLUENCE OF THE BIOMASS OF STREPTOMYCETES ISOLATED FROM THE SOILS OF THE REPUBLIC OF MOLDOVA

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The aim of this work is a comparative study of the effect of long-term consumption of biomass of strains *Streptomyces massasporeus* CNMN-Ac-36 and *Streptomyces fradiae* CNMN-Ac-11 isolated from the soils of the central part of Moldova on the process of developing defensive conditioned reflexes and conditioned reflex memory in white rats of different ages (young and old).

The studies were performed on male Wistar rats. For 90 days animals of the experimental subgroups received daily a food supplement to the standard diet at a dose of 250 mg/kg of live weight dried biomass of two local strains of streptomycetes – *Streptomyces massasporeus* CNMN-36 or *Streptomyces fradiae* CNMN-Ac-11 grown on a nutrient medium with previously determined amino acid and lipid composition. 90 days after the animals began to consume the biomass of streptomycetes, at the age of 4 months (young) and 15 months (old), they began to develop conditioned reflexes. Rats that were fed a standard diet served as controls. To study the process of associative learning the method of developing a conditioned reaction of active avoidance of a painful stimulus was used. In order to study the processes of conditioned reflex memory the dynamics of the latent period of the avoidance reaction was determined.

It was found that long-term consumption of biomass of local strains of streptomycetes – *Streptomyces massasporeus* CNMN-36 and, to a greater extent, *Streptomyces fradiae* CNMN-Ac-11 significantly stimulates the development of a conditioned response of active avoidance in young and, especially, old animals, thereby facilitating the process of conditioned reflex learning. In addition, the consumption of *Streptomyces massasporeus* CNMN-36 biomass and, to a greater extent, *Streptomyces fradiae* CNMN-Ac-11 significantly reduces the latent period of the avoidance reaction at various times after the development of a conditioned active avoidance reaction in young and, especially, old animals, thereby contributing to slowing down the extinction of traces of conditioned reflex memory.

Thus, local strains of streptomycetes *Streptomyces fradiae* CNMN-Ac-11 (primarily) and *Streptomyces massasporeus* CNMN-36 (to a lesser extent), isolated from the soils of Moldova, are promising for further research in order to isolate and identify substances with neuroprotective and nootropic properties.

Keywords: streptomycetes, conditioned reflex learning, memory, neuroprotective activity, nootropic properties.

ACTIVITY OF DIGESTIVE ENZYMES OF THE SMALL INTESTINE OF RATS WITH DIFFERENT CONSTITUTIONAL STRESS REACTIVITY

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The aim of the work is to study the activity of membrane-bound enzymes (alanine aminopeptidase, maltase, sucrase, and glucoamylase) in different parts of the small intestine of rats with different levels of constitutional stress reactivity.

Rats were divided into three groups depending on the level of constitutional stress reactivity: 1 - high stress reactivity; 2 – average level of stress reactivity; 3 – low stress reactivity. Constitutional stress reactivity was assessed using a set of behavioral tests and analysis of histopathological differences in the tissues of the stomach, thymus and adrenal glands in some animals of each group after immobilization stress. Enzyme activity was determined in the homogenates of the mucous membrane of the small intestine.

The highest values of alanine aminopeptidase activity in all parts of the small intestine were recorded in rats with low stress reactivity compared with animals with high and low stress reactivity (differences between groups reach 38%, $P < 0.05$), and the lowest values were recorded in rats with high stress reactivity except for the ileum. It should be noted that we previously recorded an increased overall proteolytic activity of the small intestine in rats with low reactivity (Sheptytsky et al., 2019). The highest activity of maltase in the parts of the small intestine with the exception of the ileum was observed in rats with an average level of stress reactivity compared to animals with high and low levels of stress reactivity, while in animals with high and low stress reactivity the values of maltase activity are similar. In the ileum the highest maltase activity was recorded in rats with high stress reactivity. The highest sucrase activity in the duodenum and proximal jejunum is observed in rats with an average level of stress reactivity compared with animals with low and, especially, high levels of stress reactivity (differences between groups reach 62%, $P < 0.05$), in the distal part of the jejunum and ileum there was a tendency to an increase in sucrase activity in rats with high stress reactivity compared to animals with high and, especially, low stress reactivity. The highest activity of glucoamylase is observed in rats with high stress reactivity in the parts of the small intestine, with the exception of the proximal part of the jejunum, where glucoamylase activity is highest in rats with medium stress reactivity, and its minimum values are observed in animals with low stress reactivity.

Thus, the results obtained demonstrate the existence of a relationship between the genetically determined stress reactivity of the organism and the digestive function of the small intestine.

Keywords: membrane-bound enzymes, constitutional stress reactivity, small intestine.

MEMBRANE HYDROLYSIS OF MALTOSE IN THE SMALL INTESTINE UNDER THE INFLUENCE OF DIETS WITH DIFFERENT CONTENT OF CARBOHYDRATES IN EARLY POSTNATAL ONTOGENESIS

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The purpose of this work is to study the membrane hydrolysis of maltose and the absorption of glucose derived from maltose in isolated loops of the rat small intestine under the influence of diets with different carbohydrate content in early postnatal ontogenesis. The studies were performed on male Wistar rats. After weaning the rat pups of the experimental groups were kept for 6 weeks on diets with a high (78.2% of energy intake) or low (27.9% of energy intake) content of carbohydrates or on a carbohydrate-free diet, then a part of the animals of each of the experimental groups were kept on a standard diet for 3 days, 2 or 6 weeks, or 4 months. Animals kept after weaning on a standard diet served as controls. For the study of digestive and transport processes, the method of perfusion of isolated loops (20 cm) of the rat small intestine in situ was used. Maltase activity was determined in the intestinal mucosa by the glucose oxidase method. It was found that a high-carbohydrate diet leads to an increase in maltose hydrolysis and absorption of the resulting glucose (by 1.2–1.5 times) depending on the initial maltose concentration (12.5; 25 and 37.5 mM). Under the conditions of a low-carbohydrate diet, the studied parameters do not change at a low concentration of maltose (25 mM) and at its higher concentrations they decrease by 1.3-1.5 times. Changes in glucose absorption are accompanied respectively by an increase or decrease in the active component of its transport. As a result of a carbohydrate-free diet there is a sharp decrease in maltose hydrolysis and especially glucose absorption (by 3 or more times). The transfer of animals from a low-carbohydrate diet to a standard diet leads to normalization of glucose absorption after 3 days and membrane hydrolysis of maltose after 2 weeks. The transfer of animals from a high-carbohydrate diet to a standard one leads only to a partial normalization of the intensity of hydrolysis and transport processes even after 6 weeks. The transfer of animals from a carbohydrate-free diet to a standard one causes a gradual increase in membrane hydrolysis of maltose and glucose absorption, however, even after 6 weeks, their intensity is 1.5 times lower than in the control group. The activity of maltase was reduced compared to the control immediately after leaving the low-carbohydrate and especially carbohydrate-free diets and, to a lesser extent, 4 months after the transfer of rats from the carbohydrate-free diet to the standard one. As a result of a high carbohydrate diet, maltase activity increases markedly and remains above the corresponding control values for the next 4 months.

Thus, long-term high-carbohydrate and carbohydrate-free diets in early postnatal ontogenesis in contrast to a low-carbohydrate diet, contribute to the development of disorders in the membrane hydrolysis of maltose and absorption of the glucose derived from maltose in the small intestine.

Keywords: postnatal ontogenesis, membrane hydrolysis of maltose, absorption of glucose, diets with different carbohydrate content, development of disorders in the membrane.

THE PERSPECTIVE OF THE USE OF PREBIOTICS IN METABOLIC DISORDERS

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Through metabolomics medicine, the aim is to identify the mechanisms that determine the appearance of autoimmune and chronic diseases to avoid the causes of their appearance. The breakdown of proteins, fats, carbohydrates, and fiber results, through the involvement of the intestinal microbiome, in different metabolites that have beneficial or harmful effects on the appearance of diseases: obesity, diabetes, fatty liver, etc. The paper aimed to study the impact of prebiotic substances on blood glucose levels in rats depending on the carbohydrate content in their diet.

Two complex prebiotic compositions were elaborated, and their impact on the blood glucose level and body mass of laboratory rats was studied, distributed in 5 groups. The first batch received the standard ration (control), the second - composition 1, the third - composition 1 + glucose, the fourth - composition 2, and the fifth - composition 2 + glucose. The compositions included two different sources of inulin (fructan) fibers with a hypoglycemic effect, products with soluble/insoluble fiber content and enterosorbent. The results obtained show an increase in glucose content in the batch without the administration of prebiotic compositions (control), which was 7.49% compared to the initial level. After 30 days of administration of prebiotic no. 1 in group II (without glucose) - a decrease in its level was observed by 8.88%, while in group III (with glucose) - by 5.44%, compared to the content initial. Prebiotic no.2 demonstrated a higher efficacy because the glucose level, in this case, decreased in groups IV and V, respectively by - 8.61 and 10.45%, compared to the initial one. Despite the administration of glucose for 30 days, concomitantly with prebiotics in groups II and IV, an increase in body mass was observed only by 7.14 and 8.75%, being however higher compared to the control group, but lower than batches that did not receive glucose. This is due to prebiotic fibers, which facilitate the absorption of intestinal fats, leading to control of body mass. The more pronounced hypoglycemic effect of prebiotic no. 2 is explained by the role of the constituent elements. Inulin slows down digestion, allowing carbohydrate sugars to be gradually released into the bloodstream, balancing blood sugar. Co-administration of the pectin and a small amount of insoluble fiber lowers total cholesterol and low-density lipoprotein (LDL) cholesterol. Enterosorbents absorb harmful substances and neutralize toxins, absorb the products of the vital activity of pathogenic microorganisms, and improve metabolism.

Thus, the elaborated compositions have a different hypoglycemic effect, lead to a moderate decrease in blood glucose levels in rats, and have shown efficacy even in conditions of a major carbohydrate intake in their diet. The selected prebiotic components, serving as a substrate for the normal intestinal microbial flora, have a substantial role in the body's ability to assimilate carbohydrates (glucose tolerance) in regulating body mass, which opens the prospect of their use as a component of symbiotics in human metabolic disorders.

Keywords: prebiotics, metabolic disorders, autoimmune and chronic diseases, component of symbiotics, intestinal microbial flora.

3D PRINTING MACROMOLECULAR MODELS FOR TEACHING AND DEMONSTRATION

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Advancement in three dimensional (3D) printing hardware and software makes this additive manufacturing technique rather cheap and easy to implement in schools and universities. 3D printing can be used to fabricate various teaching materials for numerous disciplines, and it has a great application in teaching life sciences. Starting with the atomic coordinates downloaded from RCSB PDB, one can create a physical model of any biomolecular structure that would help better understand/demonstrate key aspects such as molecular size, symmetry, or shape. Besides that, 3D printing of molecular models can also be used to engage students in designing their own custom printable molecule of choice, offering a great opportunity to firsthand study molecular structure. After designing and printing of more than 30 different models of molecules and macromolecular complexes (including the T7 replisome, the DNA sequencing protein nanopore, the 20S Yeast proteasome or an alpha-amylase with amylopectin), several lessons have been learned. First, surface renderings of macromolecules are extremely easy to create and print and can be tackled by any user. Surface representation of subunits of the same multimeric protein can be printed separately in different colors with a high visual impact. Unfortunately, the subunits cannot always be assembled back into the complex due to frequent overhangs and tight channels. Cartoon representations can be used only for rather small proteins as are harder to print. A higher rate of success in printing complex cartoon models can be obtained with a multilateral capable 3D printer and using soluble plastic as support. Balls and sticks models are suitable for small molecules. Printing a balls and sticks or a surface model of DNA in flexible TPU has a great visual impact when demonstrating the assembly of the nucleosome. In the end, a set of models with printing instructions are provided which could serve as a starting point for anyone wishing to print its own macromolecular models on the cheap.

Keywords: 3D printing, macromolecular models, teaching materials.

MANAGEMENT ISSUES OF CHRONIC LYMPHOCYTIC LEUKEMIA

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Chronic lymphocytic leukemia (CLL) is a substantial problem of public health and oncology due to the increase of morbidity rates, frequent and refractory relapses, development of infectious complications, often leading to poor outcomes.

The aim of the study was to determine the diagnostic features and evaluate the management outcomes of infectious complications in CLL.

The study enrolled 82 patients with CLL aged 45 to 86 years (median age 66.2 years), who were treated and followed up at the Institute of Oncology of Moldova from 2000 to 2021. There were 47 men (57.3%), women – 35 (42.7%). The diagnosis was confirmed according to the IWCLL criteria based on the study of a complete blood count with the detection of lymphocytosis $\geq 5 \times 10^9/l$, myelograms with the bone marrow infiltration $\geq 30\%$ and immunophenotyping. According to Binet Classification, 54 (65.9%) patients were diagnosed and followed up in stage A, and 28 (34.1%) patients – in stage B. The study was related to the out-patient and hospitalized care at the comprehensive cancer center.

All immunophenotyped CLL cases proved to be CD20-positive. The study of the age distribution of CLL revealed the predominance of patients aged 60-79 years. The study of the disease evolution showed that out of 54 stage A patients, 22 (40.8%) developed transformation of CLL into stage B in the period from 1 to 4 years. Transformation into stage C was revealed in 10 (35.7%) cases. The study of the frequency of infectious complications noted that in 36 (43.9%) cases there was at least one of their event. Bacterial infectious complications involved the respiratory system more commonly (29 patients, or 80.6%): acute pneumonia in 10 (27.8%), acute bronchitis in 7 (19.4%), exacerbation of chronic obstructive bronchitis in 11 (30.6%) and tuberculosis in 1 (2.8%) patient. Viral herpetic complications were diagnosed in 2 (5.6%) cases: each of them on the lips and genitals. The infectious complications with the affection of the other organs and systems were detected in 5 (13.8%) patients: in 3 (8.2%) – kidneys and urinary tract, in 2 (5.6%) – acute otitis. Over a period of 3–19 years, the fatal outcomes occurred in 16 (19.5%) patients: due to the infectious complications – in 6 (37.5%), as a result of CLL progression – in 5 (31.3%), because of the secondary tumors – in 4 (25.0%), and in one (6.2%) – due to the acute cerebro-vascular stroke. The overall survival rates after 3 and 5 years were 100% and 95.7% in stage A patients, and 84.8% and 55.4% – in stage B patients, respectively. The diagnosis of CLL was frequently proved in males aged 60-79 years due to the detection of the increased lymphocyte count in the complete blood count and bone marrow aspirate. Infectious complications may be considered as common evolution features and causes of death in CLL, especially in stage B.

Keywords: chronic lymphocytic leukemia, diagnostic features, infectious complications, increased lymphocyte count, disease evolution.

THE MODIFICATION OF FREE AMINO ACIDS RATS FED WITH PROTEIN RATION

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Proteins are known to be an essential component of most body tissues, acting as an energy substrate in the development of catabolic processes. Free amino acids in physiological fluids act as metabolic regulators, and their metabolism in the cell is extremely extensive because their amount significantly increases the number of amino acids involved in protein biosynthesis. In the literature, although enough data are known about the changes that take place in the intermediate metabolism of free amino acids, the impact of physical exertion on the background of a protein-rich diet on the body is little known. The research aim is the modification the summary content of the functional groups of free amino acids (ALL) in the blood of senile rats fed protein ration in combination with physical effort (PE).

The investigations were performed on ten white laboratory rats (males-old -24-30 months), divided into two experimental groups: group I – with protein ratio (PR) (25% - proteins, 55% - carbohydrates, 20% - lipids) without PE; group II – with protein ration with PE. The animals practiced maximum dynamic PE – swimming daily for 31 days. The water temperature is +27 °C. Duration of the experiment – 31 days. The content of free amino acids in the serum and erythrocytes was determined by the ion-exchange liquid chromatography method. The analysis of the total content of AAL was performed by functional groups, depending on the physiological role of amino acids: non-essential, essential, immunoreactive, glycogenic, ketogenic, proteinogenic, and sulfur amino acids.

Analysis of the AAL serum concentration, by functional groups, in group II senile animals compared to group I, which depends largely on the absorption possibilities of enterocytes from the small intestine and partially from the large intestine, shows a decreasing tendency of the group non-essential amino acids (by 4.7%), and a significant decrease in immunoreactive, essential, glycogenic, ketogenic, proteinogenic and sulfur amino acids (respectively by 48.0; 39.2; 25.7; 17.6; 36.7; 33.1%). This decrease is possibly due to disorders of the absorption processes of amino acids in food in senile animals, as well as changes in the microbiome of the small and large intestine, which actively synthesize essential AALs. The analysis of free amino acids in erythrocytes of rats from group II compared to group I identified a marked decrease in the content of amino acids. Intensive PE in senile rats causes a downward trend in the summary content of sulfur amino acids by 13.2%, and a significant decrease in other functional groups of AAL (non-essential - by 38.7%; essential - by 64.4%; immunoreactive - by 48%; glycogens - by 44.7%; ketogenic - by 66.8%; proteinogenic - by 50.8%) compared to their content at animals without PE. The low content of free amino acids in the animals subjected to dynamic physical effort, fed with protein ration, is probably conditioned by the predominance of energetic mechanisms determined by PE. Thus, the impact of protein ration in association with PE in senile rats is directly related to the AAL content, depending on the functional group, in serum and erythrocytes, causing a decrease in the AAL content in all functional groups.

Keywords: protein ration, modification of free amino acids, physical effort, functional groups, intermediate metabolism.

PSYCHOGENIC STRESS IN HOSPITALIZED PERSONS

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People react very differently when they are sick and there are only weak correlations between the intensity of the immune response and disease behavior, and psychogenic stress is an obligatory component of the clinical picture when the patient is aware of the consequences of the disease to their life activities ensuring.

According to the concept of psychosomocreatology, any internal or external factor, or situation if it is assessed as significant and dangerous for oneself and others, or as interfering with daily activities, causing emotions, becomes a trigger for the development of psychogenic stress. And patients who are hospitalized are quite deeply aware of the danger of the physiological state of their organism to life activities ensuring, are highly disposed to the development of psychogenic stress, which predetermined the study of the features of its manifestation.

It has been found that the criteria for psychogenic stress in hospitalized patients are irritability, aggressiveness, disinhibition, muscle excitability, experiencing anxiety, fear, panic, phobias, difficulty concentrating, decreased attention, impaired social relationships, deviant behavior, frustration, heart palpitations or heart and breathing rate fluctuations, sleep disorders, dark thoughts, memory impairment. Hypo- and hyperkinetic variants of stress reactions are distinguished: dissociative stupor, which is manifested by sudden motor retardation, profuse cold sweating, expression of horror on the face, eyes wide open; hyperkinetic variant, exteriorizing with pronounced general agitation, psychomotor agitation, unfocused movements, chaotic autonomic reactions (tachycardia, pallor, sweating, etc.). The duration of such reactions is on average up to 48 hours; the symptoms begin to decrease on average after 6-8 hours.

In decompensated patients hospitalized for conservative treatment, psychogenic stress most often provokes the following types of disorders: 1) anxiety disorders: the patient is constantly focused on anxious thoughts and ideas, their phobia. This state differs from delirium in the fact that these ideas are quite real, and the patient cannot get rid of the fear of losing their job, incurable illness, etc. They realize that their fears are most likely unfounded, however, they cannot stop thinking about them; 2) panic disorder, characterized by sudden and causeless panic attacks - signs of uncontrolled fear. Patients experience inexplicable horror, begin to suffocate; sweating, dizziness, fear of death, tremors occur. The attack lasts from several minutes to half an hour; 3) depressive disorder - the patient is in a state of apathy, a loss of interest in everything that previously seemed important and interesting; now it seems to the patient meaningless; they see the future exclusively in black colors, are prone to self-deprecation. Depression is often accompanied by drowsiness, loss of appetite, and vegetative vascular dystonia.

Attenuation of psychogenic disorders caused by the stress of hospitalization, depending on the severity of the exacerbation of the nosological disease, should be carried out with psychotherapy or pharmacological preparations.

Keywords: psychogenic stress, hospitalized persons, immune response, clinical picture, disease behavior.

ANALYSIS OF Y CHROMOSOME MICRODELETIONS AND CFTR GENE MUTATIONS AS GENETIC MARKERS IN MALE INFERTILITY

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Globally it is estimated that about 15-20% of couples are affected by infertility current data from the literature show that in 50% of cases the male causal factor is involved. Genetic abnormalities cause 15%–30% of male factor infertility. Y microdeletion and CFTR gene mutations are the most frequent known molecular genetic causes associated with azoospermia and severe oligozoospermia.

The purpose of the study was to evaluate genetic markers of Y-chromosome microdeletions of the AZF and cystic fibrosis transmembrane conductance regulator (CFTR) gene mutations in azoospermic infertile men.

The study was carried out on infertile men with azoospermia recruited among infertile couples referred for reproductive treatment. All patients signed an informed consent. The endocrine markers: FSH (Follicle-stimulating hormone); LH (Luteinizing Hormone); and testosterone were evaluated. They were investigated by molecular testing for AZF and CFTR gene. Multiplex Polymerase chain reaction (PCR) was performed using Y-specific markers for AZF region: AZFa (sY84, sY86, DBY1, sY620), AZFb (sY117, sY127, sY134, SY143), and AZFc (sY254, sY255, sY153, SY158). The

detections of sY14 (SRY) and ZFX/ZFY were employed as internal controls. Two common mutations $\Delta F508$ and G542X were tested of the CFTR gene.

Deletions of Y chromosome were identified in 9 (9,9%) of 91 patients with azoospermia. Deletions of AZFc - sY153, sY158, sY254 and sY255 locus were observed in four of nine azoospermic patients 55,5%. In two (22,2%) patients were detected with deletion of AZFb region, deleted markers were sY117, sY127, sY134, sY143. Deletions affecting both AZFb and AZFc loci were found in two patients 22,2%. The average level of FSH was $6,4 \pm 3,5$, LH $6,2 \pm 3,2$ and testosterone $3,3 \pm 1,4$, of patients with microdeletions. Three (3,3%) patients were identified with CFTR gene mutation $\Delta F508$, for calculating the risk of recurrence in offspring was investigated and his wife, found himself homozygous.

Molecular genetic screening is important to define the cause of spermatogenesis defect. This is important to provide a correct diagnosis and more effective solutions to couples with infertility before reproduction treatment is applied.

Keywords: molecular genetic screening, spermatogenesis defect, Y chromosome microdeletions, reproduction treatment, CFTR gene mutations, genetic markers, male infertility.

MOLECULAR-GENETIC ASPECTS OF MIGRAINE

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Migraine is a complex genetically determined disorder characterized by episodes of moderate to severe headaches that are estimated to affect approximately 1 billion people worldwide, predominantly women. According to the 2016 Global Burden of Disease Study, migraine is the second leading cause of disability and accounts for more disabilities than all other neurological disorders combined. Migraine is a multifactorial pathology, disabling, recurrent, neurovascular headache disorder. It can be classified into subtypes, according to the International Headache Society's headache classification committee: migraine without aura, migraine with aura and chronic migraine.

The genetic basis of migraine is complex with the involvement of several loci and genes in pathogenesis; it can rely on more than one genetic source in different genomic locations that act in tandem with environmental factors to determine the susceptibility and characteristics of the disease. Identification of mutations in the following genes: CACNA1A, ATP1A2, SCN1A, PRRT2, SLC4A4, NOTCH3 could predict targeted prophylactic treatment of migraine. Many of these genes encode proteins involved in regulating glutamate neurotransmission and the normal formation of synaptic plasticity. Molecular, anatomical, and functional modifications induce high sensitivity to homeostasis fluctuations, low adaptability in neural substrate, and recurrence of headaches. Understanding the genetic predisposition to migraine and the discovery of several susceptible gene variants and genetic polymorphism define the most compelling hypothesis for generalized neuronal hyperexcitability and generalized neuronal changes in migraines. Concerning the headache itself, studies on the understanding of molecular mechanisms show that the activation of the trigeminovascular pathway is a precondition to explain why pain is limited to the head, often affects the periorbital area and the eye and is intensifying with increasing intracranial pressure.

Changes in vascular caliber are the result of abnormal secretion of chemicals present in the brain. It has been determined that in the pathogenesis of migraine, although incompletely understood, the trigeminal nerve and its axonal projections to intracranial vascularization are involved. Nociceptive signals from the trigeminovascular system are transmitted to areas of the brain that produce the perception of migraine pain.

Keywords: migraine, multifactorial pathology, nociceptive signals, genetic predisposition, generalized neuronal hyperexcitability.

USE OF AI-ASSISTED TOOLS IN HEREDITARY DISEASE DIAGNOSTIC

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One of the most difficult parts of medical genetics is analysis of a huge amount of patient's data and presumption of diagnostic. It can be even harder if we take in account multiplicity of hereditary diseases, presense of many phenocopies at some of them and purely human factors as tiredness or some form of unconscious prejudices, all of them having possibility to interfere and impair ability to put correct diagnosis, what definitively is not beneficent for patient.

This situation began to change in 1980's, when first expert systems in domain of human genetics appeared. But those systems included only limited amount of data, were statical, not able to self-improve and were composed by humans, so being influenced by all human errors.

Major changes in this field began in 2011, when FDNA company was founded and began to develop Face2Gene program, based on next-generation phenotyping and artificial intelligence technologies. This program compares patient's face photo and clinical signs with a database of previously diagnosed patients using AI and proposes a list of possible diagnoses with ranking for similarity with photo and with clinical signs.

This software helped us to diagnose such diseases as Coffin-Siris, Bloom and Myhre syndromes and much more. In this publication we summarize our experience of Face2Gene software and next-generation phenotyping use.

Keywords: AI, next-generation phenotyping, medical genetics.

OVERLAP OF CLINICAL MANIFESTATIONS IN MITOCHONDRIAL DISEASES

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Mitochondrial diseases (MD) are a clinically, biochemically and genetic heterogeneous group of disorders caused by mutations in genes that encode proteins involved in mitochondrial function. Neuropathy, ataxia, and retinitis pigmentosa (NARP) and Leigh syndrome (LS) are both established mitochondrial syndromes; sometimes the clinical manifestations in these diseases may overlap.

LS is defined as an early-onset progressive neurodegenerative MD typically characterized by subacute onset of psychomotor regression and encephalopathy associated with the development of bilateral symmetrical lesions in the basal ganglia, thalami, subthalamic regions, mesencephalon, and brainstem, which are the hallmarks of the disease. In contrast, NARP is another mitochondrial syndrome characterized by sensory axonal neuropathy, ataxia, retinitis pigmentosa, epileptic seizures, cognitive impairment, sensorineural hearing loss, diabetes mellitus, and cardiomyopathy. Our patients manifested with recurrent seizures, muscle weakness, psychomotor retardation, feeding difficulties, elevated lactate and alanine; meanwhile, additional symptoms include exercise intolerance and partial atrophy of the optic nerve in the case of the patient with NARP syndrome, as well as tremor, ataxia and abnormal ECG in the patient with LS. Considering the multisystemic impairment and the presence of predominantly neurological clinical manifestations, an underlying mitochondrial respiratory chain disorder was suspected taking into account the clinical phenotype of the patients, and confirmed by biochemical, instrumental and genetic examinations. Evaluating the clinical criteria for mitochondrial diagnosis according to the Nijmegen Mitochondrial Disease Criteria Scale, there were counted 8 points for both patients as scoring for definite mitochondrial disorder. The first-line investigations used in this case of inborn error of metabolism indicated elevated lactate in blood [2.2-2.4 mmol/L (LS) and 3.7-7.8 mmol/L (NARP); ref. val. 0.7-2.1 mmol/L], hyperalaninemia Ala [594 μ mol/L (LS) and 1038 μ mol/L (NARP), ref. val. <450 μ mol/L], Ala/ Lys ratio [3.44 (LS) and 11.8 (NARP), abnormal if >3] and hyperaminoaciduria. Neuroimaging findings consisted of pathological foci in the bilateral basal nuclei in the case of NARP, and a symmetrical distribution of lesions along thalamus, mesencephalon, brainstem, medullary tegmentum and cerebellar hemispheres (periventricular) and medulla oblongata, in a pattern that is characteristic of LS.

Genetic analysis revealed the m.3243A>G mutation in the *MT-TL1* gene in the case of the patient with LS and in the patient with NARP syndrome the m.8993T>G mutation was identified in the *MT-ATP6* gene of the mitochondrial genome. Early onset in the presence of complete health, the polymorphism of clinical manifestations, such as a central nervous system lesion, muscle weakness, impaired psychomotor development, and seizures in a child should prompt the clinician to consider a mitochondrial disorder namely LS or NARP, and conduct further investigations such as measurement of blood lactate, magnetic resonance imaging, and if possible, genetic analysis.

Keywords: mitochondrial diseases, mutations, syndrome, genetic analysis, genome.

THE BENEFITS OF L-ARGININE IN PREECLAMPSIA

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Preeclampsia is a multisystem complex disorder with an incidence of 3-7% for nullipara and 13% for multipara and is a major contributor of both fetal as well as maternal morbidity and mortality. Endothelial nitric oxide synthase (e-NOS) promotes the synthesis of NO which dilates the arteriolar bed whereas in preeclampsia there is deficiency of e-NOS resulting in vasoconstriction of the placental vasculature as well as the vasculature of the other organs.

L-arginine is the substrate of nitric oxide (NO), a potent vasodilator, which may play a major role in regulating blood pressure. Its level in the blood during pregnancy is increased and its availability is very important to ensure proper and adequate synthesis of nitric oxide in endothelium and since there is increased destruction of NO in abnormal endothelium, the requirement of L-arginine increases.

Hypertension, proteinuria, fetal growth retardation, and glomerular damage could be induced by blockade of NO synthesis, while hypertension induced by NO synthesis inhibition could be reversed by L-arginine supplementation. Studies show that daily intravenous infusion of Arg (20 g/day) for days to women with unknown causes of IUGR increased birth weight at term by 6.4% during later weeks of gestation. In another study conducted by Neri et al., they examined the effect of L-arginine on utero-placental circulation in pregnancy complicated by intrauterine growth restriction in the third trimester and observed an enhanced level of nitrates/nitrites and the growth hormone. They also observed significant changes in various other Doppler parameters and improved fetal outcome. It was observed that after the supplementation of L-Arginine, there were significant changes in the mean values of various Doppler parameters from 1st to 3rd visit which were higher in the study group as compared to the placebo group. In different studies it has been seen that treatment with exogenous

Arginine increases fetal and neonatal outcome as well as improvements in Doppler parameters also helps in prolonging the pregnancy, but larger studies are required to further validate effects of L-Arginine. The mainstay of treatment in these patients is strict control of blood pressure using anti-hypertensive drugs and strict fetal and maternal monitoring. By examining the maternal and fetal vessels using Doppler ultrasound it is possible to determine, the risk of complications developing in the course of pregnancy long before clinical signs of preeclampsia as well as it helps in adequate fetal monitoring so that preventive and therapeutic measures can be undertaken early. L-Arginine proved to be an efficient intervention to improve the fetal and neonatal outcomes in patients with hypertensive disorders of pregnancy.

Keywords: preeclampsia, multisystem complex disorder, pregnancy, L-arginine, clinical signs.

COMPARATIVE *IN SILICO* ANALYSIS OF PRIMERS USED FOR VERTEBRATE METABARCODING

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DNA metabarcoding is a relatively new method of biomonitoring and refers to the molecular analysis of a mixture of intra- and extracellular DNA from various organisms. It allows the use of different target sequences for the simultaneous characterization of several organisms and has a big potential in biodiversity research. The environmental DNA (eDNA) analysis began to be applied for studying vertebrate species and like any metabarcoding research requires a correct selection of primers. In this study we aimed to perform *in silico* PCR and compare the degree of conservation, taxonomic coverage (Bc) and specificity (Bs) of 2 universal primers pairs used for vertebrate DNA analysis. The first pair (Kitano-16S) was proposed in 2006 by Takashi Kitano and co-workers as universal primers for identifying vertebrate species, based on 16S mitochondrial DNA sequencing. The second pair (12S-V5) targets the 12S mitochondrial region and was designed by Tiayyba Riaz and collaborators in 2011. The 12S-V5 has been validated using bioinformatics algorithms (*ecoPrimers*) and experimental laboratory approaches. The current study includes 191 NCBI mitochondrial genomes of 140 species of birds, 42 species of mammals, 5 species of amphibians and 4 species of reptiles, most species being part of the fauna of the Republic of Moldova.

In order to perform *in silico* analyses, there were used following bioinformatics platforms and tools: *NCBI*, *OBITools*, *ecoPCR*, *SO Ubuntu*. The assessment of the primers conservation level and graphical representation were performed using R language and *ROBITools*, *ROBITaxonomy*, *ROBIBarcodes* libraries.

The *in silico* PCR of the studied 191 mitogenomes showed that the pair Kitano-16S represents a higher degree of conservation compared to the second pair; however 12S-V5 demonstrated a clearly superior performance with a taxonomic coverage of 97.91% and a specificity of 76.47%. Respectively, for the Kitano-16S pair resulted a 21.99% taxonomic coverage and a 90.48% specificity. Also, the 12S-V5 pair has a maximum of 2 mismatches for both primers while the Kitano-16S - a maximum of one mismatch for the forward primer with a total lack of mismatches for most amplicons. The metabarcodes length is about 200 bp for Kitano-16S pair and varies between 96 and 106 bp for the 12S- V5. Finally, the *in silico* amplification of the studied vertebrate mitogenomes is more efficient with the use of universal primers 12S-V5. This pair can be used to perform metabarcoding research that targets above mentioned mitochondrial genomes.

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Keywords: DNA metabarcoding, biomonitoring, *in silico* PCR, mitogenomes, primers.

DIFFERENTIATION OF THE FORMS OF PHENYLKETONURIA BASED ON GENETIC DETERMINISM

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Phenylketonuria(PKU) is an inborn error of metabolism, inherited in an autosomal recessive fashion resulting in >98% of cases from phenylalanine-hydroxylase(PAH) gene defect that leads to mental retardation if not treated.

Early diagnosis of PKU is possible due to neonatal screening, initiated in 1989 in the Republic of Moldova. Patient genotyping for PAH gene mutations has been used in predicting the evolution and individualization of disease treatment based on evidence from the BIOPKU database (<http://www.biopku.org>) and literature. 127 patients with PKU are registered in the Republic of Moldova. The analysis of pathogenic mutations was performed by PCR/RFLP methods and Sanger sequencing of the PAH gene. In 6 cases the DNA samples were damaged. The 121 patients, 45 genotypes were identified. Only 30 genotypes were reported in the BIOPKU database, 11 genotypes were found in the literature and 4 genotypes were not included in any of the sources. In 73.33% they corresponded to the classic PKU forms, requiring the hypophenylalanine diet as the treatment of choice, and 26.67% - Hyperphenylalaninemia. Responsiveness to BH4 was assessed based on evidence from the databases used. Thus, 5.10% of the total group were considered probably responsive at BH4, and 13.26% - responsive.

This study allows to conclude that about 20% of patients with PKU in the Republic of Moldova can be tested with BH4, with a potentially positive result, thus improving treatment with BH4 therapy. In addition, the BIOPKU database could be supplemented with the cases of 4 patients whose genotypes have not yet been registered.

Keywords: Phenylketonuria, genotype, responsiveness to Biopterin.

Session D

**ENVIRONMENT
PROTECTION AND
NATURAL RESOURCES
MANAGEMENT**

DENDROLOGICAL ASPECTS OF VEGETATIVE REPRODUCTION OF SOME WALNUT GENOTYPES

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Walnut (*Juglans regia* L.) is the most widespread tree nut in the world. There is a great diversity of genotypes differing in forestry, productivity, physical and chemical nut traits. Using a range of methodologies, from morphological markers to the most recent advances in genome analysis, many genetic studies of walnut have been conducted during the past years, including examination of diversity, determination of relationships within or among germplasm collections and populations, phylogenetic and origin elucidation, genetic map construction, and biotic or abiotic stress investigations. The genetic improvement of walnut has undergone great evolution. Some of them have been evaluated as promising and may serve as germplasm sources for breeding. Germplasm diversity is commonly evaluated with the help of morphological descriptors. This is usually the first step in classifying and describing germplasm and in studying heritability of traits for a new breeding program and selection of superior genotypes.

Cultivated walnut varieties, generally well adapted to climatic conditions of the different production zones, often lack some important agronomic characteristics. It would therefore be useful to select in natural populations or create through hybridization new cultivars combining characters of improved climate adaptation, early fruiting, high productivity, disease tolerance and quality fruit production. This is possible given the very large and so far unexploited variability within the *Juglans regia* L. species. Long juvenility period and high variation between trees in terms of different characteristics, usually make it impossible to establish uniform orchards. Yield and quality of fruit and kernel is low and could not compete with the production of the countries that have used the cultivars. So, breeding and introducing of new suitable walnut cultivars is necessary for walnut development. However, both the common walnut *Juglans regia* and black walnut *J. nigra* are quite particular in terms of optimal growing conditions. Hybrid walnuts provide an exciting alternative. It is possible to encourage and control hybridisation through tree breeding programmes. Hybrid species tend to have greater than either of their parents, and may be more tolerant to a wider range of conditions.

Root systems have a strong influence on the vigor of the tree. Vigorous rootstocks are commonly used to increase productivity or to decrease it. This feature is especially important when dealing with trees, as most of the research in breeding with walnuts has been looking for very vigorous scions that must be grafted on seedling rootstocks of unknown and heterogeneous growth capabilities. Once selected scions are shown to be very vigorous and productive on their own roots, it can be assumed that micropropagated selections of walnuts must result in more homogeneous and vigorous trees than the same selections grafted onto seedling rootstocks.

Plant breeding has always impacted food production and played a vital role in improving human nutrition. However, this has also increased uniformity within the world's agricultural crops, contributing to increased genetic vulnerability to biotic and abiotic stresses. For these reasons, it is important to understand better the impacts of modern plant breeding on genetic diversity. In the same way, intelligent management of this diversity could be of valuable assistance to breeders.

Keywords: *Juglans regia* L., vegetative reproduction, varieties, breeding, introducing.

GROUNDWATER CHEMISTRY OF SARMATIAN AQUIFERS IN RABNITA AND DUBASARI DISTRICTS

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In Rahnita and Dubasari districts the aquifers in the strata of Neogene age are used for the water supplies of the local population. The strata are represented by limestones, and in some areas and levels by marls and clays. Regionally, these strata belong to the Lower and Middle Sarmatian stage of Middle and Upper Miocene. In most cases there are no waterproof strata between Lower and Middle Sarmatian limestones, and the rocks of both sub-horizons represent a single, united aquifer. The limestones of Lower and Middle Sarmatian often are almost identical lithologically, and the limit between them is identified only by fauna. In Rahnita district the rocks of Lower Sarmatian are uncovered by the erosion of the River Dniester, and the Middle Sarmatian limestones lie above the level of erosion. Thus, the Middle Sarmatian is drained by water springs and the principal water horizon is located in the rocks of Lower Sarmatian only. In Dubasari district the Lower Sarmatian strata lie below the level of Dniester waters; they show low transmissivity, and these waters are not used exclusively for the supply of population. The principal water horizon is Middle Sarmatian only. The most wells in the district were drilled down until the limit of Lower Sarmatian or even Upper Badenian aquifers, but pump mostly the waters of Middle Sarmatian. There are no waterproof strata between the two Sarmatian horizons, but Middle Sarmatian waters dominate because of higher transmissivity values of the Middle Sarmatian limestones. In Rahnita district there are many wells in and around the district centre proper (Rahnita, Smalena, Harjau). The chemical composition of Lower Sarmatian groundwaters is mixed. The dominating anion is HCO₃, with about 40-60%, the second is SO₄, with about 25-40%, Cl almost never exceeds 20%. Among the cations, all 3 are presented above 20%, in the majority of wells Na is first, sometimes Ca or Mg. In Varancau, the chemical composition is HCO₃-Na-Mg, in Jura HCO₃-SO₄-Na-Mg, in Ofatinti HCO₃-SO₄-Mg-Na-Ca, in Crasnencoe HCO₃-Cl-Na. The waters are fresh, mineralisation rarely exceeds 1 g/l. In Dubasari district there are wells which pump only the Lower Sarmatian aquifer, with the upper part of the water-bearing strata below the sea level – in Cosnita, Lower Sarmatian is found on -6.8 m. The waters are fresh, mineralisation below 0.75 g/l. In Cosnita, the composition is HCO₃-SO₄-Ca-Mg-Na, in Dubasari HCO₃-Na. In many areas of Dubasari district the wells use both Upper Badenian, Lower and Middle Sarmatian aquifers. In the district centre proper, the chemical data show a mixed, 3-anion and 3-cation composition of groundwaters, with HCO₃ on the first place, and dominant Mg and Na among cations. In Tabuleuca, HCO₃ is 76% and Na 62%, in Molovata Noua HCO₃ stays at about 60-70%, Mg at 25-50% and Na more than 50%. In Cocieri SO₄ is dominant among anions at about 35-50%, and Mg at about 40-50%. In Crasnai Vinogradari and Alexandrovca Noua HCO₃ is at 50- 60% and Mg at about 60%. In Cosnita the waters of the Middle Sarmatian aquifer show the dominance of HCO₃ at 65-70%, Mg at about 35-40%, Na at about 25-40%, and Ca at 25-35%. The mineralisation of the Middle Sarmatian groundwaters in Dubasari varies from 0.5 to 1.35 g/l. In both districts the concentration of Mg very often exceeds normal, about 6-7 mg-equi/l. This, together with some concentration of Ca, makes the waters in the majority of areas and wells minerally hard, in many cases above 8-10 mg-equi/l. Of other components, both Cl and SO₄ do not exceed normal quantities established by the standards of state. In both districts the groundwaters used in the communal sector are taken from the first, the uppermost aquifer in the pre-Quaternary rock strata.

Keywords: *Sarmatian aquifers*, groundwater, chemical composition, mineralization.

**CONTRIBUTIONS TO THE STUDY OF SAPROXYLIC BEETLES
(INSECTS: COLEOPTERA) FROM THE REPUBLIC OF MOLDOVA**

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The paper includes the first analysis of saproxylic beetles stored in 4 entomological collections in Chisinau city, as well as own materials collected between 2005 and 2022 from the reserves Codrii, Plaiul Fagului, Pădurea Domnească and Prutul de Jos, landscape reserve Codrii Tigheci, Hârbovăț, Telița and some forest protection curtains in different country's districts.

As a result of investigation in the Republic of Moldova, were revealed 318 species of saproxylic beetles belonging to 212 genera and 44 families. Of these, 151 species are xylophagous, 57 polyphagous, 48 zoophagous, 40 mycophagous and 22 species are attributed to saprophagous, lichenophagous, parasitoid, phytophagous and coprophagous groups. Most of analyzed species are widespread: Palaearctic (80 species), Western Palaearctic (73), European (66), followed by Holarctic (21), Euro-Caucasian (19), Euro-Siberian (17) and cosmopolitan (14) species. The other 17 species are Mediterranean, Eurasian, Trans-Palaearctic and Near-Arctic.

The wide diversity of saproxylic insect species in forest ecosystems is an indicator of the state of the quality of the environment and also reflects the level of its functioning. For their development, saproxylic species require decaying wood. In the Republic of Moldova, forest management includes the evacuation of dead wood, most of which is removed from forest ecosystems, as a result, the number of saproxylic beetles in recent years has decreased dramatically.

According to the IUCN classification, only 14 species of saproxylic beetles are protected in the Republic of Moldova, of which 7 are critically endangered (CR), 2 endangered (EN) and 5 vulnerable (VU).

Acknowledgments: The study was supported by the projects 22.00208.7007.05/PD1 and 220.80009.7007.02.

Keywords: *Saproxylic beetles*, entomological collections, forest ecosystems, indicator, quality of the environment.

STUDY IN THE FIELD OF REDUCING THE HARMFULNESS OF A BIODIESEL POWERED ENGINE

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Given that energy requirements are growing, that fossil fuel reserves are being depleted, and the polluting effects of their use on the ecosystem are catastrophic, it has become imperative to find new ways to produce energy from alternative sources to replace these classic fuels. One of the main directions for reducing toxic emissions is the use of alternative fuels for internal combustion engines. Particular emphasis is currently being placed on reducing the emissions of exhaust gases from diesel engines, due to the extension of their scope and the increase in the total number of vehicles equipped with diesel engines. Therefore, together with the improvement of the economic performance of diesel engines, reducing the toxicity of their exhaust gases becomes a major problem.

The studies are aimed at studying the influence of biofuel on the ecological performance of the engine, as well as identifying the practical possibility of using the optimal composition as fuel for MAC. For this, the values of the concentration of pollutant emissions from the exhaust gases generated by the combustion of biofuels were measured. The purpose of this research was to:

- establishing the values of experimental tests on pollutant emissions for various loads and speeds of diesel engine operation;
- analysis of exhaust emissions (carbon monoxide (CO), carbon dioxide (CO₂), hydrocarbons (C_nH_m) and particulate emissions (fumigation)) in the operation of the engine with fuels tested.

When carrying out the experimental tests in laboratory conditions (on stand), the gas analyser of the Cartec type CET 2000 series was used, in order to determine the concentration of the polluting emissions in the exhaust gases. As fuels, diesel was used, the mixtures B20 (diesel 80% + biodiesel 20%), B50 (diesel 50% + biodiesel 50%) and B100 (biodiesel 100%) transesterified from rapeseed oil at the biofuel production plant M8- KPB-01 developed by S.A. „Alimentarmaş”, t. Chisinau.

Research results have shown that:

- The use of biodiesel as fuel for the DC4 11,0/12,5 engine allowed reducing the CO emission concentration by 25% at the engine operating mode at an average load of 70-75%.
- With the increase of the biodiesel concentration in diesel, the emission of C_nH_m in the exhaust gases decreases, which shows us that in the cylinder the fuel practically burns completely. Hence, it is mentioned that the use of B100 biodiesel reduces the concentration of C_nH_m emissions in the exhaust gases compared to diesel by 37,5%.
- With the increase of the biodiesel concentration in diesel, it leads to a decrease of the smoke emission concentration in the exhaust gas of the engine DC4 11,0/12,5, which significantly improves the ecological parameters.

Experimental on-site research into the concentrations of pollutant emissions from the exhaust gases of the DC4 11,0/12,5 engine powered by B100 biodiesel and a mixture of biodiesel and diesel (B20, B50) reveals that the results obtained are dependent on complex, contradictory. The establishment of the best performances of the concentrations of harmful emissions in the exhaust gases is achieved by optimizing the composition of such biofuels.

Keywords: alternative sources, harmful emissions, biofuels, engine powered, biodiesel, diesel.

EVALUAREA TENDINȚE ÎN MODIFICAREA STRUCTURII PEISAGISTICE PE EXEMPLUL BAZINULUI HIDROGRAFIC CUBOLTA

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Structura peisagistică caracteristică teritoriului Republicii Moldova a fost puternic influențată de presiunea tehnogenă asupra terenurilor. Documentele de politici de dezvoltare durabilă la nivel național au apărut pe ordinea de zi practic pe parcursul ultimilor două decenii, deși elementul de vulnerabilitate a teritoriului și capacitate de adaptare a acestuia a devenit indispensabil în ultimii ani ai perioadei indicate. Faptul indicat a fost determinat de dezvoltarea instrumentelor de analiză geospațială și apariția unor noi tehnici de evaluare și modelare în baza tehnologiilor informaționale geografice.

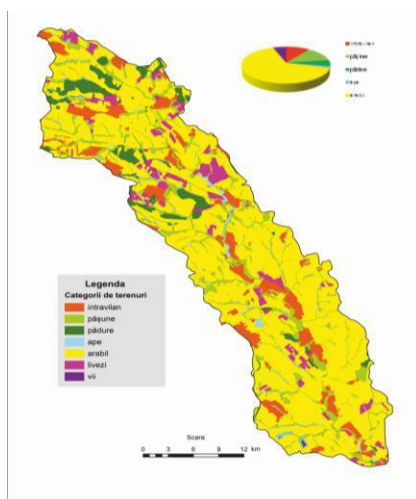


Figura 1. Model de utilizare a terenurilor din cadrul bazinului hidrografic Cubolta

Astfel, ca rezultat al interpretării imaginilor Landsat și aplicării clasificării în conformitate cu Sistemul FAO de Clasificare a acoperirii Terenului – Corine Land Cover Land Use a fost realizată harta privind modul de utilizare a terenurilor din cadrul bazinului hidrografic Cubolta. În rezultat se constată ponderea înaltă a terenurilor arabile (56464,5 ha) și frecvență mare a pășunilor (11266,7 ha). Metodologia de evaluare aplicată permite atribuirea bazinului hidrografic la terenuri cu ecologie instabilă (conform coeficientului de stabilitate ecologică), fiind evidentă capacitatea slăbită a teritoriului de absorbție a presiunii tehnogene.

Keywords: structura peisagistică, bazinul hidrografic Cubolta, sistemul FAO, ecologie, presiunii tehnogene.

IRIS VARIEGATA (IRIDACEAE) IN THE FLORA OF LANDSCAPE RESERVE "CĂRBUNA"

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It is recognized that biological diversity is a natural wealth that underlies the functioning of ecosystems, the provision of ecosystem services essential to human well-being and contributes to the economic development of society.

Iris is the largest genus in the Iridaceae family with up to 300 species. Eight species have been indicated for the flora of the Republic of Moldova.

In recent years, during the planned research within the landscape reservation "Cărbuna" and the adjacent territories, the protected by law in the republic species *Iris variegata* L. – Variegated Iris has been reported. It is a plant with a thick brown rhizome. The stem is usually branched at the top or in the middle, wrapped at the base with the long sheaths of the leaves. Two or three leaves dry out during flowering, and the green ones have a length of 20-30 cm, at the top being narrow and acute. Flowers more frequent in number of 6 on the flowering stem. The leaflets of the back are slightly swollen, acute green or obtuse. External perigonial lacinae rounded at the tips, barbellate, golden- yellow, with narrow, yellow-rimmed edges with dark lilac or light-lilac venation.

According to our investigation in the reserve during 2020-22, the Variegated Iris grows alone or in small groups in clearings or in White oak forest edges, in plots 12, 13, 17, 18 and 19, in association with *Chrysaspis aurea* (Poll.) Greene, *Coronaria coriacea* (Moench) Schischk. et Gorschk., *Cotinus coggygria* Scop., *Crataegus monogyna* Jacq., *Cruciata laevipes* Opiz, *Dianthus armeria* L., *D. carthusianorum* L., *Erysimum cuspidatum* (Bieb.) DC., *Falcaria vulgaris* Bernh., *Ferulago galbanifera* (Mill.) Koch, *Gagea paczoskii* (Zapal.) Ghrossh., *G. villosa* (Bieb.) Duby, *Galatella linosyris* (L.) Reichenb. fil., *Galium mollugo* L., *G. octonarium* (Klok.) Soo, *Hypochaeris maculata* L., *Inula conyza* DC., *Leopoldia comosa* (L.) Parl., *Scorzonera cana* (C.A.Mey.) O.Hoffm., *Silene bupleuroides* L., *Ranunculus illyricus* L., *Salvia nemorosa* L., *S. verticillata* L., *Stachys recta* L., *Teucrium chamaedrys* L. etc.

In this context, the populations identified in the "Cărbuna" Landscape Reserve were monitored, the coordinates were taken and the limiting factors were identified. As conservation measures, we propose to protect the growing sites of this species, monitoring existing populations and recording parameters for the study of their dynamics. The inclusion of this taxon in the Red Book of the Republic of Moldova (4th edition) will contribute significantly to the protection and further distribution of the species both *ex-situ* and *in-situ*.

Acknowledgments: The research was supported by the NARD through the Project "Research and conservation of vascular flora and macromycobiota of the Republic of Moldova", 20.80009.7007.22.

Keywords: *Iris variegata* L., "Cărbuna" Landscape Reserve, protect of species, monitoring, Red Book.

ADAPTATION OF THE METHOD FOR DETERMINING VITAMIN B₉ IN AQUEOUS SOLUTIONS

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Assessing the impact of natural waters pollution with chemical compounds is a complex and difficult process, especially at the stage of quantitative determination of pollutants in water composition, which are complex and multiphase systems. From a chemical point of view, pollution can be interpreted as the entrainment of the polluting compound in the processes of chemical self-purification and the shift of the dynamic equilibrium in the sense of the dominance of the reducing compounds. The pharmaceutical industry can become a dangerous source of natural waters pollution because pharmaceutical preparations contain biologically active substances. Once in the water, such substances not only upset the balance of self-purification processes, but can also show toxicity to hydrobionts.

Thus, in order to evaluate the intake of vitamin B₉, on the processes of chemical self-purification and the redox state of natural waters, the optimal conditions for modeling the systems in which vitamins have the role of potential pollutants were established. Vitamin B₉ is marketed as tablets, and 5 mg tablets were used in the study. Given that the modeled systems simulate the redox and photochemical processes of water self-purification, in which other substrates are present, such as hydrogen peroxide, transition metal ions (Cu, Fe), the selected spectrophotometric method must meet certain criteria, namely that other substrates must not interfere with the determination of the vitamin concentration. Secondly, it is necessary to select the optimal conditions for "preparation" of the vitamin solution from the pharmaceutical forms used, in order to obtain true results.

Vitamin B₉ in the form of tablets contains the following excipients: glucose and stearic acid. Therefore, the solution prepared in distilled water has turbidity. In order to eliminate the turbidity caused by stearic acid, 2 methods were applied: filtering through a simple paper filter and centrifugation ($V = 3500$ rpm, $t = 15$ min), and subsequently recorded the UV absorption spectra in the UV domain for comparison. Both procedures showed the same result, the absorption maximum wavelength for vitamin B₉ solution is 281 nm in both cases. The maximum absorption of vitamin B₉ solution allows its direct determination with the construction of the calibration curve $A = f(C)$, because this wavelength does not coincide with the absorption maximums of other substrates in the modeled systems. In order to demonstrate that the pH of the system does not influence the proposed spectrophotometric method, the UV absorption spectra were recorded at different pH values, respectively the wavelength of the absorption maximum is in all cases the same - 281 nm.

Further in the study of the legitimacy of the participation of vitamin B₉ in chemical self-purification processes of aquatic systems for the fast and effective quantitative determination, the spectrophotometric method will be used, based on the own absorption of the substrate in the UV spectrum.

Keywords: vitamin B₉, chemical self-purification, redox state, natural waters, photochemical processes.

OPEN SCIENCE PRACTICE USED IN STRENGTHENING THE NATIONAL COLLECTION OF NON-PATHOGENIC MICROORGANISMS

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Microbiology is a field of biological science that studies microorganisms, most of interest and important are includes in collections for maintenance and distribution. But the requirements to the collections of microorganisms as a reliable and informationally associated biological material are increase. National Collection of Non-Pathogenic Microorganisms (CNMN) functions at the Institute of Microbiology and Biotechnology and present an important depository of scientific and industrially valuable strains of microorganisms (fungi, yeasts, actinomycetes, bacteria, cyanobacteria, microalgae) in the Republic of Moldova. The digitalization, processing and standardization of CNMN scientific data facilitates their sharing and reuse between science and industry fields.

Research aim is to strengthen the National Collection of Non-Pathogenic Microorganisms by adopting open science practices on access, preservation, sharing and reuse of scientific data. The objectives:

- Conceptualization of CNMN according to the FAIR principles for scientific data (Findability, Accessibility, Interoperability, and Reuse of digital assets)
- Digitization, processing and standardization of Collection scientific data
- The development of CNMN digital tools (using Agile principles)
- Facilitating the integration of CNMN into the pan-European digital infrastructure MIRRI (Microbial Resource Research Infrastructure)

Open science is a policy priority for the European Commission (EC) and the standard method of working under its research and innovation funding programmes as it improves the quality, efficiency and responsiveness of research. It enables to store, curate and share research knowledge between partners from across academia, industry, public authorities, citizen groups and countries [1].

As a result of the project implementation, digital tools of National Collection of Non- Pathogenic Microorganisms information system will be developed, respecting the Open Science practices regarding the management of scientific data. This would enable preparing and submitting international projects related to microbial biodiversity and the application of microorganisms in various branches of the economy.

Acknowledgments: The research was funded out within the project 21.70086.38\$D (ANCD)

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Keywords: non-pathogenic microorganisms, national collection, conceptualization of CNMN, digital tools, information system.

BACTERIAL VIABILITY AFTER 15 YEARS STORAGE

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Lyophilization, or freeze-drying, is a process is a widely applied effective method for the preservation of microorganisms. The National Collection of Nonpathogenic Microorganisms (NCNM) contains *Pseudomonas* sp., *Bacillus* sp. which are known for its antimicrobial activity, also lactic acid bacteria strains with important biotechnological properties for the dairy industry and prospective for starter cultures composition. It conservation and preservation for long-term storage present the purpose for the collection.

The aim of the research was to check the viability and stability of pure strains *Bacillus* sp. CNMN-BB-01, CNMN-BB-02, *Pseudomonas* sp. CNMN-PFB-01, CNMN-PsB-03 and lactic acid bacteria strains after 15 years storage in NCNM. Lactic acid bacteria pure cultures used in this study comprised *Lactococcus* sp. from CNMN-01 to CNMN-14 strains, *Streptococcus thermophilus* CNMN-15 and CNMN-16 strains.

The number of viable cells was determined as colony forming units per ml (CFU/ml). The survival rate was calculated as CFU/ml after freeze-drying divided by CFU/ml before freeze-drying. Therefore, the results of this work show that *Bacillus* sp. and *Pseudomonas* sp. strains are viable and their titer was ranged from 6,8 to 7,6 $\log_{10}\text{UFCml}^{-1}$ for *Bacillus* and from 7,9 to 8,1 $\log_{10}\text{UFCml}^{-1}$ for *Pseudomonas*. It is known that *Pseudomonas* and *Bacillus* bacteria can be stored for over 30 years in freeze-dried form with no changes of high level cell viability at 6-7 $\log_{10}\text{UFCml}^{-1}$.

Similar research results on lactic acid bacteria strains after 15 years of storage in freeze-dried form demonstrated viability more than 80% with titer ranged from 6,2 to 8,3 $\log_{10}\text{UFCml}^{-1}$. According to other studies viability of species *Streptococcus*, *Staphylococcus*, *Brevibacterium*, *Pseudomonas*, *Corynebacterium*, *Lactobacillus*, *Salmonella*, *Bacillus* after freeze-drying amount to min. 70%. Thus, the numbers of viable cells remaining in the ampoules are sufficient to maintain the culture.

Microscopic smear appearance of strains investigated confirmed the purity of cultures, cell cultures represented rod-shaped cells typical of Gram-positive *Bacillus* sp. and Gram-negative *Pseudomonas* sp. Lactic acid bacteria cells present cocci and diplococci in the medium and long chains. Morphological and cultural properties correspond to the technological requirements for lactic acid bacteria species. All strains were able to active milk coagulation, formed firm, gel dense consistency, without eliminating of whey, without gas eruption.

Freeze-drying provides higher cell viability, is used for the long-term preservation, also it plays a fundamental role in scientific and practical fields. The obtained results confirm effectiveness of freeze-drying as a method of conservation through high ability to strains regeneration.

Acknowledgments: The research was funded out within the project 20.80009.7007.09 (ANCD).

Keywords: bacterial viability, lyophilization, national collection, nonpathogenic microorganisms, long-term preservation.

THE RISK OF LATE SPRING FROSTS FOR AGRICULTURE OF THE REPUBLIC OF MOLDOVA

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In the group of weather-climatic phenomena unfavorable to agriculture, frost occupies an important place. They cause not only the slowdown of plant development and the premature end of the vegetation cycle, but even their partial or total death. On the territory of the Republic of Moldova, according to the data of the State Hydrometeorological Service analyzed in this study for a period of over 60 years, late spring frosts are generated in anticyclones and their ridges formed in the Arctic air masses, as well as in high pressure areas with insignificant barricade gradients, oriented from west to east. They can also occur as a result of the advection of cold air behind cyclones. Most often these frosts are advective-radiative. The mentioned frosts usually affect the layer in the immediate vicinity of the soil during the vegetation period of the plants, when the average daily air temperature remains positive.

In spring, frosts pose a great danger to agricultural crops, which are signaled after the stable passage of the average daily air temperature through 10°C in the direction of its increase (April 16-23). Spring frosts on average remain on the territory of Moldova until April 5-21, at the ground surface until April 22 - May 6. But the average dates of frost disappearance depend, to a large extent, on local conditions (fragmentation and height of landforms), sometimes reaching variations of 20-30 days. The latest date of frosts in the air in the north and center of the republic was reported on May 21-24 (1980), and in the south of the republic during the period 1-10 May (1990). Frosts are possible at the surface until June 1 (1955). In 1977 such a phenomenon took place between May 22-28.

Late spring frosts cause considerable damage to fruit crops during flowering. The probability of frost damage to apricot flowers and fruits is on average 15-40%, for other fruit crops - up to 15%. A special danger for the vine is the late spring frosts after the buds open. The most sensitive to frost are vegetable crops - such as tomatoes, peppers, eggplant and others are the most demanding to heat. Frosts with an intensity of 0-1°C cold lead to their extinction. The probability of frost damage to sprouted corn and sunflower plants is relatively low.

In practical interest, the risk interval for frost has been established, when the respective phenomena are the most dangerous, in order to avoid some of their serious consequences. The risk interval represents the interval between the average and extreme date of frost production. The risk range varies depending on the intensity of the genetic factors of the frost, as well as the local conditions, both as production time and as a place of manifestation. All methods used to reduce the consequences and control frost (fumigation, shelter, plant protection curtains, sprinkler and micro-sprinkler irrigation, air ventilation, hot air heating) must contribute to the reduction of radiative cooling, destruction of the thermal inversion layer from soil, homogenization of the air temperature in the microclimatic layer and consequently, maintenance of the air temperature and on the soil surface higher than 0°C.

Acknowledgments: The study was supported by the project 20.80009.5107.01 (NARD).

Keywords: frosts, agriculture, damage, crops, risk.

ON THE MECHANISM OF CHEMICAL SELF-PURIFICATION OF VARIOUS WATER SYSTEMS OF THE LOWER DNIESTER BASIN

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Identification of the participation of iron and copper in redox processes that occur in natural aquatic ecosystems allows us to make an assumption about the presence or absence of free radicals OH, intensively oxidizing organic substances of autochthonous and allochthonic origin, which is the essence of the mechanism of radical chemical self-purification. The presence of such a self-purification channel along with biological, can ensure the ecological prosperity of aquatic ecosystems and a high level of biodiversity of flora and fauna [1].

To identify the presence of a chemical self-purification channel in aquatic ecosystems of the Lower Dniester basin (small rivers, Dniester, reservoirs), a correlation and regression research method was used based on a database, including concentrations of various forms of metal migration and quality indicators of natural water.

The study carried out suggests that the oxidation of dissolved organic matter in the small rivers Ichel and Raut takes place by an ion-molecular mechanism, without the formation of free radicals. The migration of the studied metals is dominated by mineral forms, the dynamics of migration is of a pronounced seasonal nature.

In the segment of the Dniester from Dubasari to Vadul-lui-Voda, a change in the nature of the forms of migration of metals from mineral-organic to organic has been revealed. At the Vadul-lui-Voda cross-section, considerable involvement of copper in the processes of chemical self-purification was established. Iron, apparently, accumulates in bottom sediments and does not play a decisive role in the intensification of the radical processes of chemical self-purification in the studied segment of the Dniester.

The priority role of iron in the intensification of radical processes of chemical self-purification has been identified for the aquatic ecosystems of the reservoirs Ghidighici and Danceni. Copper, apparently, as a necessary trace element is accumulated by intensively developing biota of reservoirs.

As a result of the study, it is possible to conclude about irreversible problems with the ecological prosperity of the waters of Ichel and Raut, on the establishment of the Dniester cross-sections, where chemical self-purification processes are not intensive and about the greatest contribution of iron to the processes of chemical self-purification of reservoirs.

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Keywords: aquatic ecosystems, chemical self-purification, Lower Dniester basin, redox processes, biodiversity of flora and fauna, metal migration, quality indicators.

SPATIAL MODELING OF DANGEROUS FROSTS IN THE REPUBLIC OF MOLDOVA

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For the agriculture of the Republic of Moldova, the late spring and early autumn frosts present a great danger, because they can harm agricultural crops in the early stages of development or towards its end, thus creating frost damage, sometimes quite serious, depending on their resistance to frost. Being located in the southeast of the continent, the territory of the republic is at the cross-road of several trajectories of the air masses movement that ultimately cause such phenomena as frost.

The possibilities of interconnecting the modules of GIS software, of analyzing the data obtained from field measurements and of organizing these data in structures such as meteorological models, lead to obtaining good results compared to the classical methodology.

For the spatial distribution of dangerous frosts, we used the Regional Geographic Information Systems. The date of manifestation of the first and last frost was interpolated, from 18 meteorological stations for the period 2005 - 2020.

The numerical model was obtained based on the regression equation (1, 2), without using the constant in the model, when performing the calculations, we used the number of calendar days (Julian date calendar).

The regression equation based on which the spring frosts' spatial distribution was modeled is: $Y=0,0505557*exp-0,0327863*habs-0,0305571*hrel+0,89103*panta-0,00000496262*x+0,00002212*y$ (1), where *exp* - slope's exposition, *habs* - absolute altitude, *hrel* - relative altitude, *panta* - the degree of inclination of the slopes, *x*, *y* - *x* and *y* coordinate respectively. Latitude *X* and longitude *Y* both are expressed in meters in WGS84 Transverse Mercator projection with 27° central meridian and false easting 500000 m.

The regression equation for the autumn frosts' spatial distribution is: $Y=0,0281752*exp+0,045071*habs+0,0166217*hrel-0,639479*panta+0,000070738*x+0,0000461509*y$ (2), where *exp* - slope's exposition, *habs* - absolute altitude, *hrel* - relative altitude, *panta* - the degree of inclination of the slopes, *x*, *y* - *x* and *y* coordinate respectively. Latitude *X* and longitude *Y* both are expressed in meters in WGS84 Transverse Mercator projection with 27° central meridian and false easting 500000 m.

As a result of the calculations performed, we conclude the following:

1. Dangerous frosts occur every year and have a spatial distribution throughout the study area.
2. A more in-depth study is required to be carried out for late spring frosts because both their duration and intensity threaten agricultural crops. For example, those registered in Soroca and Camenca on May 10, 2017.
3. The date of manifestation of early frosts as well as of late frosts has a negative impact on the vegetation, especially on agricultural crops, if they are manifested during the important phenological phases, causing economical damage to agriculture sector.

Keywords: agricultural crops, damage, frosts spatial distribution, meteorological models.

**CONTRIBUTIONS TO THE KNOWLEDGE OF LEAF BEETLES
(CHRYSOMELIDAE) FROM ALFALFA**

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The leaf beetles (*Coleoptera*, *Chrysomelidae*) family is one of the biggest of the order Coleoptera. The practical importance of the leaf beetles in the human activity draws the attention of the researchers permanently.

The faunistic materials were sampled in the alfalfa culture from Horești, Nișcani, Piatra Albă, Telița, Svetlii, Vulcănești, Vadul lui Isac, Călinești, Trifești and Saharna during May and June 2022. Most of the investigated fields were adjacent to a forest sector. The entomological net was used for collection. Some specimens were also gathered by hand from the leaves or soil surfaces.

A total number of 10 leaf beetles species belonging to 9 de genera and 5 subfamilies were identified all over the studied fields: *Cassida denticollis* Suffrian, 1844, *C. rubiginosa* Müller, 1776, *Chrysolina marginata* (Linnaeus, 1767), *Entomoscelis adonidis* (Pallas, 1771), *Galeruca tanacetii* (Linnaeus, 1758), *Gastrophysa polygoni* (Linnaeus, 1758), *Gonioctena fornicata* (Brüggeman, 1873), *Oulema melanopus* (Linnaeus, 1758), *Pachybrachis fimbriolatus* (Suffrian, 1848) and *Plagioderma versicolora* (Laicharting, 1781).

The species *Gonioctena fornicata*, a known pest of alfalfa was represented in all of studied fields. The lucerne leaf beetle usually feed more intensively at the stage of larva, that's why the larvae cause more damage than adult insects. Often the feeding on green parts of the plant leads to the worsening of the physiological condition of the plant attacked, and consequently to the decrease of harvest or to the plant growth decrease.

Considering that other herbaceous plant species can be found in the alfalfa fields, this explains the variety of leaf beetles collected: *Cassida denticollis*, *C. rubiginosa*, *Chrysolina marginata*, *Entomoscelis adonidis*, *Galeruca tanacetii*, *Gastrophysa polygoni*, *Oulema melanopus* and *Pachybrachis fimbriolatus*. The presence of *Plagioderma versicolora* is accidental because it feeds exclusively on leaves and pollen of willow and poplar trees and can be explained by the existence of forest next to the field.

The investigations will continue throughout the growing season of alfalfa and the list can be supplemented with other leaf beetles species.

Acknowledgments: The study was performed under the project 20.80009.7007.02 „Evolutionary changes of economically important terrestrial fauna, of rare and protected species under anthropogenic and climatic changes”.

Keywords: *Chrysomelidae*, alfalfa, decrease of harvest, leaf beetles species, variety.

**INERSPECIFIC BEHAVIOUR BETWEEN *APODEMUS FLAVICOLLIS*
AND *APODEMUS SYLVATICUS* FEMALES FROM URBAN ECOSYSTEMS
OF CHIȘINĂU CITY, REPUBLIC OF MOLDOVA**

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Recently many studies have described the effects of urbanization on species behavior and adaptation. The species *Apodemus uralensis* and *A. sylvaticus* were captured with live traps in urban ecosystems of Chișinău city. The study of the interspecific relations between the two species were carried out by the method of pair encounters.

In interspecific relations of *A. uralensis* and *A. sylvaticus* various types of olfactory contacts were established: naso-nasal, naso-lateral, naso-dorsal, naso-ventral. In the interspecific contacts of the females, aggressive elements were registered, but in small numbers. *A. sylvaticus* females explored the environment (36.9% of the experiment duration) and actively tried to familiarize themselves with the partner – the number of contact initiations increased 2 times compared to intraspecific contacts and was 2.4 times higher than the analogous index of females *A. uralensis*. In *A. uralensis* females the complex of defensive behavior increased considerably: practically at each contact initiation, they responded through aggressive postures or shrill sounds. The duration of the freezing was 2 times longer than that of the exploratory activity, while the last one decreased by 1.3 times. Compared to intraspecific contacts, the noise level increased 3.7 times and the number of aggressive postures - 5.2 times.

According to the obtained data, the reciprocal olfactory contacts (naso-nasal, naso-anal, naso-lateral) have a great importance for the social relations of rodents and depend on the social status, season and sex of the partners. When the specimens of different species are placed in pairs, the olfactory contacts last 10 - 40 sec. The number of olfactory contacts is maximum in the first 5 minutes of the experiment, during the establishment, the formation of mutual relations between animals.

All the listed characteristics of representatives of *A. sylvaticus* allow us to conclude that this species has high adaptive abilities, great potential and resistance to overcome the negative conditions of “tension” community.

Acknowledgments: The study was carried out within State Program project 20.80009.7007.02.

Keywords: *Apodemus sylvaticus*, *Apodemus uralensis*, adaptation, behavior, intraspecific contacts, urban ecosystems.

CLIMATIC AND HYDROMETEOROLOGICAL PHENOMENA OF RISK ON THE TERRITORY OF THE REPUBLIC OF MOLDOVA

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The paleoclimate of the planet Earth demonstrates the cyclicity and evolution of the climate for certain geological periods of the Earth manifested in time and space, where flora and fauna have confronted and adapted to certain climatic conditions and hydrometeorological phenomena.

Weather and climate have been and are the basic elements of the planet Earth that condition and influence the daily existence and evolution of both man and the environment.

The weather and climate parameters monitored for a period of more than 30 years have become climatic norms, and serve as benchmarks both for anthropogenic activities and for the state and evolution of the environment.

But deviations from the climate norm are classified as climatic and hydrometeorological risk phenomena, so the greater the gap from the climate norm, the greater the intensity and damage caused by hydrometeorological risk phenomena, sometimes reaching catastrophic levels.

The territory of the Republic of Moldova in the context of climate change is increasingly affected by risky climatic and hydrometeorological phenomena, such as: storms, heat waves, cold waves, gusts of wind, storms, torrential rains, floods, hail, droughts, late frosts. In this article I made the statistical analysis of the evolution and of the damages produced as a result of the production of the climatic and hydrometeorological phenomena of risk from the period 2007-2021.

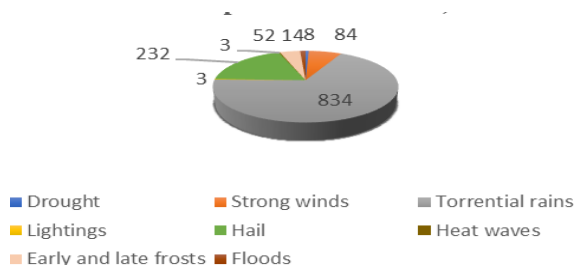


Fig.1 Number of climatic and hydro meteorological phenomena of risk (2007-2021)

The material damages caused by the climatic and hydrometeorological phenomena of risk have a direct impact on the socio-economic development of the country. World experience has shown that the most vulnerable countries to climate and hydrometeorological risk are poorly developed or developing countries such as the Republic of Moldova, because they do not have the financial support for national programs to adapt to climate change.

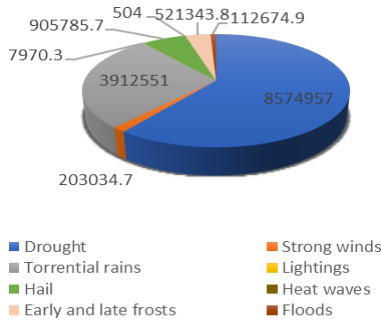


Fig. 2. Material damages caused by climatic and hydrometeorological phenomena of risk (2007-2021)

At the same time, the statistical analysis of the data provided by the General Inspectorate for Emergency Situations showed which of the climatic and hydrometeorological phenomena most often have deviations from the climatic norm and consequently cause major material losses and also what is their tendency.

Keywords: climate change, climate norm, climate parameters, risk phenomena, material damages.

PARASITE FAUNA IN PYGMY FIELD MOUSE FROM VARIOUS BIOTOPES OF THE REPUBLIC OF MOLDOVA

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The pygmy field mouse (*Apodemus uralensis*, Pallas, 1771) inhabits the forest edge, forest shelter belts and the open type biotopes: meadows, pastures, agrocenoses, fallow ground. It is well adapted to more arid conditions in comparison to other *Apodemus* species, therefore it is widely spread over the republic territory. It is a species with a lower frequency compared to other small rodents and has accessory ecological significance. The rodents are the basic trophic component of carnivorous mammals and prey birds, being the hosts (intermediate, definitive) that contain various invasive forms of a large range of parasitic species specific to other animals and humans.

The aim of the paper was the ecoparasitological study of *A. uralensis*, in order to establish the structural diversity of parasite fauna from various biotopes of the Republic of Moldova.

The results of parasitological investigations showed a prevalence of *Catenotaenia cricetorum* of 5.0%, respectively of *Hydatigera taeniaeformis larvae* – 10.0%, *Taenia pisiformis larvae* – 10.0%, *Rodentolepis straminea* – 5.0%, *Paranoplocephala omphaloides* of 10.0%, *Skrjabinotaenia lobata* – 10.0%, *Syphacia obvelata* – 20.0%, *Syphacia stroma* – 5.0%, *Capillaria hepatica* – 15.0%, *Heligmosomoides polygirus* – 5.0%, *Mastophorus muris* – 20.0%, *Trichocephalus muris* – 15.0% and invasion with *Strongyloides ratti* – 15.0%.

The taxonomic structure consists of 3 classes, 10 families, 12 genera and 13 species, of which 6 parasitic species belong to the Cestoda class (*T. pisiformis larvae*, *S. lobata*, *H. taeniaeformis*, *C. cricetorum*, *R. straminea*, *P. omphaloides*), with a share of 46.1% of the species, 5 species - to the Secernentea class (*S. obvelata*, *S. stroma*, *H. polygyrus*, *M. muris*, *S. ratti*) with a share of 38.5%, and 2 species - to the Adenophorea class (*T. muris*, *C. hepatica*), constituting 15.4% of the total identified species.

The data obtained prove the potential of the parasitic pollution risk of the interfering area between natural and anthropized ecosystems and as a result the transmission of invasive forms from wild animals to domestic animals and to humans. At the same time, the rodents as component of the trophic chain are vectors of invasive forms in the environment and ensure the functional stability of the host-parasitic systems within the investigated biocenoses.

Acknowledgments: The studies were performed within the State Program projects 20.80009.7007.12 „Diversity of hematophagous arthropods, zoo- and phytohelminths, vulnerability, strategies for tolerating climatic factors and elaboration of innovative procedures for integrated control of socio-economic interest species” and 20.80009.7007.02 „Evolutive changes of economically important terrestrial fauna, of rare and protected species in the conditions of anthropic and climatic changes”.

Keywords: *Apodemus uralensis*, host-parasitic systems, parasite fauna, parasitic pollution risk.

**CONTRIBUTION TO THE STUDY OF *FAGOPYRUM ESCULENTUM*
MOENCH IN THE REPUBLIC OF MOLDOVA**

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The flora of the Republic of Moldova is rich in species of honey plants, with high productivity of honey, plants that provide food for insects throughout the year, except for the cold season. In the collection of Honey and Forage Plants of the "Alexandru Ciubotaru" National Botanical Garden (Institute) (NBGI), there is a wide range of valuable plants, with multiple potential uses, in particular – as honey crops. Among them, there are species that start vegetating in early spring and start blooming in early May. The study of flowering dynamics allows highlighting the generative stages of plants and the most active periods when honey insects collect nectar and pollen. *Fagopyrum esculentum* Moench (family Polygonaceae Juss.), common buckwheat, is a valuable herbaceous, annual plant, with edible seeds, included in the group of pseudocereals. It is also cultivated as a honey plant. Depending on the weather conditions, in 12-14 days after sowing, the cotyledons of the seedlings emerge on the soil surface. At this moment, the vegetative phase begins, consisting of the seedling, immature and virginal stages, characterized by the active growth of the vegetative organs of plants (stems, lateral branches and leaves), which lasts about 20-25 days from the emergence of seedlings. Buckwheat plants can reach up to 125 cm in height, the stem is erect and branched. Usually, there are 6-12 leaves on each branch. The generative phase begins with the development of floral buds. Flowering is long, staggered and lasts about 33-48 days. In the full flowering stage, on a medium-sized shoot (85 cm), with 3-4 branches, there are 22-24 inflorescences at different stages of development. On one inflorescence, at the same time, there can be flower buds, flowers and developing fruits. The number of flowers on a shoot varies between 375 and 400, and the fruits – about 92-125. A buckwheat plant can produce around 1-1.5 thousand flowers [1]. The flowers are attractive to pollinating insects, as a source of nectar and pollen, especially under conditions of high temperatures and humidity. A flower lives for about 24 hours and produces up to 0.1 mg of sugar [2]. Pollinating insects are more active on buckwheat flowers until 11.00 on days with high temperatures. Common buckwheat, *Fagopyrum esculentum* Moench, can be sown as a primary crop or as a successive crop, the potential honey productivity reaches values of 70-90 kg honey/ha [1]; the dehulled seeds contain 12.6% protein, large amounts of threonine and lysine, vitamin E, folic acid. The identification, mobilization and acclimatization of new high-potential honey plants, will contribute to the expansion of the range of honey crops and ensure a continuous source of pollen and nectar, very important for the health and well-being of honeybee families.

Acknowledgments: The study has been carried out in the framework of the project: 20.80009.5107.02 “Mobilization of plant genetic resources, plant breeding and use as forage, melliferous and energy crops in bioeconomy”.

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Keywords: *Fagopyrum esculentum*, flowering, honey crops, honey productivity.

CYANOBACTERIUM *NOSTOC LINCKIA* GROWTH UNDER DIFFERENT CONCENTRATIONS OF COPPER(II) IONS

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Cyanobacteria are ecologically significant prokaryotes that can be found in heavy metals contaminated environments. In these situations, they can act as bioremediators of pollutants, especially heavy metal ions, by sequestering the metal ions on the surface of their cells due to the presence of negatively charged hydroxyl (OH), carboxyl (COOH), carbonyl (C=O), sulfhydryl (SH), and other functional groups that are available to bind with positively charged metal ions. In cyanobacteria, copper plays an essential role as a structural component of plastocyanin, which mediates the electron transport chain, and also an essential cofactor of enzyme superoxide dismutase. However, at higher concentrations (more than 3.0 mg/L), copper is reported to be toxic to microorganisms. Consequently, higher metal contents are transferred to higher trophic levels of the food chain. The permissible limit of Cu²⁺ in water is between 0.05 and 1.5 ppm, and has many health risks associated with the exposure to excess Cu²⁺ in humans. Abnormally high copper levels have been associated with a number of diseases, including neurodegenerative disorders such as Alzheimer's, Parkinson's, and prion diseases.

In this work, the effect of divalent copper ion (Cu²⁺) exposure was studied on cyanobacterium *Nostoc linckia* growth. A sulfate salt of copper, namely copper(II) sulfate pentahydrate, was used as a source of metallic ions. The copper concentrations chosen for the study were 0.1, 0.5, 1.0, 1.5, 2.0, 2.5, 5.0, 10 and 20 mg/L, which were added to the culture in the exponential growth phase of the life cycle. *Nostoc* growth was determined spectrophotometrically at the end of cultivation cycle, which lasted 12 days.

The data showed no significant decrease in the amount of biomass in the case of *Nostoc* growth under the lowest concentrations of copper ions. Hence, in metallic monosystems with 0.1, 0.5 and 1.0 mg/L copper ions, cyanobacterial biomass produced during cultivation cycle was about 1.0 g/L, which means a reduction of up to 15% compared to control. In the case of further metal concentrations such as 1.5, 2.0 and 2.5 mg/L, *Nostoc* growth has sharply decreased up to 32% in comparison with control. Then, the cultivation of cyanobacterial strain under conditions of higher concentrations of copper 5.0, 10.0 and 20.0 mg/L did not have a significant effect on the reduction of biomass production. At the same time, the data were comparable with the cultivation of the culture under conditions of low concentrations of copper ions. Thus, we can see a wave type effect of *Nostoc linckia* growth under various concentrations of metallic copper.

The release of polysaccharides by some cyanobacterial strains has been one of the mechanisms to overcome metal toxicity. The increase in the amount of exopolysaccharides synthesized by *Nostoc* in response to higher concentrations of copper and the protective role of these molecules may explain the wave type effect of *Nostoc linckia* exposure to different concentrations of copper.

Keywords: *Nostoc linckia*, bioremediators, concentrations of copper ions, cultivation cycle, food chain, health risks, heavy metal ions,

THE ROLE OF CLIMATE FACTORS IN THE PROCESS OF AGRICULTURAL PRODUCTION IN MOLDOVA

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The agricultural sector of the Republic of Moldova is most influenced by climate variability, given the dependence of agriculture on the evolution of weather on those periods of crop vegetation, as well as increasing the frequency, duration and intensity of weather and climate risks in the context of regional climate change. The effects of regional climate change are significant and expressed by changing the main environmental variables (air temperature and precipitation), the impact on agricultural plant development being increasingly evident.

The daily temperature regime is the one that imprints the biological evolution of the cultivated plants and offers a closer image of the reality of the temperature variations that can occur, sometimes reaching extreme values, which constitute climatic risks through negative effects. Thus, the persistence and continuity of positive daily average temperatures between April and September for the territory of the Republic of Moldova, indicate favorable thermal conditions for various annual or perennial crops. At the same time, tropical temperatures, above 30-35°C, impose restrictions on plant development, reducing agricultural production.

Atmospheric precipitation registers relatively low multiannual averages (470-620 mm), this being the main characteristic of the climate in the region. The results of our investigations note that the drought limiting factor that manifests itself on the largest agricultural area of the country, and in this context, data indicate that the most vulnerable agricultural areas are located in the south and center of the republic, except for partial land, located in the meadows of large rivers.

According to the analyzed data, in the last two decades there have been several years with strong and very strong droughts (2003, 2007, 2009, 2012, 2015, 2020), which have led to the decrease of fruit in most agricultural crops, causing significant direct losses, estimated at about \$ 1.0-1.25 billion each. Also, analysis of the results obtained on road losses to the main agricultural crops, it was found that in all the dry years of the analyzed decades, sunflower recorded the lowest losses compared to other field crops, due to ecological plasticity and potential, greater adaptation of sunflower to regional climate change.

It is very important that the knowledge and identification of meteorological risks with negative effects on crop stability be based on both early warning systems on dangerous meteorological phenomena and on technological practices applied in relation to the current evolution of climatic conditions and foreseeable future scenarios. Thus, possibilities to adapt to the effects of climate change aim to reduce the impact on the agricultural production process and are mainly based on optimizing the duration of the vegetation period of crops, resistance of genotypes to extreme temperatures (heat, cold / frost), deficits / excesses water in the soil and increase their risk of phytopathogens, ecological plasticity, tolerance to the effects of extreme weather events.

Keywords: agriculture, climate variability, climate risks, droughts, vulnerable areas.

MICROBIOLOGICAL TOOLS FOR ASSESSING IMPACTS ON SOIL ORGANIC MATTER CONTENT

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Soil organic matter (SOM) is fundamental to soil quality and has been suggested as the single most important indicator of soil quality and productivity. For any given soil and climate, the amount of SOM is determined by land use and soil management, but the usefulness of SOM data for soil quality monitoring is constrained by the difficulty of experimentally verifying changes over short periods of time. Long-term experiments seem to offer only a partial solution to this problem.

Soil microorganisms accounts for only 1-3 per cent of total soil organic carbon, but they play a key role in the soil forming processes, including in SOM transformations.

Significant correlations between SOM content and soil microbial parameters (soil microbial biomass, and/or basal respiration, and/or metabolic quotient) were observed in the moderately and poorly humified Typical chernozems from the Moldovan long-term field experiments that included 6 traditional (10-field) and 5 ecological (7-field) crop rotations (with and without alfalfa, mineral fertilizers and/or farmyard manure), continuous black fallow, and 5 continuous crops (with and without mineral fertilizers with farmyard manure). These correlations open practical possibilities of using soil microbial parameters as a tool for timely SOM related assessments and predictions. Once a new soil management practice is introduced, the substantial difference in the soil microbial biomass (SMB) and SOM turnover rates will allow the soil microbial parameters to reach a new equilibrium (reflecting the peculiarities of this management) much sooner than the SOM content. As soon as that equilibrium of the microbial parameters is established, the future SOM content becomes predictable/calculable from the established correlational relationships, assuming that given enough time SOM will tend to fit the same correlational relationship that was observed in the long-term field experiments. These predictions may be beneficial in such important fields as protection and enhancement of soil quality and biodiversity, carbon sequestration, development/assessment of sustainable soil management practices, and others. The implementation of the possibility may provide farmers with better opportunities for investing into soil quality/biodiversity and may contribute to solving problems related to the climate change and others.

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Keywords: soil organic matter, soil quality, sustainable soil management.

STORAGE POTENTIAL OF TRITICALE ACCESSIONS - INDICATOR OF THEIR VIABILITY UNDER *EX SITU* CONSERVATION

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The purpose of this research was to study the morphophysiological and biochemical parameters of tritcale seeds after accelerated aging (AA) test. The storage potential (SP) of collection accessions in active collections of the gene bank was characterized with the purpose of their *ex situ* conservation, and to reveal genotypic differences of genotypes. According to the plan, tritcale seeds genotypes should be grouped by their potential capability to preserve viability of samples exposed to high temperature and humidity.

Objects of studies were collection accessions of tritcale seeds from active collection of the plant gene bank. SP of seeds was determined by two tests: test for accelerated aging of seeds by Safina & Filipenko method (2013) and test for electrical conductivity of solutions (EC), both are included in the International Rules for Seed Testing (ISTA). AA-test of seeds was conducted under the increased air temperature (37°C), humidity of seeds 13%, high relative humidity (98%), aging time was 30 days. During this test various morphophysiological parameters of seeds and seedlings were measured every week (dynamic measurement) according to the ISTA: germinating power (GP) and germinability (G) of seeds, radicle length (RL), fresh and dry biomass of radicles. EC of solutions with normal and aged seeds was measured by conductometer N5721M. With regard to biochemical parameters, the activity of the peroxidase enzyme (PO) was measured in radicles of seedlings. The experiment was conducted in quadruplicate. Aged seeds were sprouted in Petri dishes in thermostat at 25°C. Experimental data were processed using the software *Statistica 7*. According to the set of different parameters of tritcale seeds after the AA-test of seeds, the following genotypes had the highest SP: *Ingen 3*, *Ingen 33*, *Ingen 35*, *Ingen 40*, *Ingen 54*, *Ingen 93* (loss of seed germination after AA-test by 35%) and the lowest storage potential was found in *Ingen 2* and *Ingen 4* (loss of seed germination by 70%). The smallest drop of other parameters was in genotypes with high SP (raw root biomass-by 100 mg, root length-by 2 mm). Conclusions: 1. Use of AA-test of seeds and determination of their different parameters allowed assessment of SP of tritcale collection accessions from the active collection of the gene bank. 2. It was demonstrated that EC of solutions with aged seeds of the most genotypes of tritcale is higher than that of solutions with normal seeds. 3. Significant positive correlation of germinability of tritcale seeds with germinating power, radicle length, fresh and dry biomass of seedlings was revealed after conduction of test for AA of seeds.

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Keywords: accelerated aging test, *ex situ* conservation, morphophysiological parameters of seeds, tritcale.

THE IMPACT OF PRECIPITATION ON THE SUNFLOWER CROP IN THE NORTHERN REGION OF THE REPUBLIC OF MOLDOVA

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In the conditions of the continental climate with excessive nuances, characterized by a great variability in time of the climatic elements, specific to the territory of the Republic of Moldova, the amount of precipitation is one of the most important factors that determine the optimal growth and development of agricultural crops, including sunflower, determining the greatest differentiations in time and space of the average harvest value per hectare. Thus, in the conditions of the Republic of Moldova, the amount of precipitation, most of the times is rather a limiting factor.

Knowing the amount of atmospheric precipitation during the growing season, forecasting assessments of the productivity value can be carried out. The results obtained by Cojocari R., reveal that they vary on the territory of the country between the limits of 340-450 mm. Starting from these values, we find that a more favorable situation is outlined during the vegetation period of the sunflower, when, especially in the northern part of the region, there are also some quantitative surpluses of precipitation compared to the optimal required. Thus, the isohieta with the value of 375 mm – the optimal quantity for the period April- October – crosses the districts of Riscani, Edinet, Donduseni and Ocnita. Within the region, based on the data from the period 2007-2020, it appears that the most favorable districts for the cultivation of sunflower are Glodeni, Ocnita and Edinet. The districts with the lowest performances in the development of the sunflower culture are Singerei, Floresti and Soroca.

In sunflowers, although rainfall amounts in the cold half of the year correspond as a spatial distribution with the average fruit distribution, in terms of quantity, they are insufficient for the entire region. Instead, the precipitation amounts from the growing season allow to highlight the north of the country where the average values of the period 2007-2020 are within the optimal needs.

The use of modern agrotechnical processes that help to effectively use the moisture accumulated in the soil. The mechanized work of the soil reduces the use of moisture by evaporation, improves the aeration of the soil. But the use of the appropriate agricultural technique and the application of the correct methods of rotation are possible only on large agricultural areas. So, it would require a consolidation of agricultural land.

Acknowledgments: This study was funded by the project of the State Program 20.80009.5107.01 - Genetico-molecular and biotechnological studies of the sunflower in the context of sustainable management of agricultural ecosystems.

Keywords: agricultural technique, amount of precipitation, isohieta, limiting factor, sunflower, spatial distribution.

ECOTOURISM POTENTIAL OF BIODIVERSITY FROM CRIULENI DISTRICT, REPUBLIC OF MOLDOVA

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The biodiversity of the living world, through the richness of floristic and faunal species, has always been the points of tourist attraction. A special interest for tourists who practice ecological tourism, presents the protected natural areas of this district. Thus, according to the field studies and in accordance with the Law on the fund of natural areas protected by the state no. 1538-XIII of 25.02.98, in the Criuleni district, the following protected natural areas of interest for the practice of ecological tourism are: **I. Monuments of nature (A. Geological and paleontological: Fossil soils on the Transnistrian terraces; Goian outcrop; Surprises cave** (The fauna of the cave is made up of several species of bats (mainly of the *Myotis* and *Rhinolophus* genera), but also terrestrial invertebrates such as pseudoscorpions, arachnids, myriapods, terrestrial crustaceans et al. There are also several species of flora in the cave. Saprophytic fungi grow here (including a species of phosphorescent mushrooms); **B. Hydrological: The mineral springs from Onitcani village** (The protection zone is sprinkled with clumps of trees, the adjacent land being covered by forest plantations); **C. Botanical - Representative sectors with forest vegetation: Poplar forest** (from trees comprises 94 specimens of *Populus alba*, 2 of *Ulmus laevis* and 1 of *Pyrus pyraeaster*, from shrubs are present *Prunus spinosa*, *Swida sanguinea*, *Crataegus monogyna* and *Rosa canina*); **Pogoreloe** (15 species of trees were identified – *Acer campestre*, *Acer negundo*, *Acer platanoides*, *Acer tataricum*, *Carpinus betulus*, *Cerasus avium*, *Fraxinus excelsior*, *Morus alba*, *Populus alba*, *Pyrus pyraeaster*, *Quercus robur*, *Salix alba*, *Tilia cordata* et al.; 12 species of shrubs including *Corylus avellana*, *Crataegus monogyna*, *Euonymus europaea* et al.); **Secular trees - 3 in number**. According to Annex no. 3 of the Law on the fund of natural areas protected by the state, in Criuleni district there are 3 secular trees - 2 pedunculate oaks and a poplar from Canada. **II. Forest nature reserves: Dubăsari, Zoloceni, III. Monuments of landscape architecture: The park from Bălăbănești village** (from trees, the species are still preserved are: *Sophora japonica*, *Quercus imbricaria*, *Koelreuteria paniculata*, *Maclura aurantiaca*, *Pinus silvestris*, *Pinus strobus* et al., and from planted shrubs – *Rosa canina* and *Syringa vulgaris*), **The park from Miclesti village** (among the species of trees and shrubs attested in the park are: *Acer platanoides*, *Acer pseudoplatanus*, *Acer campestre*, *Aesculus hippocastanum*, *Juglans regia*, *Fraxinus excelsior*, *Populus alba*, *Populus pyramidalis*, *Ulmus pumila*, *Ulmus laevis*, *Cerasus avium*, *Tilia tomentosa*, *Buxus sempervirens*, *Pyrus communis*, *Robinia pseudoacacia*, *Abies nordmanniana*, *Pinus nigra*, *Picea abies*, *Biota orientalis*).

Thus Criuleni district has a rich ecotourism potential consisting of objectives and natural complexes taken under state protection: 3 monuments of geological and paleontological nature, 1 monument of hydrological nature, 2 monuments of botanical nature represented by representative sectors with forest vegetation, 2 forest nature reserves, 2 monuments of landscape architecture – rich in a significant variety of floristic and faunal species, as well as 3 secular trees.

Keywords: biodiversity, ecological tourism, landscape architecture, protected natural areas.

ASPECTS REGARDING THE ECOLOGICAL SITUATION OF SOME PROTECTED AREAS IN CHISINAU MUNICIPALITY

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Currently, it is necessary to address the problems of biodiversity conservation at the geosystemic, ecosystemic and species levels. Particular attention must be paid to protected natural areas intended for the protection and conservation of plant communities and valuable wildlife habitats, and this is clearly felt in urban areas as well.

In Chisinau municipality, according to the Law on the fund of state protected natural areas no. 1538-XIII of 25.02.1998, there are 4 natural objects protected by the state: Zoo – 24,27 ha; Dendrological Garden - 83 ha; National Museum of Ethnography and Natural History - 0.075 ha; “Valea Morilor” culture and leisure park - 113.9 ha, as well as secular trees - 63 units.

The Zoo is a Scientific Institution for culturalization and training. During 2021, the correctness of the requirements for the rational use of the faunal collections as well as of the species from the animal kingdom in captivity was kept under control and verified.

Dendrological Garden has rich vegetation. During 2021, the administration of the institution did not request the Environmental Protection Agency to coordinate the forestry works. At the same time, it should be mentioned that the ecological condition of the park is satisfactory.

The National Museum of Ethnography and Natural History is a Monument of landscape architecture and guarantees the conservation and protection of the faunal and floristic collections within the botanical garden and the vivarium of the institution. The secular trees managed by the respective Museum are in a satisfactory condition, their protection regime is respected, information plates are installed.

The “Valea Morilor” culture and leisure park has the status of an architectural and landscape monument, the owner being the Chisinau City Hall. During 2021, forestry works were coordinated in relation to the vegetation proposed by the Municipal Enterprise of the Green Spaces Management Association. The vegetable kingdom is relatively rich. There are 63 secular trees in the field. The Agency for Environmental Protection for the period 2021, drafted letters to the beneficiaries of the lands in whose management are secular trees, through which it was made known the need to ensure the protection regime. Protected natural areas operate in accordance with the Framework Regulations, developed for each of their categories. The regime for the administration of the protected areas fund represents a unitary set of protection, ecological and technical-organizational measures that regulate the activity carried out within the natural objectives. The specific elements of the protected areas in Chisinau municipality correspond to the category of protection and their condition is satisfactory.

Keywords: biodiversity conservation, protected natural areas, secular trees, urban areas.

**ASSESSMENT OF THE ANTHROPOGENIC IMPACT ON THE WATER
QUALITY OF THE LOWER DNIESTER TRIBUTIES**

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It has been established that in all sections of the rivers under study, cases of pollution by biogenic substances have been identified, which differ both in the concentrations of their excesses and in the cases of their detection. The excess in the nitrogenous group dominates in the tributaries: Bull, Ikel, Botna. In the Reut tributary, the groups of nitrogen and phosphate pollutants are equal.

The most unfavorable sections of the river were identified, which were Botna, Ikel, where excesses were recorded for all nutrients and had a degree of pollution higher than in the tributaries of the Reut and Byk. It is shown that the assessment of water quality in rivers from an ecological point of view, inflows according to the same indicators had different quality classes. However, class V was defined in all river ecosystems, which indicates the fact of an increased anthropogenic load, associated primarily with the entry of pollutants into the rivers, both recent and permanent pollution.

Keywords: biogenic substances, lower Dniester tributies, nitrogen pollutants, phosphate pollutants, water quality.

MICROBIOLOGICAL POLLUTION OF SMALL TRIBUTIES OF THE LOWER PART OF THE DNIESTR

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It has been established that bacteriological pollution with concentrations of thermotolerant coliform bacteria and total coliform bacteria was recorded in all tributaries, however, the level of pollution varies significantly, which indicates a significant degree of anthropogenic load along the rivers and different volumes of pollutants.

It has been proved that the highest degree of pollution on the right side of the Dniester is for the Byk tributary, and on the left side of the river, it was found for the Svetlyi stream, but its degree of bacteriological pollution is lower than in the Byk tributary. According to the estimated bacteriological indicators, the degree of pollution of the tributaries was characterized and classified from “moderate” to “extremely high” pollution level, which indicates a possible discharge of domestic wastewater into tributaries from households located along the coast that do not have a centralized network of domestic sewerage.

Keywords: anthropogenic load, bacteriological indicators, bacteriological pollution, lower Dniester tributies, thermotolerant bacteria.

CYTOTOXIC EFFECT OF NANOCOMPOSITES BASED ON VANADIUM OXIDES

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Nanomaterials are widely used in various fields: medical, commercial and industrial production practices [1]. Despite the rapid progress and early acceptance of nanotechnologies, their potential for various adverse effects on humans, non-human biota and ecosystems is still not well elucidated. In this context, in recent years, there has been a rapid increase in experimental studies on the toxicity and biological hazards of nanomaterials in the environment [2].

In this paper we aimed to study the cytotoxic effects of nanocomposites based on vanadium oxides on onion meristematic cells. In the experiment were used the pyroceramic plates coated with thin layers of VO₂, V₂O₃ and V₂O₅, of nanometric dimensions [3]. Onion was selected as the model specimen for cytotoxicity assessment, which was exposed to nanocomposites for different time (48h and 72h), prior to germination. For cytological studies, root with a length of 1-2 cm were taken. Washing, fixation, hydrolysis, staining and preparation were performed according to the standard methodology of staining and observation of chromosomes [4].

According to the results of the cytological study, the exposure of the seeds to the action of nanocomposites based on vanadium oxides, induces changes in the values of mitotic index, comparative control. At the same time, fluctuations in the values of the phase indices of mitosis were recorded. Disruption of cell division phases indicates that vanadium oxide-based nanocomposites alter mitotic activity by cytoskeleton disorder and interfere with mitotic spindle formation, as confirmed by the occurrence of abnormal mitoses: C-mitoses, disturbed metaphase, ana-telofase spindle disorders, sticky chromosomes with bridge, vagrant chromosomes and fragments. Cells with 1-2 micronuclei and binucleated cells have also been identified, which are an indicator of the above-mentioned mutations [5].

Our results demonstrated that exposure of onion to nanocomposites based on vanadium oxides causes cytotoxicity and genotoxicity.

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Keywords: biological hazards, chromosomes, nanocomposites, toxicity, vanadium oxides.

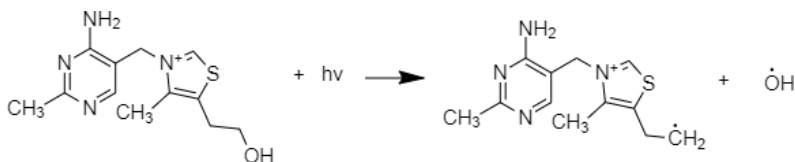
PARTICIPATION OF THE VITAMIN B COMPLEX IN PROCESSES OF CHEMICAL SELF-PURIFICATION OF THE AQUATIC ENVIRONMENT

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The product sold in pharmacies under the name Vitamin B complex after use can enter surface waters and can participate in various processes of self-purification of water. Photochemical degradation of dissolved substances takes place in the upper layers of water in case of intensive sunlight irradiation. The experimental data obtained indicate that the Vitamin B complex undergoes direct photolysis, so that it is destroyed under the influence of UV rays, with the removal of OH radicals, ensuring increased speeds in the process of photochemical destruction. The effective constant of the direct photolysis has a value $k=12 \cdot 10^{-4} \text{ s}^{-1}$ and the half-life $\tau=1\text{h } 42 \text{ min } 41\text{s}$. In other words, the studied substrate is very labile and rapidly decomposes photochemically. During photodegradation, the components of the vitamin B complex contribute to the generation of OH radicals in the environment, which is a favorable factor for the development of degradation processes. For example, in the case of direct photolysis of vitamin B₁, which is a component part of the complex, the reaction proceeds as follows:



It has been found that the rate of the induced photochemical oxidation reaction of vitamin B complex increases with increasing H_2O_2 concentration, the cause being H_2O_2 photolysis and the generation of additional amounts of OH radicals which actively participate in the destruction of Vitamin B complex. The effective constant of the induced photolysis $k=1.27 \cdot 10^{-4} \text{ s}^{-1}$ and the half-life $\tau=1\text{h } 30 \text{ min } 33 \text{ s}$, which is 12 minutes less compared to the direct photolysis of the complex.

When Cu^{2+} ions are present in the modeling system, they negatively influence the process of destroying the Vitamin B complex, so that the substance stabilizes to form a complex compound with metal ions that are more difficult to destroy. The presence of Cu(II) ions leads to two undesirable consequences: they form a complex compound that is difficult to degrade with the Vitamin B complex; by the formation of these complexes Cu(II) ions are removed from the aquatic environment and thereby diminish the intensity of the photochemical self-purification process.

Therefore, the results obtained indicate that the vitamin B complex actually participates in photochemical self-purification processes of the aquatic environment.

Keywords: Cu (II) ions, OH radicals, photolysis, vitamin B, water self-purification.

**NEW SPECIES OF MACROSTELAS (HEMIPTERA,
AUCHENORRHYNCHA) IN THE REPUBLIC OF MOLDOVA**

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The genera *Macrostelias* Fieber, 1866 is included in Tribe Macrostelini Kirkaldi, 1906. The largest genus in this tribe, *Macrostelias*, was established by Fieber (1866) with *Cicada sexnotata* Fallen as its type species. This leafhopper genus contains more than 80 species. The species of the genera *Macrostelias* are distributed in Palaearctic (Europe, Asia, North Africa), Nearctic (USA), Ethiopian (South Africa), Oriental (China, Japan, Taiwan, Korea) regions (Zhang Yalin et al. 2013). Referring to the data of fauna of the Republic of Moldova, there is information about six species: *Macrostelias fieberi* (Edwards, 1889), *M. halophilus* (Horvath, 1903), *M. laevis* (Ribaut, 1927), *M. salsolae* (Puton, 1872), *M. sexnotatus* (Fallen, 1806) and *M. viridigriseus* (Edwards, 1922).

Macrostelias is a difficult and diverse genus, most of its species are similar to each other by external signs, so it is impossible to identify them without dissection. We are using male aedeagus for the exact determination of the species. The representatives of the genus are generally slim insects with dimensions of body about 3-6 mm. The head and the pronotum are usually at the same width.

The research was realized in the northwestern part of the Republic of Moldova at the Scientific Station of Institute of Zoology in Brinzeni commune, Edineț district. The insects were collected in the period from May 25 to September 7 of 2021 in two traps with white and ultraviolet light. During the analysis of them, there were found some of well-known and spread species of genus *Macrostelias*, and also two species, that are new for the fauna: *Macrostelias frontalis* (Scott, 1875), monophage, on *Equisetum palustre* and some other horsetail species and *M. quadripunctulatus* (Kirschbaum, 1868), oliophage, on Asteraceae. The genus *Macrostelias* is important to agriculture as it contains species that feed directly on plant fluids and transmit plant pathogens, as for example *Aster yellows phytoplasma*; also this group of insects is very sensitive to the conditions of environment and reflects the specific characteristics of landscape in detail (Anufriev, 1968). The examined material is deposited in the collection of Museum of Institute of Zoology.

Acknowledgments: The study was performed under the project Nr. 20.80009.7007.02.

Keywords: *Macrostelias frontalis*, *Macrostelias quadripunctulatus*, determination, Moldovan fauna, new species.

SOME AGROCHEMICAL PROPERTIES OF CARBONATE CHERNOZEM AND DIFFERENT SOIL USE

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Soil plays a key role in the functioning of any terrestrial ecosystem. The human impact on soils has sharply increased in the face of increasing intensification of agricultural productions and leads to changes in physical, chemical and biological properties of soils. Therefore, timely control over changes in their properties is very important. The purpose of these studies is to assess changes in the chemical properties of carbonate chernozem under the influence of various types of land use: long-term fertilization, long-term crop rotation, single-crop system, forest belt, fallow.

This research was conducted on the Ketrosy (Chetrosu) Experimental Station in the Central Zone of Moldova. The soil is a Calcareous chernozem or Calcic chernozem in the World reference base for soil resources with the following characteristics: light loam with 2.5-3.0% humus, the content of mobile phosphate (Machigin) was 0.8-1.5 mg 100 g⁻¹, exchangeable potassium - 18-22 mg 100 g⁻¹ and carbonates - 1.8-2.2% in the 0-20 cm layer. In the experiment, the following variants were studied:

1. long-term fallow (over 30 years);
2. monoculture (winter wheat);
3. crop rotation without fertilization from 1953;
4. crop rotation with mineral fertilizers (N₄₇P₄₆ per year on variants with previous long-term application of N₉₀P₆₀K₆₀);
5. crop rotation with organic fertilizers (manure 18 t ha⁻¹);
6. forest belt.

Soil samples were taken from layers 0–20, 20–40, 40–60, 60–80, 80–100 cm in May, the same term for all variants. For each sample, the following agrochemical properties were determined: the content of nitrate nitrogen (NO₃-N), mobile phosphate (P₂O₅) and exchangeable potassium (K₂O).

The highest content of nitrate nitrogen was observed in the variant with minimal anthropogenic impact – with the field protective forest belt. The nitrate nitrogen content in the topsoil (0-20 cm) was minimum in the variant with monoculture – by almost 3.4 times less than in the variant with the forest belt. Long-term use of crop rotation and organic fertilizers had a favorable effect on the content of mobile phosphorus and exchangeable potassium in the topsoil, but this content sharply decreased in deeper layers. The lowest content of mobile phosphorus and exchangeable potassium in topsoil was in the variant with monoculture and in the variant with crop rotation and natural soil fertility.

Acknowledgments: This research was conducted as a part of state projects 15.817.05.13F, 20.80009.5107 “Efficient use of soil resources and microbial diversity through the use of elements of biological (organic) farming”, financed from the budget of the Republic of Moldova.

Keywords: carbonate chernozem, chemical properties, crop rotation, mobile phosphorus, nitrate nitrogen content, organic fertilizers.

THE ASSOCIATIVE AND INVASIVE IMPACT CAUSED BY COMPLEXES OF PARASITIC INSECTS AND NEMATODES WITH THE APPLICATION OF CHEMICAL MANAGEMENT IN MAIZE PLANTATIONS UNDER THE CONDITIONS OF THE REPUBLIC OF MOLDOVA

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The main reason why the area cultivated with corn has been expanded in recent years, in the Republic of Moldova, is the high profitability of this crop, taking into account the use of the most modern cultivation technologies in various cultivation systems, the implementation of modern hybrids and integrated protection management. The invasive insect species found on maize plants in their first, vulnerable stages of development, which can cause significant agro-economic impact, are the following: cutworms of the genus *Agrotis* spp., fam. *Elateridae*; Maize leaf weevil – *Tanymecus dilaticollis* Gyll., fam. *Curculionidae*, turnip moth – *Agrotis segetum*, Den. et Sch; cotton bollworm – *Heliothis armigera* Hbn., fam. *Noctuidae*; European corn borer – *Ostrinia nubilalis* Hbn., fam. *Pyraustidae*, ord. *Lepidoptera*. These species are studied annually in association with the parasitic nematode complexes detected on cereal crops, such as the representatives of the order *Tylenchida* consisting of species belonging to the genera: *Pratylenchus*, *Ditylenchus*, *Heterodera*, *Helicotylenchus*, *Paratylenchus*, *Rotylenchus*, *Tylenchorhynchus*, *Merlinius*, *Criconemella* etc., which are also the main pests noticed on maize plants, detected in all areas of phytotechnical cultivation. The attack of these pests is very dangerous when the plants are in the early stages of development (germination – the formation of 3-6 true leaves), when the crop can be completely destroyed, and the economic agents are forced to sow it again. Every year, in the most favorable areas for the established pests, the plants are attacked with varying degrees of frequency and intensity. The chemical treatment of seeds with systemic insecticides was the most effective method of protecting maize against the attack of these pests. Under the specific and unstable climatic conditions of our country, the management of chemical protection of maize provides for the use of all the available technological means to create unfavorable conditions for the development of parasitic agents, namely in the first vegetation period. The aim of this study has been to evaluate the efficacy of two new remedies with systemic insect-nematicidal action: *Curaj*, SC and *Shenzi* 200 SC, in doses of 0.15-0.20 L/ha., in comparison with the untreated control and the standard insecticide *Coragen* 20 SC, 0.20 L/ha., for combating the pest complexes that attack maize and protecting plants in the early stages of vegetation.

The results of the helminth-entomological investigations performed on maize, in the years 2021-2022, present the estimation of the phytosanitary condition of plants in the first stages of growth (germination – 3-6 true leaves), determining the parasitic impact, caused by nematodes and associated invasive insects from soil, compared by zones, sectors, periods, crop associations and cultivation technologies. The phytosanitary

surveys have shown the degree of parasitic impact, by estimating the comparative indices of numerical density (*N.d.*), the following average were obtained: insects – 3-5 individuals/m² and nematodes – 30 individuals/100 g soil, with the prevalence of higher numbers in the Center area (15-20%), in the same research period, conditions caused by climate factors, differentiated by zones. These results served as the basis for mounting research-testing experiments on the new preparations with insecticide-nematicide action: *Curaj SC* and *Shenzi 200 SC* (chlorantraniliprole 200g/kg), as a result of which a biological efficiency of 88-97% was established as compared with the untreated variant. As a result of 3 treatments applied on the complexes of Lepidoptera insects of the family *Noctuidae*, the species *Agrotis segetum*, *Heliothis armigera*, the family *Pyralidae*, the species *Ostrinia nubilalis* associated with the complexes of endoparasitic nematodes of the order *Tylenchida*, the genera *Pratylenchus* and *Ditylenchus*, semi-endoparasitic nematodes of the genera *Heterodera*, *Heterodera* and ectoparasitic nematodes of the genera *Helicotylenchus*, *Paratylenchus*, *Rotylenchus*, *Tylenchorhynchus* and *Merlinius* in the experimental sectors planted with maize, at the Institute of Phytotechny “*Porumbeni*”, Pașcani village, Criuleni district, Central Zone, Republic of Moldova, and the comparative phytosanitary investigations on maize plantations in the growing season in 2021-2022, it was found that, under the agro-climatic conditions of the Republic of Moldova, there were detected 34 harmful organisms and 15 parasitic agents that cause specific diseases, with severe parasitic impact characterized by malformations of roots, stems, leaves and cobs and differ from each other in the degree of frequency and extent, the level of damage to corn crop depending on the area, environmental factors and resistance of the planted hybrids.

Acknowledgments: The research was carried out with the support of the institutional project - state program: 20.80009.7007.12 F, 2020-2022. Project manager: Acad. Prof. Dr.hab. Ion Toderaș. The research was carried out with the support of the institutional project - state program: 20.80009.5107.15 Project manager: PhD. Pantelimon Bopozan.

Keywords: chemical treatment, efficacy, invasive insect species, maize, nematode, parasitic impact.

**CLIMATE-OPTIMIZED ADAPTIVE-LANDSCAPE TECHNOLOGIES:
CONCEPTUAL-THEORETICAL AND APPLICATIVE SUPPORT**

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The current phase of evolution of arable chernozems in the Carpatho-Danubian Pontic space is characterized by the accelerated development of quantitative-functional changes manifested in interdependent and interdetermined degradation of the organic and structural-aggregate substances system.

Quantitative expression of the level of quantitative-functional degradation is the degree of physical degradation of arable chernozems, which is currently the main factor that determines the meaning of their anthropo-natural evolution.

Direct causes of quantitative-functional degradation are the profound transformations in the organization and functioning of the soil ecosystem accompanied by the reduction of the total amount of organic substances in the soil, the transformation of soil humic system composition and the amount of organic carbon sequestered and stabilized in structural aggregates. anthropo-natural chernozemic pedogenesis (metastructuring process).

According to our research, metastructuring is manifested in increasing the degree of clodding and hardsetting of the aggregate structure, compaction of aggregates <5 mm and reduction of their porosity below 40%, reduction of aggregate content 5-1 mm below 50% in the composition of agronomic-valuable structure (0, 25-10 mm), total loss of water stability for aggregates >3 mm.

In the composition of the humic system of the soils is reduced the content of calcium humate responsible for the formation of "chernozem" aggregates (5-1 mm), increases the content of water-soluble humates formed with monovalent cations (Na^+ , K^+ , NH_4^+). In the composition of fulvic acids, the content of the fractions of "aggressive fulvic substances" increases, leading to the intensification of the mineral transformation processes and the mobilization of the organo-mineral complexes with increased humus content. The changes in the specified humic system contribute to the "eluvial humus losses" of the active agrogenic layer during the wet period of the year. At the same time, the degradation of the aggregate structure leads to a reduction in the degree of physical protection of humus in structural aggregates, intensification of structure degradation due to water action and humus mineralization processes and, respectively, to increased greenhouse gas emissions from arable chernozems. This has led to the establishment of a one-way degrading trend in arable chernozems, accompanied by a significant reduction in the capacity to adapt to climate change and severe climate phenomena, in particular drought, and to an accelerated increase in soil vulnerability to them.

This implied the need to review the paradigm of management of arable chernozem resources by placing emphasis on technological elements capable of restoring the ability to adapt to climate change and reproduce basic agroecosystemic functions: reproductive living modeling, agro-productive.

Their key objectives are:

1. Restoration of the system of organic substances and the humic profile of the soils; Enlarged reproduction of the mass, composition and diversity of soil biota;
2. Restoring the priority role of the process of humus formation and accumulation in the anthropo-natural chernozem pedogenesis;
3. Optimizing the functionality of the pedofunctional system [system of organic substances ↔ structural-aggregate system] responsible for intensifying the processes of enlarged reproduction of chernozem pedogenesis.

More recent research has shown that to these imperatives corresponds the mulch tillage > the minimum tillage system with storage of more than 30% of organic residues at the soil surface (minimum tillage) > direct sowing (no-tillage) .

An important element of these technologies is the practice of bioorganomineral fertilization with products of humic origin.

Monitoring of the structural aggregate state of clayey loamy and clayey chernozems with moderate humus content showed that the administration of bioorganomineral fertilizers in the rhizosphere and in the phyllosphere by foliar treatment contributes, even in years with severe drought, to the establishment in the agrogenic layer of such a structural aggregation that contributes to increasing of the adaptability of arable chernozems to climate change and the severe climate phenomena induced by them.

During the vegetation period, the content of aggregates 5-1 mm is maintained in the range of satisfactory values, thus ensuring the conservation and rational use of evapotranspiration of productive water reserves. In this context, the monitoring of the soil moisture regime showed even in conditions of severe drought at the end of the vegetation period of autumn crops (wheat, barley, rapeseed) the water content in the agrogenic layer is 0.45-0.52 FC (*Field Capacity*).

Acknowledgments: The researches were carried out within the Technology Transfer Project 21.80015.58.07.253T

Keywords: bioorganomineral fertilization, chernozems, calcium humate, fulvic acids, humus mineralization, quantitative-functional degradation.

**FOSSIL FELIDAE (GENUS *Metailurus*) IN THE MIOCENE OF THE
REPUBLIC OF MOLDOVA**

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Fossil Felidae of the genus *Metailurus* are widely known in Miocene deposits of Eurasia and Africa and were part of the Hipparion faunas of Vallesia and Turoly.

The earliest finds of fossil *Metailurus* in the Republic of Moldova are known from the Vallesian Hipparion fauna of Calfa (Middle Sarmatian, Calfin faunal complex, MN9; 12.0-10.8 Ma) - *Metailurus pamiri* (Ozansoy, 1965). The material is represented by the right branch of the lower jaw of the juvenile with dp3-dp4, m1, and alveolus p2, as well as isolated teeth and postcranial skeleton bones. *Metailurus cf. parvulus* Hensel 1862, a characteristic species for the hipparion faunas of the Late Sarmatian (locality of Poksesti, Poksesti faunal complex, MN 10.7-8.7 Ma) and Meotis (Early to Middle Turonian) appears in Late Vallesian. Fossil material was found during excavations in Poksesti in 2012 and is represented by a complete lower jaw of an adult.

Metailurus cf. parvulus is known from a number of localities of hipparion faunas of Meotis: *M. cf. parvulus* (= *Machairodus schlosseri* Weith.) from a locality near Cimislia (MN 12; 8.0 Ma) and *M. parvulus* (= *Machairodus parvulus* Hens.) from a locality near Taraclia (MN 12; 8.0 Ma).

In the Early Pliocene the composition of faunal complexes significantly changes and the semi-sabre-toothed Felidae of the genus *Metailurus* are replaced by representatives of the genus *Dinofelis*, found in a number of localities of Pliocene age of the Republic of Moldova.

Acknowledgments: The studies were performed within the State Program project 20.80009.7007.02 and doctoral project: "Terrestrial carnivorous mammals (Mammalia: Carnivora) from the Miocene of the Republic of Moldova".

Keywords: hipparion faunas, *Metailurus pamiri*, *Metailurus cf. parvulus*, Miocene deposits, the change of faunal complexes.

**CONTRIBUTIONS TO THE STUDY OF THE ANTS (INSECTS:
FORMICIDAE) FROM THE REPUBLIC OF MOLDOVA**

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The research was carried out in the districts of Chisinau, Calarasi, Anenii Noi, Comrat and Gagauzia in 2022. 3 types of ants were identified - *Lasius*, *Formica* and *Camponotus*. The collections were made in different habitats such as alfalfa field or with other crops, meadow in the forest or forest or river bank. Dependence on the area and the identified ant species was observed. Species of the genus *Camponotus* ants were identified only in samples collected from central Moldova (Calarasi). Habitat dependence and the species identified in the sample collected from this habitat were observed. The species *Lasius alienus* and a species of the genus *Formica* have been identified in the alfalfa plains. *Lasius emarginatus*, being a forest species, was also identified only in the samples collected from this habitat.

Acknowledgments: The study was supported by the project 220.80009.7007.02

Keywords: *Formica*, *Lasius alienus*, *Lasius emarginatus*, ants species, habitat dependence.

DEVELOPMENT OF A MATHEMATICAL MODELS SYSTEM FOR REMOTE ASSESSMENT SOIL EROSION

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The purpose of the study is developing a system for remote monitoring of the degree of soil erosion in winter wheat field crops.

For identification eroded soils was used an indicator approach, based on the sensitivity of winter wheat crop to soil erosivity. The assessment of winter wheat crops conditions was carried out according to the data of vegetation index NDVI.

The system of mathematical models developed for winter wheat production crops, based on the NDVI, is adequate and has a high information capacity. The obtained mathematical models were applied on the lands of the central part of the Grigoriopol district occupied in the period 2017-2021 by winter wheat crops grouped by the vegetation index NDVI. As a result, on an area of 16,490 hectares, the soils erosivity was determined by remote data (fig).

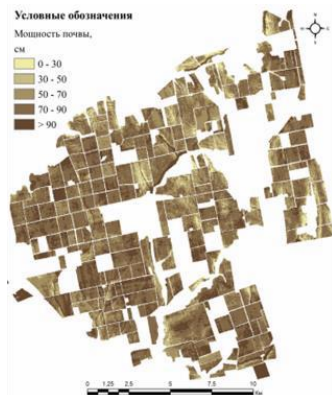


Fig. Remote estimation of soils erosivity on the lands of the central part of the Grigoriopol district

Remote estimation of soils erosivity based on the proposed system of equations better corresponds to the data of field studies of soil power compared with the data of soil maps of scale 1:50 000 and 1:10 000 and the results of soil erosion modeling by the RUSLE method.

The system of mathematical models there has been developed can be used for preliminary assessment of the degree of soil erosivity, to facilitate the work of soil scientists during soil survey.

Keywords: mathematical models, remote monitoring, soil erosion, vegetation index NDVI, wheat field crops.

BAT FAUNA (MAMMALIA, CHIROPTERA) FROM LIMESTONE MINES OF MOLOVATA NOUĂ

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The limestone mines of Molovata Nouă are located on the left bank of Nistru river (47.32 N, 29.08 E) at a distance of about 200 m from the river and at an altitude of 70 m. There are 8 entrances about the same size of 4-6 m wide and 3-3.5 m high. The corridors extend on a distance of over 700 m and are parallel, descending downwards. The ceiling and the walls have multiple cracks of 4-10 cm wide and 5-15 cm depth left after machine excavation, where the bats find shelter. This site was studied for the first time, it was not mentioned before in the literature. In the period 2020-2021 the site was visited three times: in September 2020, at the end of hibernation period in March 2021 and in June 2021. The bats were studied directly by visual observations, all observed individuals were identified.

In all the periods 11 bat species from two families were registered: *Rhinolophus hipposideros* from fam. Rhinolophidae, *Myotis myotis*, *M. blythii*, *M. daubentonii*, *M. dasycneme*, *M. nattereri*, *M. bechsteinii*, *M. mystacinus*, *Eptesicus serotinus*, *Plecotus auritus* and *P. austriacus* from fam. Vespertilionidae. The species abundance was different depending on the season. In September, when the bats are actively feeding and gather in underground sites for hibernation, 397 individuals from 9 species were recorded, dominant being *M. daubentonii* (43.94%), followed by *E. serotinus* (25.25%) and *M. mystacinus* (23.99%). Other species were observed in low amount, between 0.25% and 3.03%, while the species *M. nattereri* and *M. bechsteinii* were not registered. At the end of hibernation period the bat diversity and number were the highest – 10 species with 478 individuals, which shows the suitability of the site for many species hibernation. The dominant species was *M. daubentonii*, representing more than half of bat community, followed by *R. hipposideros* (17.15%) and *M. myotis* (15.48%), while other species had a low percent and *M. blythii* was not observed. In June, when reproduction occurs, no maternity colony was found and the bats visited the site only for daytime rest. The bat fauna was represented by 59 individuals from 6 species (*M. myotis*, *M. daubentonii*, *M. dasycneme*, *E. serotinus*, *P. auritus* and *P. austriacus*), dominant being *M. daubentonii* with more than 76%, and the rest of the species had the abundance of 1.69% to 8.47%.

All the species, except *E. serotinus*, are listed in the Red book of the Republic of Moldova. At European level all species are listed in Appendix II of Bern Convention and in Annex II of Convention on the Protection of Migratory Species. We have to mention the presence in rather large number (91 individuals) of critically endangered species *M. myotis* that was not registered in the last 50 years. The species *M. nattereri* (CR) was recorded at hibernation in low number for the first time in the last 30 years and *M. bechsteinii* (CR) was previously registered only in Cricova and Goianul Nou mines from the central part of the republic.

The Molovata Noua limestone mines represent an important site for bat diversity conservation in Moldova and the monitoring of bat fauna will continue.

Acknowledgments: The studies were performed within the State Program project 20.80009.7007.02.

Keywords: *Rhinolophidae*, *Vespertilionidae*, bat fauna, limestone mines, monitoring, species abundance.

TROPHIC SPECTRUM OF THE LONG-EARED OWL (*ASIO OTUS*) IN THE REPUBLIC OF MOLDOVA

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The long-eared owl (*Asio otus*) is a sedentary bird in Moldova and one of the most widespread in Europe. The trophic spectrum of *A. otus* was established during the winter and nesting periods in different regions of the Republic of Moldova in the period 2009-2022. During the digestion process the owl regurgitates the undigestible rests of eaten animals (bones, fur, feathers, insect chitin etc.) as pellets. The study of the pellets provides important data concerning the diet of prey birds, the small mammal fauna in the area, the density of small mammal species, their spreading etc. The small mammals and bat species were identified after the dentition, maxillary and mandibular bones.

In the northern area (Volodeni locality, Edineț district) the trophic spectrum of *A. otus* during the winter period consists only of rodent species. The field voles (genus *Microtus*) were dominant with over 70%, followed by the species genus *Apodemus* with 12.66%, *A. agrarius* with 7.59% and *Mus* species with 5.07%.

In Unguri village (Ocnita district) the trophic spectrum of the long-eared owl in nesting period also consisted only of rodents. The most abundant were the *Microtus* genus species, constituting 73.02%. *Mus* genus species constituted 11.11%, the three species of *Apodemus* genus constituted about 15%, the dominant being *A. sylvaticus* with 8.73%, and *Muscardinus avellanarius* with less than 1%. In the urban ecosystems of Chisinau, the trophic spectrum of the *A. otus* was much more diverse and consisted of mammals of three orders (Soricomorpha, Chiroptera, Rodentia) and birds, of which the share of shrews was 0.81%, of bats – 0.2%, of birds – 2.55%, while the main trophic objects were the rodents - over 95%. The *Microtus* species dominated with 70.99%, followed the *Mus* species with 10.88% and *A. sylvaticus* with 10.34%. Other species of the genus *Apodemus* accumulated less than 4%. *M. avellanarius* and *R. norvegicus* had a share of only 0.13%. Among other mammals groups in the diet of long-eared owl from Chisinau representatives of shrews and bats have been registered in very low percent. The shrews were represented by 4 species (*Crocidura suaveolens*, *C. leucodon*, *Sorex minutus*, *S. araneus*) and the bats – by 2 species (*Eptesicus serotinus* and *Vespertilio murinus*).

In the Sadaclia village (Basarabasca district) from the southern part of the republic in the pellets mammals of the orders Soricomorpha, Rodentia and birds were identified. The dominant species were the field voles (*Microtus*), which represented more than half of identified animals, followed by the *Apodemus* species with 19.71%, and *Mus* species with 16.42%. Other species had a lower percent: *A. agrarius* (2.55%), *Rattus norvegicus* (0.73%) and *Muscardinus avellanarius* (0.37%). The birds had a rather high proportion – 9.49%, while the shrews, represented by one species *C. leucodon* – only 0.37%.

Many rodent species are important pests to agriculture. The long-eared owl is a predator that exerts constant pressure on rodent density, thus regulating their number.

Acknowledgments: The studies were performed within the State Program project 20.80009.7007.02.

Keywords: *Asio otus*, undigestible rests, trophic spectrum, urban ecosystems.

THE BIOECOLOGICAL SPECTRUM OF HERBACEOUS SPECIES IN THE CENTER OF BALTI CITY

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The intensification of the processes in which permanent morphological changes of the soil cover take place has led to the need for the floristic study of the urban ecosystems. The study of the floristic diversity in the square in the center of Balti city, next to the bell tower, is of major scientific interest as it is a rather anthropized and crowded place. In the limit of the researched resort were registered 41 herbaceous species, belonging to 37 genera, grouped in 21 families. The degree of coverage is 95 - 100%. It is dominated by the Poaceae and Asteraceae families, being represented by a number of 9 and 7 species. Among the dominant species are: *Arctium lappa* L., *Lolium perenne* L., *Setaria viridis* (L.) Beauv., *S. glauca* (L.) Beauv., *Elytrigia repens* L., *Hordeum murinum* L., *Amaranthus retroflexus* L., *Poa bulbosa* L., *Cirsium arvense* L. Scop., *C. serrulatum* (Bieb.) Fisch. They constitute about 90% of the surface of the vegetal carpet. Although in the city center, including in this sector, maintenance works is periodically carried out, the flora remains fairly constant. This is due to the fact that the mentioned plants largely combine vegetative and seed reproduction, which contributes to their distribution and renewal in areas exposed to the impact of the anthropogenic factor.

The presence of two invasive species was attested within the resort: *Erigeron annuus* (L.) Pers and *Humulus lupulus* L. Their ability to adapt and multiply is quite high and as a result could contribute to the elimination of spontaneous flora species, which is represented by 18% of all species. The high share of ruderal (56%) and segetal - ruderal (26%) groups represents an index of the significant anthropogenic load on the vegetation of the study area.

In relation to the humidity conditions, the greatest diversity of the flora is represented by mesophytes - 27%, xeromesophytes, mesophytes - 27%, as well as xeromesophytes - 19%. The share of groups that require a high humidity regime is quite small.

In relation to the trophicity of the substrate, 18 indicator species were identified: Eutrophic - 14 species (78% of the total); Megatrophic - a species (5% of the total); Eutrophic, Mesotrophic - 2 species (11% of the total); Mesotrophic, Eutrophic - a species (6% of the total).

The analysis of the geobotanical spectrum shows us that the species in this resort have various centers of origin. Taxas of Eurasian and Cosmopolitan origin account for 33%, these being represented by 12 species each. Centers of origin: Circumpolar - is represented by 3 species (8% of the total); North America - 2 species (5% of the total); the other centers of origin are represented by a single species. The floristic study shows us that the flora of the Balti urban ecosystem was formed as a result of the preservation of native spontaneous species, as well as the penetration of non-native species, in different ways on the territory of the country.

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Keywords: anthropized place, invasive species, spontaneous species, urban ecosystems.

**ATMOSPHERIC BREAKDOWN CHEMISTRY OF
THE NEW GREEN SOLVENT 2,2,5,5- TETRAMETHYLOXOLANE: GAS-
PHASE RATE CONSTANTS EVALUATION TOWARDS OH AND CL
RADICALS AT 298 K AND 1 BAR**

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Aromatic solvents, unsustainably sourced from the petroleum industry, are harmful to health, often potentially carcinogenic, and environmentally hazardous [1]. Considerable research effort therefore has been directed towards developing less harmful and hazardous new "green" solvents. Such a promising green solvent with unusual properties is 2,2,5,5- tetramethyloxolane (TMO) [2]. In the present study the gas-phase reactivity of TMO towards OH radicals and Cl atoms at temperature of 296 K and total pressure on 1 bar of synthetic air was investigated using the ESC-Q-UAIC (Environmental Simulation Chamber made of quartz from "Alexandru Ioan Cuza" University of Iasi) chamber facilities [3]. Results obtained from the relative kinetic studies showed the following rate constants: $k_{(TMO+OH)} = (3.1 \pm 0.4) \times 10^{-12} \text{ cm}^3\text{molecule}^{-1}\text{s}^{-1}$ and $k_{(TMO+Cl)} = (1.2 \pm 0.1) \times 10^{-10} \text{ cm}^3\text{molecule}^{-1}\text{s}^{-1}$. Kinetic data are compared with structure-activity-relationship estimated values. Possible atmospheric implications for TMO use as a new "green" solvent are highlighted in the present study.

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Keywords: aromatic solvents, constants evaluation, environmental hazards, ESC-Q-UAIC.

**APPLICATION OF BIOTECHNICAL STRUCTURES TO IMPROVE
BREEDING CONDITIONS OF WETLAND BIRDS IN THE YAGORLYK
NATURE RESERVE**

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In conditions of unstable hydrological regime of the Dniester River, which is expressed in frequent, non-cyclical fluctuations of the water level, of great importance is the improvement of nesting conditions for wetland birds. In 2021, in accordance with the recommendations of the Reconstruction and Management Plan for the Yagorlyk Nature Reserve 4 nesting rafts were created, the target species are Great Crested Grebe and Ferruginous Duck. Rafts about 1.5 m across are made of planks and wooden slats, or from plastic containers, wire mesh and turf. A load serving as an anchor is tied to the bottom of the rafts with a rope. In the spring of 2022, 15 closed-type platforms for nesting wetland birds were also manufactured and installed among the hydrophytes. The platforms were made of wooden pallets, for buoyancy, foam material "penoplex" and plastic containers were placed under the platform. A pipe made of metal mesh and meadow hay was fixed to the platform. The dimensions of the platforms are 100/60 cm, the size of the nest tube is 80 cm in length and 30 cm in inner diameter. When nesting sites are located in dense thickets of wetland vegetation, all plants around the nest are removed by 30-40cm. The nests were located in two groups at a distance of 10 – 20 meters from each other: 1 group of 5 nests in the area of the tract "Balta"; 2 group of 10 nests in the Goyan Bay near the village of Doybany. The platforms were installed in March, immediately after the ice meltdown.

In addition to the above target species, such artificial rafts will be used by Coots and other waterfowl. Using the experience of creating biotechnical structures in conditions of unstable hydrological regime of the Dniester River it will allow to preserve and improve habitats for a vulnerable group of birds of the middle and lower Dniester in the future. Monitoring of the ornithopopulation of limnophiles it will allow tracking the dynamics of changes in the fauna of birds of the middle and lower Dniester and to identify species that are particularly vulnerable to which it is urgently necessary to apply biotechnical methods.

Acknowledgments: The studies were performed within the State Program project 20.80009.7007.02 and doctoral project «Ecological-ethological peculiarities of water birds and wader species from the Lower Dniester area»

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Keywords: Middle and lower Dniester, nesting, water level fluctuations, wetland birds.

GROWTH CHARACTERISTICS OF FUNGAL PATHOGENS IN CONDITIONS OF WATER RESTRICTIONS

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The increased impact of climate change on plant ecosystems and the security of water resources causes the prevalence of the complex fungal diseases, associated with root and stem rot, seedling rot in the early stage of growth, leaf spot, but also black embryo (black point) in mature seeds. The diversity of these diseases presents a major impediment to achieving the productive potential of the wheat globally. This research focuses on the behavior of causative agents of necrosis under water restriction conditions.

Some cultural and morphological characters of 3 strains of *Drechslera sorokiniana*, *Fusarium solani* and *Alternaria alternata* species on the osmotically modified Potato Dextrose Agar (PDA) environment were investigated. Water restrictions were adjusted with polyethylene glycol (PEG 6000) in concentrations of 5%, 10% and 20% from the volume. PDA boards, inoculated in the center with discs of uniform size (5 mm) were incubated for 7 days at a temperature of 25°C. The indices of radial growth of the colony, the texture of the aerial mycelium, the surface and reverse color, the intensity of pigmentation and the zoning were recorded on the 4th and 7th days after the initiation of the culture. The fungi *F. solani*, *A. alternata* and *D. sorokiniana* revealed, respectively, a major, medium and low growth rate over the entire 7-day interval. The radial growth of the mycelium varied in a wide range, the growth rate being more pronounced in the period 1-4 days after the initiation of the culture. The water restrictions established at the administration of PEG 6000 in 20% concentration produced significant inhibition of *F. solani* growth, but also *A. alternata* in the period of 5-7 days from the initiation of culture. The mycelium *D. sorokiniana* was tested with the most advantageous increase in various drought simulations in relation to the control. The analysis of the growth rate variance of *F. solani* mycelium demonstrated a considerable decrease in the share of the fungal strain factor (68.9%-17.0%), instead increasing the contribution of PEG 6000 concentration in the 7-day culture (18.0%-48.3%). The high level of PEG 6000 was a decisive factor for the growth of *D. sorokiniana* strains (76.5%-93.6%). The decrease in the weight of the PEG 6000 factor was accompanied by the increase of the weight fungal strain equally in the case of the growth rate of the *A. alternata* fungus. In simulated drought conditions at the administration of PEG 6000, it has been revealed the spread of the mycelium in the form of a dense submerged plate or stretched film. At the same time, the delayed appearance of the aerial mycelium, the increase or decrease of the pigmentation intensity of the mycelium, the lack or decrease of the border band show the adaptability of the growth to the water restrictions.

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Keywords: climate change, drought simulations, fungal diseases, necrosis, radial growth.

**THE IMPACT OF REGIONAL CLIMATE CHANGE ON THE
SUSTAINABLE DEVELOPMENT OF THE AGRICULTURAL SECTOR IN
THE REPUBLIC OF MOLDOVA**

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In the last two decades, the sustainable development of agriculture in the Republic of Moldova is increasingly addressed both in academic debates and in farmers' associations. The mutual support of these parties directly involved in current research on the one hand and practice on the other are strategic elements in identifying the essential needs of the population, connected to the technological limits on the ability to meet the needs of the present and the future.

Socio-cultural development, political stability, economic growth, and the implementation of effective strategies are needed by the agricultural sector at present, to bring the main branch of development of countries according to high standards in the field of production efficiency.

Efficient management of agro-climatic resources in the context of regional climate change is becoming an important condition for knowing the impact of significant weather and climate risks on the agricultural sector.

Thus, in the context of regional climate change it is important to observe and implement effective measures in the agricultural sector, effectively managing agro-climatic resources in terms of researched meteorological and climate risks, namely: drought, vegetation frosts, torrential and heavy rains, and hail massive.

At present, the socio-economic costs of natural disasters associated with climate change such as those listed above are significant. According to researchers, it is assumed that both their intensity and frequency will increase significantly in the future, which is mainly determined by climate change. The consequences of recent events have been disastrous. As an example, only the droughts of 2007, 2012, 2015, and 2020 caused significant direct losses, estimated at about 1.0-1.25 billion US dollars each.

Recent studies show that climate change is affecting all regions of the world and if extreme weather events and rainfall are becoming more frequent on the other side we are facing extreme heat waves and drought.

The analysis of the potential impact of climate change on the sustainable development of the agricultural sector in the Republic of Moldova is of enormous interest for knowing the mechanism of manifestation of these risks in order to reduce the negative effects.

Keywords: agro-climatic resources, climate risks, regional climate change, significant direct losses.

GEOPAEDOLOGICAL INVESTIGATIONS AT THE ARCHAEOLOGICAL SITE LIPOVENI II-*LA NISIPĂRIE* FROM CIMISLIA DISTRICT

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The archaeological site Lipoveni II-*La nisipărie* is located in the suburbs of the Lipoveni village from Cimișlia district. The analyzed soil samples were taken from the archaeological section in the year 2021. The investigated soil is a typical uncovered chernozem and, according to the granulometric composition, it is characterized as loamy-sandy on the loamy sand's parental rock. For comparison, soil samples were also collected from arable land, at a distance of 100 m from the archaeological site, up to the depth of the parent rock of 130 cm, on the loam's parental rock. The density of solid phase of the samples collected from the site, determined according to Petinov method, constitutes 2,53-2,56 g/cm³ up to a depth of 150 cm and 2,57-2,65 g/cm³ in the underlying horizons. The values of apparent density do not correspond to a natural genetic profile, especially from 60 cm in depth. The parameters of density of the solid phase of control profile (from arable land) was in accordance with the genetic regularities of an intact soil profile. The density of the solid phase correlates with the content of humus (organic matter) in the soil. The humus was determined using the method of V. Tiurin, based on the oxidation of organic carbon (C_{org}), with the modifications proposed by V. N. Simakov. The humus content on the entire site profile was high (2,93-4,00 %) in the upper horizons and 2,40-3,67 % in the underlying ones, correspondingly the C_{org} content also was increased. This argues for the low values of the solid phase density in the underlying horizons. For the first half meter depth, that is in the upper horizons, the humus content corresponds to this type of soil [1]. In the same time, the high content of humus and C_{org} in the underlying horizons could be originated from anthropic activity. Regarding the granulometric composition, analyzed by the pipette method of N. A. Kacinskii [2], we note the small content of fine clay (< 0,001 mm) on the site profile, the values ranging between 6,27-10,40 % in the upper horizons and 4,49-8,73 % in the underlying ones. The high content of the sand fraction (56,12-77,34 %) is due to the parent rock on which the given soil was formed – loamy sand. Special attention is paid to the segmentation of the profile into four layers: I. 0-60 cm, which according to determined parameters corresponds with the control profile from the arable land (that is to say an integral profile); the other layers show traces of human activity; layer II. 60-120 cm, with frequent presence of burnt charcoal; layer III. 120-140 cm and layer IV. 140-200 cm.

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Keywords: humus content, parameters of soil, solid phase density, uncovered chernozem.

**ELEMENTS OF AGROGENIC EVOLUTION OF THE AGGREGATIC
STRUCTURE OF CLAYEY-LOAMY AND LOAMY-CLAYEY
MODERATE HUMIFERUS CHERNOZEMS**

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The aggregate structure of soils is a distinct feature by which they differ from other bioroutinar systems and materialize in their eco/agroecosystemic functions. Through this prism of ideas, chernozems differ from other types of soils by the “chernozemic structure” represented by aggregates with dimensions mainly between 5-1 mm, porous and with increased water stability. Its formation and dynamics in conditions of natural ecosystems is the product of the interdependent and interdetermined interaction of coagulation, coprolitic and root mechanisms of aggregation-structuring that ensure the reproduction-renewal of the "chernozem structure" during the vegetation period thus ensuring the adaptation of ecosystems to the dynamics of environmental conditions.

Therefore, during the vegetation period, the content of valuable agronomic aggregates (0.25-10 mm) and agronomically precious ones (5-1 mm) are maintained in the optimal value ranges, respectively 70-80% and 50-60%. The content of aggregates > 10 mm during the vegetation varies insignificantly, and that of the aggregates < 0.25 mm does not exceed 4%.

In such a dynamic, chernozems have a great ability to adapt to environmental conditions, including climate change, especially to drought.

In agroecosystem conditions, the intensity of the reproduction-renewal mechanisms of the “chernozem structure” is significantly reduced and the decisive role in the dynamics of structural composition belongs to the thermo-compression mechanism determined by the alternation of the hydrothermal regime of arable chernozems.

In the agrogenic layer, the “agrogenic structuring” leads to the increase of the aggregates content > 10 mm and of the 5-10 mm ones. Their content increases during the growing season with maximum values at the end of it. The content of 5-1 mm aggregates during the vegetation period is reduced from 50-53% to 43-40% in the arable layer and below 35% in the subarable layer.

In the transition horizon (B) the structuring process is mainly determined by the thermo-compressive mechanism and leads to the formation of aggregates > 7 mm, their content at the end of the vegetation period has values higher than 60%.

Significantly reduces soil aggregate stability. In the arable layer (0-20-25 cm) the aggregates > 3 mm have no water stability. Aggregates 2-1 and 1-0.5 mm are characterized by maximum water stability. The aggregate content < 0.25 mm is greater than 50%.

In the subarable layer, the water stability of the 5-3 mm aggregates slightly increases due to their partial compaction and the reduction of the mesopore volume. At the same time, however, it is due to the compaction of 5-3 mm aggregates and the reduction of the volume of aggregate pores. At the same time in the subarable layer aggregates with dimensions > 10 mm with morphological and physical features not characteristic of chernozems are formed. The subarable layer is characterized by rigid placement of structural aggregates in the subarable layer in the wet period of the year creates anaerobic

conditions with negative impact on the soil biota, at the same time due to the reduction of the volume of interaggregate pores is the formation of an anisotropic profile of the porous space. This leads to a reduction in hydraulic conductivity on the soil profile and involves the seasonal over-wetting of arable chernozems during the wet period of the year.

Under conditions of low water stability of the structure, the degree of physical protection of organic carbon is reduced and as a result increase CO₂ emissions from soils. It also increases vulnerability to drought.

More recent research has shown that in order to ensure the stability of the structural-aggregate state during the vegetation period, measures are necessary to intensify the activity of the soil biota. Their number includes the treatment of soils with algal preparations based on nitrogen-fixing cyanophyte algae.

Their application to the soil in the early phases of the vegetation period contributes to the intensification of the coagulation, -radicular and coprolitic aggregation-structuring mechanisms of the soil mass with the formation of 7-1 mm aggregates and the increase of the content of water stable aggregates with a diameter of 7-1 mm. The monitoring of the structural aggregate condition during the vegetation period showed that the nitrogen-fixing cyanophyte algoflora contributes to the modeling of the soil biopedoplasm with the formation of a favorable environment for the functioning of the soil biome.

The maximum effects were found in the *Nostoc gelatinosum* variant (3 l / ha) in sunflower and corn crops.

Acknowledgments: The research was carried out within the Technology Transfer Project 21.80015.58.07.253T

Keywords: agronomic aggregates, chernozems, cyanophyte algae, hydrothermal regime, structure of soils.

CLIMATE INFLUENCES ON THE LANDSCAPES OF THE NARNOVA HYDROGRAPHIC BASIN

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Landscape structure and function can change for many reasons and in many ways. Change in a patch or a landscape can be caused by any number of factors. Some of these are intrinsic, other causes are extrinsic to the ecosystem and are imposed by outside forces, such as climate change and disturbance events. The potential causes of change may be interrelated in complex ways. A drought may make a forest more vulnerable to pathogens or a new clearing may increase the vulnerability of adjacent trees to windthrow. Global climate change will force many species to adjust their geographical distributions in the near future, with important consequences for biodiversity, conservation biology and ecosystem services. Changes in landscape structure can have several spatial and temporal forms. Patches can shrink or expand, or be lost entirely. Successional dynamics on patches can lead to a shifting mosaic of patch types through time. Some changes are nearly instantaneous and occur over very short periods of time, such as the effect of fire. Other changes occur slowly and take a longer period of time to develop, such as desertification. Patch configuration on a landscape can also change. Patches can become perforated by other patch types, and large patches can be fragmented into several smaller patches.

The varied natural conditions of the Nârnova river basin, with varying altitudes, have determined the presence of a wide variety of land categories (types of landscapes). Climatic conditions have had a direct influence on the increase and decrease of certain areas of land. Within the Nârnova river basin, during the years 1991-2020, an average temperature of 10.88 degrees C was registered, and the average rainfall was 580 mm. Recently, a series of hydroclimatic changes have appeared that have led to the reduction of the snow layer, the change of precipitation from snow type to rain. Drought phenomena have also become more numerous, which has further led to the appearance of vegetation fires (dry vegetation has become more flammable) as the climate grows warmer and drier. Within the Narnova river basin, the arid climate has led to a decrease in the number and surface of the lakes, which has directly influenced even the actual course of the river, decreasing its flow. The surface of many perennial landscapes has turned into agricultural or pasture landscapes. The latitudinal distribution of temperatures influenced the growth of forest and multiannual landscapes. The torrential rains also intensified, which led to intense soil erosion due to the rather rugged terrain. This in turn led to the transition from arable landscapes to shrubs, bushes, eroded pastures, etc. If climate change is affecting landscape processes over the multidecadal time scales of human lives and infrastructure design, that could have major implications for human health and safety, economic stability, water security, natural-resource management, and ecosystem resilience.

Keywords: causes of change, drought, landscape structure, climatic conditions

**EX-SITU AND IN-VITRO PROPAGATION OF
GENISTA TETRAGONA IN REPUBLIC OF MOLDOVA**

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Botanic gardens are the main institutions involved in *ex situ* conservation of wild plant diversity and have adopted the Target 8 of the Global Strategy for Plant Conservation (GSPC), which evokes that 75% of threatened plant species should be maintained in *ex situ* collections in order to provide genetic diversity for ecological restoration efforts.

The *ex situ* population of *Genista tetragona* Besser (Fabaceae) was created in 2000 by transplanting several vegetative specimens from the natural habitat (the vicinity of vill. Mașcăuți, Criuleni district) to the experimental plot of rare plants (National Botanical Garden (Institute), Chișinău), in order to continue on *in situ* restoration programs. The plants took root on the site, but during 2000-21 they did not form seeds, and efforts for vegetative propagation did not produce results. As a result, in 2021-22 actions were taken to propagate this species by microclonal methods. *Genista tetragona* is a xerophylous, calciphylous shrub species. Blooms in April-May and fructifies in June-August. In the Republic of Moldova species grows in the districts of Camenca, Rîbnița, Dubăsari, Grigoriopol and Orhei. It is a Podolian endemic, a tertiary relict. It is a highly decorative shrub, especially in the spring time. The species is protected globally – included in the IUCN Red list of threatened species, Annexes of Bern Convention, and regionally – protected by state in the Republic of Moldova and included in the Red Book of the republic and Ukraine.

At present, priority propagation is given to *in vitro* techniques that offer wide opportunities in the conservation of rare and endangered plant species, thus being a complementary to *in situ* conservation.

This study aimed at the introduction and multiplication in the *in vitro* culture of the species *Genista tetragona*. *In vitro* sampling, inoculation and culture methods were traditional. The MS medium (Murashige, Skoog, 1962), supplemented with BAP and TDZ cytokines (0.5 mg/l), served as the basic nutrient substrate. Apical bud segments were used as biological material. The inoculated material under aseptic conditions was transferred to the culture chamber with a specific regime for the investigated plants. In the process of *in vitro* cultivation, observations and analyzes were performed in order to microclon them. Phytohormones had different effects on the development of shoots and on the rate of seedling proliferation *in vitro*. Maximum proliferation values were obtained on the medium supplemented with BAP of 10.4 vigorous new shoots/explant, TDZ generated some symptoms of hyperhydration. *In vitro* seedlings are currently being induced to take root.

Acknowledgments: The research was supported by the NARD through the Project “Research and conservation of vascular flora and macromycobiota of the Republic of Moldova”, 20.80009.7007.22.

Keywords: *Genista tetragona*, microclonal methods, *ex situ* conservation, *in vitro*.

THE INFLUENCE OF TOMATIN BAC ON THE PROCESS OF ALCOHOLIC FERMENTATION OF WASTE BIOMAS

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The quantity of agricultural waste has been rising rapidly all over the world, many studies has revealed that fruits and vegetables are the main source of bioactive compounds; in most cases, wastes and byproducts generated by the food processing industry present similar or a higher content of antioxidant compounds. Therefore, there is an increasing interest in finding new ways for their processing toward safely upgrading these wastes for recovering high-value-added products with a sustainable approach. Among food waste, the abundance of bioactive compounds in byproducts derived from tomato suggests possibility of utilizing them as a low-cost source of antioxidants as functional ingredients. The solid residue remaining after the industrial processing of tomatoes (*Solanum lycopersicum* L.), tomato pomace, consists of large amounts of tomato peels and seeds that currently find use as animal feed and fertilizers or are sent to landfill. However, it is still rich in important antioxidant present in the ripened tomato. In this line, numerous approaches have been proposed for the valorization of the unused parts of tomato in various sectors. In the past, people added these byproducts as compost to the soil for agricultural purposes, thus allowing the recycling of nutrients. Today, instead, because of the huge increase in the accumulation of large amounts of waste matter, reducing waste is among the efforts to relieve the pressure on natural resources and move toward more sustainable food systems. So, a critical and up-to-date review has been conducted on the latest individual valorization technologies aimed at the generation of value-added by-products from industrial food wastes. Waste treatment in wine and alcohol industry is a current problem in Republic of Moldova. The annual volumes of the production and accumulation of liquid wastes are considerable.

The aim of the scientific research is to examine the associated advantages and drawbacks of each technique separately along with the assessment of process parameters affecting the efficiency of the generation of the bio-based products. Research of the influence of tomatine BAC on the process of alcoholic fermentation in the laboratory conditions. The biomass used was the waste from the ethyl alcohol production "GARMA GRUP" L.L.C. company. The testing of tomatine BAC was performed in order to explore possible activating or inhibiting effect on the alcoholic fermentation process.

Acknowledgments: The State Project 20.80009.500727 "Physico-chemical mechanisms of redox processes with electron transfer involved in vital, technological and environmental systems", running at Technical University of Moldova, Department of Oenology and Chemistry.

Keywords: agricultural waste, alcoholic fermentation, antioxidants, sustainable approach, tomato pomace, valorisation.

FIXED SOURCES OF ATMOSPHERIC AIR POLLUTION IN THE BALTI URBAN ECOSYSTEM

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Stationary sources of pollution (excluding the household sector) in the Balti urban ecosystem eliminate a huge volume of pollutants into the atmosphere. The number of economic agents registered in the municipality that eliminate pollutants in the atmosphere in the production process for the period 2014 - 2021 is decreasing: from 408 to 222. The dynamics of the volume of emissions in the municipality is increasing for the period 2014-2021 from 855.13 t to 1544.86 t. (Fig. 1).

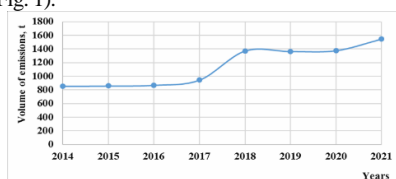


Fig. 1 The dynamics of the volume of emissions from stationary sources in Balti municipality (2014 - 2021)

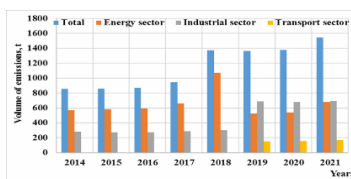


Fig. 2 The dynamics of the volume of emissions by activity sectors in Balti municipality (2014 - 2021)

For the period 2014 - 2018, the largest volumes, about 70%, were emitted by the energy sector, and the rest from the industrial sector (Fig. 2). Starting with 2019, the industrial sector emits about 700 tons of emissions into the atmosphere compared to about 300 tons by 2018. Among the largest companies in the thermal energy sector with increased emission volumes is J.S.C. "CET-Nord" with 50.29 t in 2021 and Î.M. „Termogaz Bălți” with 9.102 t. Regarding the transport services sector, there is an increase in the volume of emissions from 151.107 t in 2019 to 170.819 t in 2021. Among the biggest polluters in this field are LTD "Lukoil Moldova" - 36,069 t considering all the fuel supply points from the municipality, followed by LTD „Dominic” – 34.441 t, LTD „Petrom Moldova” – 29.382 t, LTD „Bemol Retail” - 12, 563 t, etc. In the food production sector for 2021 stand out J.S.C. „Floarea Soarelui” with 604.287 t, J.S.C. "Incomlac" – 29.548 t, J.S.C. "Vinăria din Vale" - 7,449 t, J.S.C. "Basarabia Nord" - 6,008 t. The sector of production of construction materials in the Balti municipality is represented by Î.M. MG „CMC-Knauf” with 26.953 t, LTD „Dolomita Prim” (with the specificity of producing concrete articles) with 6.673 t. and others with a lower volume. An increased volume of emissions for the Balti urban ecosystem is also represented by LTD "Gloring Engineering", which in the process of activity for 2021 emitted into the atmosphere 241.863 t. The composition of the emissions of this economic agent is 69% hydrocarbons (CH), followed by CO - 21% and solids - 3%. Nitrogen and sulphur dioxide (NO₂ and SO₂) accounted for 1% each.

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Keywords: air pollution, polluters, economic agents, urban ecosystem, emissions.

EUROPIUM BIOACCUMULATION BY *ARTROSPIRA PLATENSIS* AND ITS EFFECT ON BIOMASS

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Europium is one of the most reactive rare-earth elements, which due to its physical and chemical properties is widely applied in many technological processes. Nowadays, it is imperative to develop sustainable, eco-friendly and promising extraction techniques and explore alternative europium resources, along with protecting the environment and human health. The ability of cyanobacteria, *Artrospira platensis*, to accumulate europium and its effect on biomass biochemical composition was evaluated. Europium accumulation in biomass was traced by ICP-AES technique. At addition of europium ions in the cultivation medium in concentrations 10-30 mg/L, its accumulation in biomass was 9.8-29.8 mg/g (removal efficiency being) 98-99%. Europium ions in studied concentrations range did not affect productivity and content of carbohydrates and pigments in biomass, and led to the decrease of the content of protein and an increase in the amount of MDA. *Artrospira platensis* can be considered as a potential bioremediator for treatment of wastewater polluted with europium.

Keywords: *Artrospira platensis*, alternative europium resources, bioremediator, biomass, extraction techniques.

FINDING OF THE REMAINS OF A FROG AND VARAN FROM THE PLIOCENE LOCALITY PRIOZERNOE

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During the 2021 season fieldwork, reptile and amphibian remains were found in the early Pliocene alluvial sediments of the Dniester River valley in a sandstone quarry near the village of Priozernoye (Moldova, Transdnistria).

The reptile remains, represented by a vertebra of the trunk, belonged to a large lizard, identified by us as *Varanus* sp. From the Pliocene deposits of Moldavia, there is only an indication of the presence of *Varanus* sp. from the localities Etulia and Suvorovo (Zerova & Chkhikvadze, 1984) (description and photos are not provided), which requires clarification. The closest Pliocene faunal localities from which remains of a representative of this family have been previously described are located in the Odessa Region, Ukraine (Kotlovina-1 and Kotlovina-2) (Ratnikov, 2002).

At the end of the field season, a torso vertebra of a large amphibian of the order Anura was found. The vertebra is very large, more than 2 cm wide. It belonged to a frog of the family Palaeobatrachidae. The *Palaeobatrachus* cf. *langhae* (biozones MN 14-15) was a characteristic species of the Pliocene of Europe (Hungary, Poland, Russia and Slovakia) (Ratnikov, 1997, 2002; Villa, Roček at al., 2016). The remains have been tentatively identified as *Palaeobatrachus* sp. Thus, for the first time for the Pliocene of the Republic of Moldova (Transdnistria) a new species of large frog of the family Palaeobatrachidae was identified. The presence of lizards of the family Varanidae in the Pliocene in the Dniester River valley was also reliably established.

These findings supplement our understanding of the fauna of our region during the Early Pliocene and allow us to clarify the paleogeographic and paleoecological conditions of the early stages of formation of the large paleo-river valleys of the Dniester and Prut rivers.

Acknowledgments: The studies were performed within the State Program project 20.80009.7007.02 and doctoral project «Fossil fauna complexes and the evolution of vertebrate fauna in the early stages of the formation of the Dniester Valley (Pliocene-Early Pleistocene)».

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Keywords: *Palaeobatrachus* cf. *langhae*, paleoecological conditions, Pliocene deposits

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Session E

**CHEMISTRY AND
CHEMICAL
COMPOUNDS IN
BIOLOGY,
AGRICULTURE AND
MEDICINE**



OBTAINING ACTIVE DRY WINE YEAST BIOMASS AT THE MICROPILOT LEVEL

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A *Saccharomyces cerevisiae* strain, isolated from “Fetească regală” grapes cultivated in “Pietroasa” Viticulture and Winemaking Research and Development Center, belonging to the University of Agronomic Sciences and Veterinary Medicine of Bucharest (USAMV) and identified by molecular sequencing, was used for cultivation on a synthetic medium to obtain active dry yeast biomass. The scale-up from laboratory level at micropilot level was performed in this study, following the optimization of the process at laboratory level. The pre-inoculum was based on pure culture of *Saccharomyces cerevisiae* which was used to inoculate a liquid YSP medium (yeast extract, hy-soy peptone, and sucrose) to obtain the liquid inoculum. The liquid inoculum was used to inoculate the fermentation medium (carbon source - sugar and nitrogen source - yeast extract) from the bioreactor (working volume 4L). The cultivation parameters were monitored online (temperature 30°C, stirring rate 200-300 rpm, pH 4-5) and off line (total soluble dry matter, pH; wet cell biomass and dry cell biomass). Finally, following the post-fermentation process, a wet biomass of 46.5 g/L was obtained, respectively active dry wine yeast 14.79 g/L, which will be used in the experiments regarding the production of “Fetească regală” wines from “Pietroasa” Viticulture and Winemaking Research and Development Center, USAMV, Bucharest for assessing the terroir behavior.

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Keywords: active wine dry yeast, *Saccharomyces cerevisiae*

THE USE OF ^2D NMR SPECTROSCOPY FOR ANALYTICAL EVALUATION OF LAVENDER EXTRACTS. DETERMINATION OF ROSMARINIC ACID

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NMR spectroscopy is a powerful method for structural identification of both individual compounds and complex natural products mixtures. The main advantages of this modern analytical tool are, among others, high resolution power, as well as simultaneous qualitative and quantitative evaluation of the investigated sample [1].

The current work presents the determination of rosmarinic acid (RA) content in lavender extracts basing on fast and reliable 2D NMR correlations. Few data on the quantitative determination of RA in lavender are known in the literature, even though it is an abundant and highly valuable secondary metabolite [2]. A pure sample of RA was isolated from dried leaves of lemon balm (*Melissa officinalis* L.) and its ^1H and HSQC spectra recorded. Purity grade of the obtained sample was calculated basing on ^1H NMR and internal standard (IS) method. Methyl 4-nitrobenzoate was used as an IS, both for ^1H and HSQC measurements. Two diagnostic signals of RA have been chosen on the basis of their separation from the IS peaks. The HSQC relaxation delay parameter (D_1) was optimized basing on the relaxation times T_1 determined for both diagnostic signals. The calibration curve was drawn on the basis of HSQC correlation, plotting the peak volumes of the chosen diagnostic peak, normalized to the peak volume of IS versus molar ratio between RA and IS. A perfect linearity was achieved. The quantitative determination of RA in a lavender extract was easily performed, thanks to an excellent resolution of RA diagnostic signals from all other extract components cross peaks. The elaborated method can be routinely used for the quality control of herbal pharmaceuticals or related products. The analysis time is comparable with the time of an average HPLC experiment. The sample preparation procedure and interpretation of results are very simple and can be easily adapted for any modern analytical laboratory.

Acknowledgments: This work was funded by the National Agency for Research and Development (ANCD) of the Republic of Moldova, projects PLANTERAS, code 20.80009.8007.03.

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Keywords: diagnostic peak, NMR spectroscopy, rosmarinic acid, quality control, quantitative method.

SELECTIVE EXTRACTION OF POLYPHENOLIC COMPOUNDS FROM *HIPPOPHAE RHAMNOIDES* SEEDS

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The antioxidant properties of natural products are generally connected with the content of polyphenolic compounds, which can efficiently quench active free radicals and diminish oxidative stress of the living cells. This is very important both for maintaining and amelioration of cellular function and regarded as a relevant tool in wound tissue repair [1]. The oil obtained from the sea buckthorn (*Hippophae Rhamnoides*) represents a relevant remedy used in folk medicine for wound healing [2]. We report in the current communication the selective preparation of a polyphenol – rich extract from sea buckthorn seeds which are primarily used for oil production. The total content of phenolic compounds in the obtained extract was determined by Folin-Ciocalteu method against both galic and chlorogenic acids as standards. The high free radical scavenging ability was determined basing on the DPPH antioxidant assay. The cell-proliferation properties have been investigated by *in vitro* MTT tests on mesenchymal stem cells from rabbit bone marrow. The elaborated preparation method is simple and can be up-scaled for large production and following pre-clinical studies.

Acknowledgments: This work was funded by the National Agency for Research and Development (ANCD) of the Republic of Moldova: projects PLANTERAS, code 20.80009.8007.03.

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Keywords: antioxidant, buckthorn seeds, DPPH antioxidant assay, extraction, *Hippophae rhamnoides*, method, polyphenolic compounds.

STOPPED-FLOW STUDIES OF THE INTERACTION OF DFH₄ AND ITS DERIVATIVES WITH DPPH[•]

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In the course of this research, ascorbic acid, dihydroxyfumaric acid (DFH₄) and ten of its newly obtained derivatives were investigated in the modified DPPH[•] assay using the stopped-flow method.

Fig.1. shows five most potent antioxidants synthesized from DFH₄.

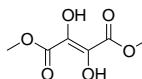
Obtained curves demonstrate the existence of three types of kinetic behaviors: fast, mixed (the fast stage is followed by a slower stage, which can be visually distinguished) and slow kinetic behavior.

The kinetics is determined both by the molecular structure of the studied compound and by the stability of the antioxidant radical formed following the reaction with DPPH[•].

Ascorbic acid is the only compound exhibiting rapid reaction with the radical, which is determined by the presence of active functional groups, which allow the rapid transfer of hydrogen atoms.

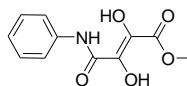
Analysis of the obtained kinetic curves shows that DFH₄, ester 2, acid 12 and diol 13 (Fig.1) show mixed kinetics. Upon interaction with DPPH[•], a rapid decrease in radical concentration is observed in the first 3-10 seconds (1 min for ester 2), but the steady state is reached in a longer time (several minutes), compared to ascorbic acid.

Anilide 5 and fumaramide 6 exhibit a slow kinetics in the DPPH[•] assay, with a reaction time of 45-60 minutes.



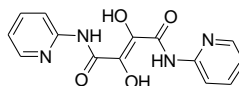
2

dimethyl 2,3-dihydroxifumarate



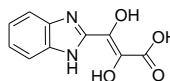
5

(*E*)-methyl 2,3-dihydroxy-4-oxo-4-(phenylamino)but-2-enoate



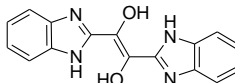
6

2,3-dihydroxy-N¹,N⁴-di(pyridin-2-yl)fumaramide



12

(*E*)-3-(1H-benzo[*d*]imidazol-2-yl)-2,3-dihydroxyacrylic acid



13

(*E*)-1,2-di(1H-benzo[*d*]imidazol-2-yl)etene-1,2-diol

Fig.1. Most potent obtained derivatives of DFH₄

ESTIMATION OF ADMET PROPERTIES OF DFH4 AND ITS NOVEL DERIVATIVES

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Computer assisted prediction of absorption, distribution, metabolism, excretion and toxicity (ADMET) was performed using the SwissADME online tool, for the dihydroxyfumaric acid (DFH₄) and its synthesized derivatives, which showed good antiradical activities in previous studies (Fig. 1).

The colored part of the radar shows the desired physico-chemical surface suitable for oral bioavailability of the compound: LIPO (lipofilicity): $-0,7 < XLOGP3 < +5,0$; SIZE: $150\text{g/mol} < \text{molecular weight} < 500\text{g/mol}$; POLAR (polarity): $20\text{\AA}^2 < \text{TPSA} < 130\text{\AA}^2$; INSOLU (insolubility): $-6 < \log S \text{ (ESOL)} < 0$; INSATU (insaturation): $0.25 < \text{ration of sp}^3 \text{ C atoms} < 1$; FLEX (flexibility): $0 < \text{number of rotatable bonds} < 9$.

All studied compounds satisfy *Lipiski's* rule of five, and have a good estimated bioavailability (0.56), while Fig.1. also shows a good drug-likeness.

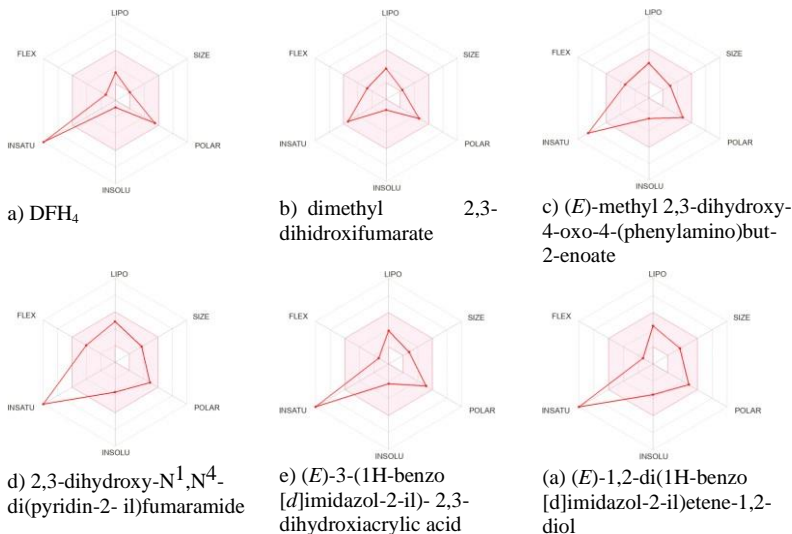


Fig.1. Bioavailability radars for the studied compounds.

Keywords: dihydroxyfumaric acid, DPPH• assay, functional groups, newly obtained derivatives, kinetic behaviors.

CAROTENOID CONTENT IN PLANT PRODUCTS OF *CASSIA OCCIDENTALIS* (L.) LINK SPECIES

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The genus *Cassia* contains about 60 species, including *C. angustifolia*, known as medicinal plant. In recent decades the other species *C. occidentalis* has become the object of phytochemical studies, which allowed the identification of various chemical compounds with therapeutic effects such as anthraquinones, phenolic compounds, and carotenoids. This species was introduced into the Collection of Medicinal and Aromatic plants of the Institute of Genetics, Physiology and Plant Protection of the Republic of Moldova.

Objective of the study was the determination of carotenoid content in different vegetal products collected from the *C. occidentalis* species, grown in climate conditions of the Republic of Moldova.

Carotenoid dosing was performed at *Metertech* UV/VIS SP 8001 spectrophotometer (absorbance 448 nm) in different plant products – *Folia*, *Flores*, *Fruits* and *Herba* of *C. occidentalis* species. Extraction was carried out in 2 extractants (95% ethyl alcohol and hexane) on a water bath at 60°C. Content was expressed as mg% FW.

The experimental data denote that, for all analyzed vegetal products, the extraction of carotenoids in 95% ethyl alcohol was more efficient than in hexane. The obtained results show that total carotenoids in recalculation to β -carotene varies in the analyzed plant products from 18.48 to 88.10 mg% in 95% ethyl alcohol and from 2.09 to 28.7 mg% in hexane extractant. The highest values recorded in both extractants were reported for *Folia* – 88.00 mg% in 95% ethyl alcohol and 28.70 mg% in hexane, followed by *Flores* (respectively) – 58.82 and 18.47 mg% and *Herba* – 55.23 and 17.17, but the lowest in *Fructus* 18.48 and 2.09 mg%.

Ethyl alcohol 95% is much more effective than hexane for extracting carotenoids from all plant products of *C. occidentalis* species. All analyzed plant products are characterized by carotenoid content and are of interest for the cultivation and valorization of this species for pharmaceutical purposes.

Acknowledgments: This study was carried out with the support of the project "Diminishing the consequences of climate change by creating, implementing varieties of medicinal and aromatic plants with high productivity, resistant to drought, wintering, disease, ensuring sustainable development of agriculture, guarantees high quality products, predestined for the industry perfumery, cosmetics, pharmaceuticals, food ", code 20.80009.5107.07

Keywords: antiradical activities, bioavailability, dihydroxyfumaric acid, SwissADME.

CONTENT OF TANNINS IN PLANT PRODUCTS OF SOME SPECIES FROM GENUS ACTINIDIA

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The genus *Actinidia*, native to the temperate eastern Asia, contains about 80 species. Many species of this genus have interes for pharmacy, food, green space ornamentation and cosmetics. Plant products collected from 3 species – *A. kolomikta* (Rupr. & Maxim),

A. argura (Siebold & Zucc.) Planchand and *A. deliciosa* (A. Chev.) C.F. Liang & A.R. Ferguson, which were introduced into the plant collection of the *Alexandru Ciubotaru* National Botanical Garden were subjected to phytochemical analysis, including tannin content.

Objective of the study was the determination of tannin content in different vegetal products collected from 3 species of genus *Actinidia*, grown in climate conditions of the Republic of Moldova.

Tannin dosing was performed by titrimetric method in different plant products – *Radices, Cortex, Folia and Fructus* of *A. kolomikta*, *A. argura* and *A. deliciosa* species. Content was expressed as % FW.

The experimental data, obtained in 3 replicates, show that all the products analyzed contain tannins, but the content varies from 0.575 to 11.361% depending on the type of plant product and species. The highest tannin values were recorded for juvenile vegetal products of *Cortex* (11.361%) and *Folia* (8.563%) from species *A. kolomikta*. For the other vegetal products such as mature leaves, roots, mature bark, fruits of *A. kolomikta* as well as the other 2 species *A. arguta* and *A. deliciosa* the tannin content ranged between 0.575 and 2.403%.

From all plant products (*Radices, Cortex, Folia, Fructus*) analyzed from 3 species of *Actinidia* (*A. kolomikta, A. arguta and A. deliciosa*), grown in the climate conditions of the Republic of Moldova, the juvenile leaves and bark of *A. kolomikta* species were found to have a high tannin content, which can be used for pharmaceutical purposes.

Acknowledgements: This study was carried out with the support of the project "Introduction and development of technologies for the propagation and cultivation of new woody plant species by conventional techniques and *in vitro* cultures", code 20.80009.7007.19

Keywords: *Actinidia sp.*, tannin content, titrimetric method, vegetal products.

PRELIMINARY PHYTOCHEMICAL ANALYSIS OF CRUD EXTRACT FROM *TANACETUM CORYMBOSUM* (L.) SHI. BIP.

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Tanacetum corymbosum (Corymbflower tansy) is an herbaceous perennial plant growing solitary or in small groups in the spontaneous flora of the Republic of Moldova. This specie is used in digestive disorders, as antiprotozoal and antibacterial remedy. Unlike, its chemical composition is less studied and this study is a continuation of the previous works, when the elemental analysis of *T. corymbosum* plants [1], chemical composition of its essential oil and antimicrobial analysis of crud extract were reported [2].

The aim of the present research was to investigate phytochemical profile of *T. corymbosum* extract using GC-MS analysis. According to this, the chemical composition of the species is extremely varied and includes constituents of many and diverse classes of organic compounds. The aliphatic hydrocarbons are presented by higher alkanes (C₉-C₂₁), together with others cyclic, unsaturated or aromatic compounds. Also, a small number of alcohols (e.g. cetylic), aldehydes (e.g. caproic) and more numerous carboxylic acids (e.g. capric, myristic, stearic) and esters (e.g. methyl palmitate, ethyl linoleate, bornyl acetate) were detected.

However, terpene compounds determine the chemical profile of the species. The most numerous are monoterpenes, more exactly hydrocarbons (e.g. limonene, *p*-cymene) and oxygenated forms (e.g. linalool, camphor). They a followed by sesquiterpene hydrocarbons (e.g. caryophyllene, humulene) and oxygenated forms (e.g. farnesol). Other superior terpenes have been identified in smaller numbers: diterpene hydrocarbons (e.g. neophytadiene) and oxygenated forms (e.g. phytol), or triterpenes (e.g. squalene).

Based on the present investigation, we conclude that *T. corymbosum* species because of its active principles can be used for pharmaceutical purposes.

Acknowledgements: This work is part of the project PLANTERAS 20.80009.8007.03 "New substances with preventive and therapeutic potential based on natural compounds of plant origin and modern methods of organic synthesis" within the State Program (2020-2023) financed by the National Agency for Research and Development.

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Keywords: organic compounds, phytochemical profile, *Tanacetum corymbosum*, terpene compounds.

SYNTHESIS OF N-CYCLOHEXYL-2-[(3-ETHOXY-2-HYDROXYPHENYL) METHYLIDENE] HYDRAZINE-1-CARBOTHIOAMIDE

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The recently studied thiosemicarbazones have exceeded researchers' expectations by demonstrating advanced pharmacological effects, such as anticancer, antibacterial and antioxidant effects.

The paper presents the synthesis of new ligand N-cyclohexyl-2-[(3-ethoxy-2-hydroxyphenyl) methylidene] hydrazine-1-carbothioamide (**H₂L**) obtained through the addition of 2-ethoxy-6-[hydrazinylidenemethyl] phenol to isothiocyanatocyclohexane.

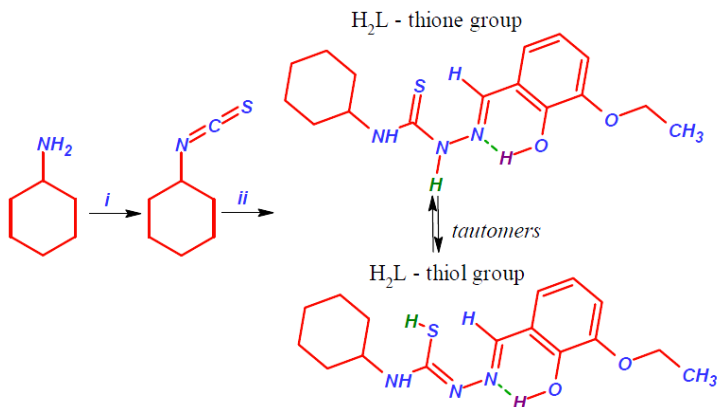


Figure. Synthesis of N-cyclohexyl-2-[(3-ethoxy-2-hydroxyphenyl) methylidene] hydrazine-1-carbothioamide (**H₂L**)

i- CSCl_2 , CaCO_3 , H_2O , (extracts with CH_2Cl_2), 2 h;

ii- 2-ethoxy-6-[hydrazinylidenemethyl]phenol (preferred name - hydrazone to 3-ethoxyalicylaldehyde) tetrahydrofuran, 1.5 h.

Thiosemicarbazone **H₂L** was characterized on the basis of various spectroscopic techniques like FTIR, ¹H and ¹³C NMR studies, elemental analysis. The compound was subjected to antimicrobial and antifungal activity screening using serial broth dilution method. The thiosemicarbazone **H₂L** showed moderate activity against: *Staphylococcus aureus*, *Bacillus cereus*, *Escherichia coli*, *Acinetobacter baumannii* and *Candida albicans*.

Keywords: antifungal, antimicrobial, dilution method, pharmacological effects, thiosemicarbazone **H₂L**.

JINMIAO TARGET INHIBITION OF *O. CUMANA* THE MECHANISM OF PARASITIC SUNFLOWER

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Jinmiao Target is a new kind of anti broomrape induced resistant agent. It has been shown that stem and leaf spray and root drip irrigation can inhibit the parasitism of sunflower broomrape, but the mechanism is unclear. The mechanism of the inhibition of broomrape on sunflower resistant variety (JK103) and susceptible variety (LD5009) by root watering was studied. Compared with the control, the number of tubercles decreased by 95.5 and the parasitism rate decreased by 98.20% after treatment with LD5009+Jinmiao Target, the fresh weight and dry weight of tubercles decreased by 94.60% and 81.63%. The height and stem diameter of sunflower increased by 2.09 cm and 0.52 cm and the growth rates were 14.92% and 15.29%. However, the number of tubercles in the resistant variety JK103+Jinmiao Target reduced by 37.5 and the parasitism rate decreased by 98.04%, the fresh weight and dry weight of tubercles decreased by 97.06% and 82.69%. The height and stem diameter of sunflower increased by 2.07 cm and 0.39 cm and the growth rates were 12.26% and 9.70%. After irrigation the corpus callosum deposition in the roots of the resistant and susceptible cultivars was increased. However, the resistance variety JK103 showed the most significant increase after 48h.

The results showed that after root irrigation in both resistant and susceptible varieties, the content of H₂O₂ after 24h reached the maximum, 3.53μmol/g and 2.68μmol/g respectively. The most significant increase was recorded in the susceptible variety LD5009, an increase of 208.05%. The activities of four different ROS scavenging enzymes showed an initial trend of increasing and then decreasing after treatment. The maximum value was reached at 48h after JK103+Jinmiao Target treatment, the maximum values of SOD, POD, CAT and PPO were 69.77g⁻¹, 5.44g⁻¹·min⁻¹, 1.88g⁻¹·min⁻¹ and 527g⁻¹·min⁻¹ respectively. However, after LD5009+ Jinmiao Target treatment, the activities of the above four ROS scavenging enzymes were increased by 25.91g⁻¹, 13.16g⁻¹·min⁻¹, 0.50g⁻¹·min⁻¹ and 313g⁻¹·min⁻¹. Transcriptional analysis of related resistance genes indicated that the resusceptible variety was induced in different degrees after target treatment. However, induction degree of LD5009 with Jinmiao Target was the most obvious, especially *CAT*, *Mn-SOD* and *XTH6*. The relative expression levels were more than 50 times higher than the control. In conclusion, both resistant and susceptible varieties, the host resistance was induced at different levels after Jinmiao Target treatment, the degree of induction varies from cultivar to cultivar.

Keywords: antibroomrape agent, Jinmiao Target, mechanism of the inhibition, resistant agent, sunflower.

STUDY OF THE ANTIOXIDANT PROPERTIES OF SOME METHYLPHENYLTHIOSEMICARBAZONES

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Antioxidants are important compounds that reduce or neutralize free radicals, thus protecting cells from oxidation. Considerable research has been directed towards the identification of new antioxidants to prevent radical damages.

Due to the presence of donor atoms such as N, S, the thiosemicarbazide backbone has been extensively studied over the last 70 years, Therefore numerous thiosemicarbazide derivatives as substituted thiosemicarbazones at N (4) and N (1) of aliphatic, aromatic and heteroaromatic carbonyl compounds were synthesized and evaluated for antitumor, antimicrobial, cytotoxic and antioxidant activity.

In order to supplement the data on agents with potential biological activity, three N4-n- methylphenylthiosemicarbazides were synthesized, and subsequently condensed with 2- hydroxy-3-methoxybenzaldehyde (o-vanillin). The antioxidant activity of the compounds was then evaluated by analysis of 1,1-diphenyl-2-picrylhydrazyl DPPH and 2,2'-azino-bis (3- ethylbenzothiazole-6-sulfonic acid) ABTS and compared with that of Trolox.

From the results obtained from the antioxidant activity of the synthesized compounds it can be concluded that the introduction of a substituent such as the phenyl group at N (4), in the thiosemicarbazide backbone, leads to biologically active compounds. The antioxidant properties are amplified with the substitution at N (1), by introducing the carbonyl fragment. All the synthesized compounds showed higher values than the control sample, against the radical cations ABTS and the radical DHHP. They are in the next phase of testing for use as medicines.

№	Name of new compounds	ABTS•+ radical cation scavenging activity IC ₅₀ , μM/L	DPPH• radical scavenging activity IC ₅₀ , μM/L
1	<i>N</i> -(2-methylphenyl) hydrazinecarbothioamide	16,2	32,1
	2-(2-hydroxy-3-methoxybenzylidene)- <i>N</i> -(2-methylphenyl) hydrazinecarbothioamide	15,2	12,6
2	<i>N</i> -(2,4-dimethylphenyl) hydrazinecarbothioamide	15,3	34,2
	2-(2-hydroxy-3-methoxybenzylidene)- <i>N</i> -(2,4-dimethylphenyl) hydrazinecarbothioamide	11,2	28,5
3	<i>N</i> -(2,4,6-trimethylphenyl) hydrazinecarbothioamide	14,4	31,4
	2-(2-hydroxy-3-methoxybenzylidene)- <i>N</i> -(2,4,6-trimethylphenyl) hydrazinecarbothioamide	11,8	43,8
4	Trolox/ control sample	26,3	48,9

Keywords: antioxidant properties, DPPH• radical scavenging activity, free radicals, N4-n-methylphenylthiosemicarbazides.

NATURAL DYES FROM POKEWEEED BERRIES: EXTRACTION PROCEDURES AND STABILITY

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Natural dyes extracted from berries of pokeweed (*Phytolacca americana*) were used as colorants in food and textile industries because of their nontoxic and ecofriendly characteristics, as nutraceuticals with antioxidant properties and sensitizers for dye-sensitized solar cells. Chemical structure of natural dyes from pokeweed berries belong to the betalains class, namely to betacyanins that provide the red-violet color and betaxanthins with yellow-orange color. Complexity of betalains extraction consist in poor stability of natural dyes and depends on various factors (temperature, pH, and concentration of dye, water activity, light and UV radiation, metal chelators, mordant and pretreatment of raw materials). The purpose of this work was to describe the different approaches to optimize the extraction technique for obtaining the natural dyes from pokeweed berries in stable form based on previous researches. The cheapest methods of dyes obtaining is extraction using different solvent systems (water-alcoholic, weak acid solutions, buffer solutions, etc.) by liquid-solid extraction in ratio 5:1. Our studies were shown that water extracts from pokeweed berries contained 5.5 times more betalain colorants when compared to ethyl alcohol (95%) extracts. While it is necessary to take into account that colorants from pokeweed maintain the stability in limits of pH from 3.0 to 7.0. The optimal pH for maximum stability of betalains is 5-6. To enhance the betalain extraction yields, the different methods of pretreatment were proposed, such as pulsed electric fields, gamma irradiation, and low-direct current electric fields, ultrasonic or microwave assisted extractions. Their advantages consist in processing at low temperatures due to the high sensitivity of betalains to heat, which decompose at temperatures above 50°C. However, in our studies it was determined that the concentration of natural dyes in pokeweed berries did not change during the year of storage at a temperature of -12-16°C. Encapsulation of extracted betalains with polysaccharides (pectin, guar gum) as the wall material led to increased stability and longer shelf life of natural dyes. Based on the described approaches and our data, the developed technological procedure for obtaining and stabilizing of natural dyes from pokeweed berries will be proposed for patenting as a food colorant.

Acknowledgments: The authors are grateful to the International Visegrad Fund for the support of this research by grant #52210146.

Keywords: betalains, extraction technique, natural dyes, optimal pH, *Phytolacca americana*, stabilizing, obtaining.

SYNTHESIS OF NEW POTENTIAL ACTIVE HOMODRIMANE SESQUITERPENOIDS WITH BENZIMIDAZOLE FRAGMENT

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Ciocarlan Alexandru, Aricu Aculina

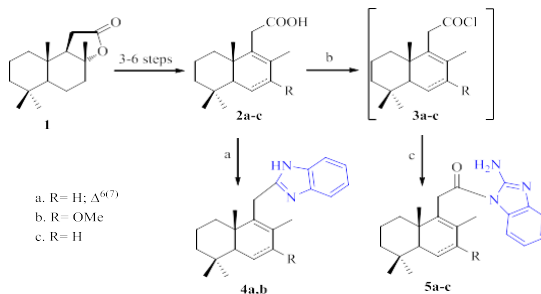
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Sesquiterpenoids, including those with a homodrimane skeleton, are natural compounds with a wide range of biological activities. Furthermore, the benzimidazole and its derivatives are considered interesting heterocycles since they possess important pharmacological activities. For this reason, synthesis of terpene compounds with heterocyclic fragments, especially benzimidazoles is a perspective direction of research in the field of biologically active compounds.

As starting material for the synthesis of title compounds 11-homodrim-6(7),8(9)-dien-12-oic acid **2a**, 7 α -methoxy-11-homodrim-8(9)-en-12-oic acid **2b** and 11-homodrim-8(9)-en-12-oic acid **2c**, previously obtained from commercial (+)-sclareolide **1** in 5, 3 and 5 steps, were used. After that, two methods of synthesis were proposed. The first method consists of interactions of one of the acids **2a,b** with *o*-phenylenediamine in the presence of triphenylphosphine (Ph₃P) and triethylamine (Et₃N), giving desired compounds **4a,b**. The second method includes the interaction of 2-aminobenzimidazole with one of the acid chlorides **3a-c** generated *in situ* from acids **2a-c**, giving desired compounds **5a-c** (Scheme).

The structure of all obtained compounds has been established using modern methods of analysis (ATR-FTIR, ¹H, ¹³C and ¹⁵N RMN spectroscopy).



Scheme. Synthesis of terpeno-benzimidazole hybrid compounds.

Reagents and condition: a. *o*-Phenylenediamine, Ph₃P, Et₃N, CCl₄, Δ , 4h, 46-57%.
b. (COCl)₂, C₆H₆, r.t. 1h, Δ , 1h; c. 2-Aminobenzimidazole, CH₂Cl₂, r.t. 3h, Δ , 4h, 49-85%.

Acknowledgements: This work is part of the project PLANTERAS 20.80009.8007.03 within the State Program (2020-2023) financed by the National Agency for Research and Development.

Keywords: biologically active compounds, pharmacological activities, sesquiterpenoids.

IRON(III) CLUSTERS WITH 3-FORMYLSALICYLIC ACID

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In the presence of additional ligands and polar solvents, the well-known μ_3 -OXO bridged triangular iron carboxylates easily reorganize and stabilize large complexes. We chose 3-formylsalicylic acid (H₂L) in order to explore its coordinative ability, due to its multiple coordination donor groups like –COOH, –OH and –CHO. Herein, new hexanuclear iron(III) clusters $[\text{Fe}_6\text{O}_2(\text{OH})_2(\text{L})_2(\text{H}_2\text{O})_2(\text{O}_2\text{CR})_8] \cdot \text{MeCN}$ (R = CMe₃ (**1**); CHMe₂ (**2**)), where L is the dianion of 3-formylsalicylic acid, are reported. Compounds **1** and **2** were prepared from the reaction of μ_3 -oxo trinuclear iron(III) precursors $[\text{Fe}_3(\mu_3\text{-O})(\text{H}_2\text{O})_3(\text{O}_2\text{CR})_6](\text{O}_2\text{CR}) \cdot 2\text{HO}_2\text{CR}$ with 3-formylsalicylic acid in acetonitrile. Both structures comprise six Fe atoms in an almost planar arrangement that can be described as two oxo-centered triangular units $[\text{Fe}_3(\mu_3\text{-O})]^{7+}$ joined together by two bridging hydroxide and two bridging carboxylate groups. The asymmetric units contain only half of an Fe₆ molecule. All Fe atoms adopt distorted octahedral coordination geometries and are in the +3 oxidation state. The peripheral ligation of metal ions is completed by six carboxylate molecules and two aldehyde ligands which are in their monoanionic and dianionic forms, respectively. The two 3-formylsalicylic ligands act as tridentate, bridging Fe1 (Fe1') and Fe2 (Fe2') ions by the alkoxo groups within each $[\text{Fe}_3(\mu_3\text{-O})]^{7+}$ unit. The oxygen atom of the formyl group is also coordinated, which was not reported so far for this ligand. All carboxylate groups adopt the bridging $\mu_2\text{-}\eta^1\text{:}\eta^1$ coordination mode: two of them join the edges of the triangular $[\text{Fe}_3(\mu_3\text{-O})]^{7+}$ units and the remaining six link Fe atoms within the latter (Fig. 1). Detailed characterization of compounds **1** and **2** will be described in a forthcoming paper.

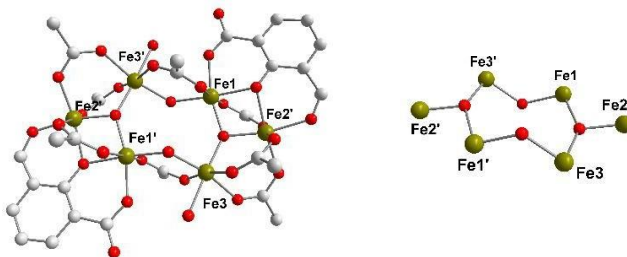


Figure 1. The molecular structure of complex **2**·CH₃CN (left) and its $[\text{Fe}_6(\mu_3\text{-O})_2(\mu\text{-OH})_2]^{12+}$ structural core (right).

Color code: Fe^{III}, olive green; O, red; C, grey. The hydrogen atoms, methyl groups and solvent molecules are omitted for clarity.

Keywords: 3-formylsalicylic acid, asymmetric units, hexanuclear iron(III) clusters, peripheral ligation.

SYNTHESIS OF SOME THIOSEMICARBAZONES DERIVED FROM 1-PYRIDIN-2-YL-3-PHENYL-PROP-2-EN-1-ONES (CHALCONES) SUBSTITUTED IN THE PHENYL RING

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Thiosemicarbazones are organosulfur compounds with the general formula $H_2NC(S)NHN=CRR$, which possess a wide spectrum of medicinal properties, they are studied for their antibacterial, antiviral and antifungal activities. Because of these activities thiosemicarbazones have been clinically tested for a variety of diseases, such as tuberculosis, viral infections, malaria and cancer. Also, their complexes with metals show a wide range of biological activities. Chalcones or 1,3-diarylprop-2-en-1-ones belong to the flavonoid family, in which the aryl rings are linked by an α - β unsaturated ketone moiety. Their wide distribution in nature, as well as the simple methods of synthesis and the accessibility of the precursors has always encouraged the investigation of the possibilities of their application in medicine due to their activity: antimicrobial, anti-inflammatory, antimalarial, antitumor. Although there is a multitude of thiosemicarbazones in the literature, those derived from chalcones are very little studied. And the known structures show serious potential to be studied as biologically active substances.

Thiosemicarbazones of chalcones were obtained by the following method (Figure 1): to the molar equivalent of chalcone and thiosemicarbazide diluted in alcohol, a few drops of HCl are added to catalyze the reaction. The reaction mixture is vigorously stirred for 2-3 hours at temperatures between 35-45°C. The obtained solids were filtered and dried to be subjected to their physical and chemical studies. The yields are between 50-80%.

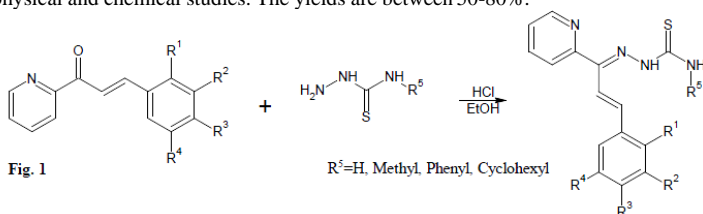


Fig. 1

The listed chalcones were obtained by aldol condensation (Fig.2) which is a synthesis within the field of green chemistry, where a solution of NaOH or Na_2CO_3 is added dropwise to the mixture of 2-acetylpyridine and the appropriate aldehyde in alcohol. The reaction mixture is stirred from 1 to 6 hours at room temperature. A solid is obtained, which is filtered, washed with water, then recrystallized from alcohol.

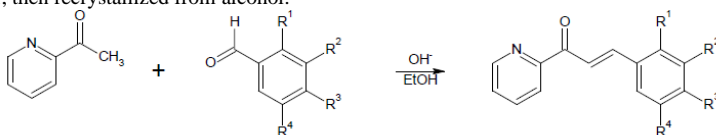


Fig. 2

I. $R^1=R^4=OCH_3$, II. $R^2=R^3=OCH_3$, III. $R^2=R^3=R^4OCH_3$

Keywords: Aldol condensation, biological activities, medicinal properties, thiosemicarbazones.

MICROMORPHOLOGICAL AND ANATOMICAL CHANGES INDUCED BY SALINE STRESS IN *OCIMUM BASILICUM* L. (VAR. GENOVESE), IN EXPERIMENTAL GROWTH CONDITIONS

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Today, saline stress is a major concern in the agricultural field, generating numerous research. Increased salinity inhibits the uptake of nutrients by plants and decreases water quality, causing ionic toxicity, nutritional disorders and osmotic stress and limits agricultural production, thus becoming a global problem^{1,2,3,4,5}. The present study aims at the adaptation and resistance of basil to different degrees of salinity, a taxon of interest in research to identify plants capable of supporting and developing in conditions of high salinity^{3,6,7}. The biological material used in the experiment was obtained from basil seeds, *Ocimum basilicum* L. (var. Genovese), purchased commercially, grown under experimental laboratory conditions.

A peat-based substrate was used for germination, and when the plant individuals reached dimensions of about 4 cm, they were transplanted into pots with universal soil, the saline treatment being applied after the appearance of the first true leaves.

After consulting the literature, in the present experiment were used 3 concentrations of sodium chloride, considered high concentrations for plant cultivation: 100 mM, 150 mM and 200 mM, the saline solutions being administered in 3 repetitions. At the end of the cultivation interval, micro-morphological and anatomical determinations were performed at the root, hypocotyl, epicotyl and leaves, to identify any morphological and structural changes caused to these organs by artificially induced saline stress.

Micro-morphological analyses showed, at the level of the lower and upper surface of the leaves, a decrease in the number of secretory hairs with the increase of the salt concentration in the cultivation substrate. At the same time, the number of stomata showed a similar evolution, but it was observed that, with the decrease of the number of stomata (more accentuated effect at high NaCl concentrations), their surface area increased. From an anatomical point of view, an increase in the number of conducting vessels was observed in plants grown on substrate with high concentrations of NaCl. At the root level, a premature suberification was observed, a phenomenon that leads to the loss of water absorption properties and is also confirmed by the increased number of secondary roots formed. This can be interpreted as a response of test plants to physiological drought caused by experimentally induced saline stress⁸.

The ability of basil to respond to saline stress and counteract its effects through various defence mechanisms make this taxon a potential candidate for cultivation on saline soils.

Further investigations are needed at different stages of plant development, to obtain a clearer picture of the effects of salinity on the development and adaptation of basil to conditions of saline stress.

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Keywords: stomata, *Ocimum basilicum*, physiological drought, resistance, saline stress, suberification.

CHEMICAL COMPOSITION OF DAMASK ROSE (*ROSA DAMASCENA* MILL.) CONCRETE OF MOLDAVIAN ORIGIN

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The Damask rose (*Rosa damascena* Mill.) is an important aromatic plant and one of the most important rose species used for commercial production of rose essential oil, rose water, concrete and absolute. These products are widely used in perfumery and cosmetics, foodstuffs, as well as in pharmaceutical industry. It was reported, that individual rose products or some rose preparations possess antimicrobial, antioxidant, relaxant, anti-inflammatory, insecticidal, anti-HIV and hypnotic properties, and are frequently used for treatment various diseases [1-3].

The main World producers are Bulgaria and Turkey, followed by France, Italy, Ukraine, Russia, Morocco, Egypt, Libya, Iran, China and India. In the former USSR, Moldova was the major producer among the Soviet republics. Currently, here are only a few small growers and rose processing plants [1-3].

The chemical composition of rose essential oil and rose water has been well studied. In opposite, the rose concrete composition is less studied. For this reason, herein the chemical composition of rose concrete of Moldavian origin is reported.

The concrete was obtained industrially in 2020 by extracting freshly collected rose petals (Pervomaisc village, Causeni district) with petroleum ether followed by complete distillation of the solvent under reduced pressure. Its overall yield was of about 0,2% recalculated for starting material. Next, the concrete was subjected to GC-MS analysis on an Agilent 7890B system, equipped with VF-WAXms 30m x 250µm x 0.25µm column during 30 minutes.

About 44 constituents of rose concrete, which belong to several groups of compounds were identified. The major constituents are as follows: monoterpene alcohols - citronellol (2.06%), nerol (2.99%) and geraniol (5.62%); monoterpene pyrans - rose oxides (0.44%); higher hydrocarbons: nonadecane (1.26%), heneicosane (1.78%), tricosane (3.09%) and aromatic alcohol 2-phenylethyl alcohol (60.42%).

The rose concrete is a valuable substance, and the quality of the product that can be obtained from Moldovan plantations should be maintained and controlled.

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Keywords: Cchemical composition, citronellol, extracting, geraniol, nerol, *Rosa damascene*, rose essential oil.

SYNTHESIS AND STUDY OF Zn TETRA-SUBSTITUTED PHTHALOCYANINE WITH CHALCONIC GROUPS

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The chalcones are considered derivatives of flavonoids and izoflavonoids, which have in their structure aromatic rings connected through a fragment of prop-2-en-1-one. The modification of aromatic rings by different heterocyclic rings creates a wide variety of new substances with a large spectrum of proprieties which can be used in pharmaceutical industry and the production of new medicines. The possibility to play with the chaconnes structures favors the obtaining of pharmaceutical products with low toxicity and high pharmacological action [1]. The chalcones are spread in plants and possess a wide spectrum of uses, due to their antioxidants properties, antibacterial, antifungal, insecticides and anticancer [2].

The antimicrobial resistance is one of the most important threats for our health. This requires efforts to discover new ways to completely eliminate these microbial pathogens. The use of photosensitizes and light is a complementary way to fight against pathogenic microbes. Nowadays, the metalphthalocyanins are intensive studied due to their structures which are rich in π conjugated systems, and have potential applications in various technological areas as optoelectronic devices, gas sensors, static induction transistors and photo receptors in laser beam printers [3].

In the literature are described a series of chalconic derivatives transformed into complexes with non-transitional metals. This type of products is wide studied by the fact that they possess more functional groups which allow the chemical connection with metals, modifying in this way the spectral and medicinal properties.

Also, are known syntheses methods of Zn and Co(II) metalphthalocyanins which has as substituents chalconic fragments. These products are obtained through the following steps: a) syntheses of chalcones with –OH groups by Claisen-Schmidt method, using the KOH as a base and ethanol as solvent; b) the interaction of the respective chalcones with 4-nitrophthalonitrile in the presence of K_2CO_3 , nitrogen atmosphere and DMSO for obtaining tetrasubstituted phthalocyanine with chalconic fragments; c) treatment of tetrasubstituted phthalocyanine which contains chalconic fragments with $ZnCl_2$ and $CoCl_2$ salts to give metalphthalocyanins and as catalyst is used the N,N-dimethylethanolamine [4].

The aim of this study is the synthesis of Zn tetrasubstitutedphthalocyanine with chalconic groups by the following scheme:

Zn Tetranitrophthalocyanine (1) was synthesized from 4-nitrophthalonitrile. The formed mixture of 4-nitrophthalonitrile, $Zn(CH_3COO)_2$ and DBU (1,8-diazobicyclo [5.4] undec-7-ene) was refluxed in pentan-1-ol for 8 hours. For purification (1) is treated with HCL (5%) at the temperature of 95 °C for 30 min. Zn tetranitrophthalocyanine (1) is treated with Na_2S aqua solution to give Zn tetraaminophthalocyanine (2). The purity of (2) was verified by thin layer chromatography on silica gel, eluent chloroform: tetrahydrofuran 6:1 [5]. 3-(4-Dimethylamino) phenyl-1-(4-isothiocyanatophenyl) prop-2-en-1-one was synthesized from 4-dimethylaminobenzaldehyde and 4-isothiocyanatoacetophenone, as catalyst concentrated HCL at 45 °C for 2 hours [6]. The presence of –NH₂ group in (2) gives the opportunity to chemically connect the 3-(4-dimethylamino) phenyl-1-(4-isothiocyanatophenyl) prop-2-en-1-one (3) with Zn tetraaminophthalocyanine (2) to give tetrasubstituted Zn phthalocyanine with chalconic groups. The reaction mixture formed from 3-(4-dimethylamino) phenyl-1-(4-isothiocyanatophenyl)prop-2-en-1-one (3) and Zn tetraaminophthalocyanine (2) 4:1 ratio, is

dissolved in DMF and heated to 60-65 °C for 5 hours. The reaction product is obtained with a yield of 57%. For obtained compounds IR analysis was performed.

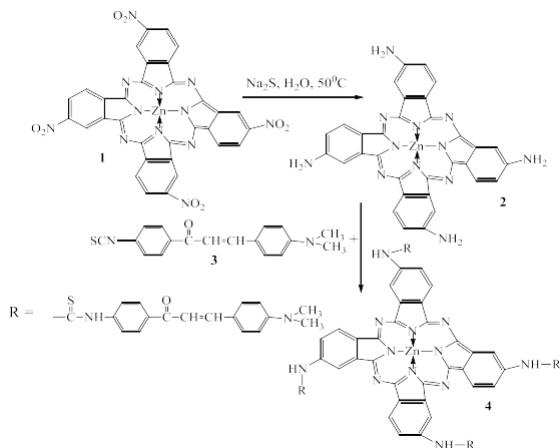


Fig. 1. Synthesis of Zn tetra-substituted phthalocyanine with chalconic groups

The synthesis of one Zn tetrasubstitutedphthalocyanine with chalconic groups was realized and the presence of functional groups was confirmed by IR spectra. These types of compounds can be used as fluorescent substances. The fluorescent characteristics are due to the presence of chalconic groups.

Acknowledgement: This work was supported by the Ministry of Education, Culture, and Research of Republic of Moldova, research grant 20.80009.5007.16.

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Keywords: antimicrobial resistance, chalcones, pharmaceutical industry, Zn tetrasubstitutedphthalocyanine.

DESIGNING OF THE BEST SLEEP PROTOCOL ACCORDING TO THE PSYCHOPHYSIOLOGICAL NEED

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While many of us think of sleep as a stretch of time where nothing happens, sleep is actually, at least neurologically. Although the importance of sleep cannot be disputed, scientists do not know exactly why so important to our survival. A rested body is much stronger and can defend itself better against infections and colds. Chronic sleep deprivation can lead to long-term mood disorders, such as depression and anxiety. Scientists have found that people who usually sleep less than 5 hours a night have a increased risk of developing diabetes. Lack of deep sleep changes the way the body produces glucose. Increases sexual appetite and decreases the risk of heart disease. Lack of sleep for long periods of time is associated with an increased level of blood pressure, rhythm increased heart rate and higher levels of certain chemicals that increase the risk of inflammation.

Considering literature data indicating psychophysiological variations during and due to sleep, the studying methods for this experiment are:

1. Surveys and psychological tests.
2. Polysomnography.
3. Hormone levels lab results.

A survey concludes that people who sleep less of 6 hours a night, or sleeping more than 9 hours, had a 30% higher death rate than those who usually sleep 7 - 8 hours. Even those who slept less than 6 hours and generally did not had health problems but had a 1.8 times higher death rate than those who slept "normal" hours.

In January 2017, a study was made public according to which insufficient sleep exposes the body to diseases by weakening the immune system.

The immune system works best when has enough sleep. Seven or more hours of rest are recommended for optimal health. Adolescents, in March 2016, showed that one in ten children are at risk of breathing disorders in sleep time. 9.6% of children in Romania are at risk for breathing disorders during sleep, of which the most severe form is obstructive sleep apnea. Children who are at risk for this problem can have signs during the night or during the day.

Humans have an internal circadian rhythm, a routine and behavioral processes that occur approximately every day for 24 hours. Despite this fact, what is the right time for a person to sleep, we know which one is the best and healthier type of sleep for a person's overall health.

Keywords: behavioral processes, circadian rhythm, disorders, optimal health, sleep protocol.

COPPER COMPLEXES WITH N⁴(2-ETHYL BENZOATE) THIOSEMICARBAZONE OF 2-ACETILPYRIDINE

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Thiosemicarbazones are an important class of organic compounds that have attracted attention in the pharmaceutical industry due to their high biological activity such as antibacterial, antifungal, antiviral, antimalarial, antitumor and others. The paper presents the synthesis of new ligand and coordination compounds of Cu(II), with N⁴(2-ethyl benzoate) thiosemicarbazone of 2-acetylpyridine (**3**) obtained through the addition of ethyl 2-isothiocyanatobenzoate (**2**) with 2-(1-hydrazinylideneethyl)pyridine.

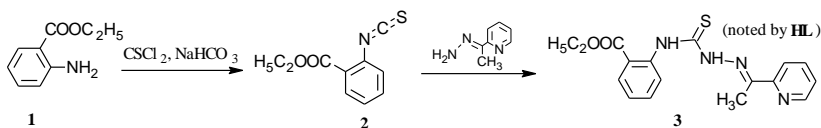
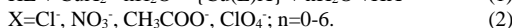
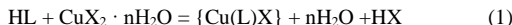


Fig. 1. Synthesis of N⁴(2-ethyl benzoate) thiosemicarbazone of 2-acetylpyridine (**3**)

Based on the synthesized thiosemicarbazone HL, five coordination compounds were obtained by mixing, the salts of Cu(II) solutions in ethanol and ligand solutions in ethanol ratio of 1:1, reflux for half an hour. Obtained as a green microcrystalline complexes according to the reaction written below.



The characterization of new compounds was done by ¹H NMR, ¹³C NMR, IR spectroscopy, conductivity and elemental analysis. In addition, the structures of the ligand (HL) and complexes [Cu(L)NO₃], [Cu(L)H₂O]₂·2ClO₄·4H₂O were determined by single-crystal X-ray diffraction.

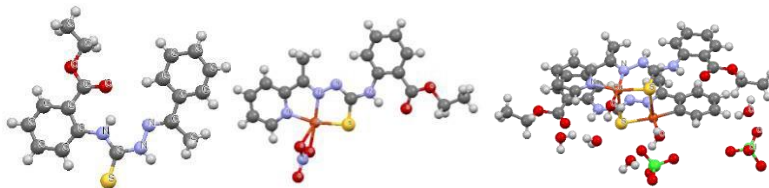


Fig. 2. Crystalline structure of ligand (HL), coordination compound [Cu(L)NO₃] and [Cu(L)H₂O]₂·2ClO₄·4H₂O

The effect of ligand and complexes on antioxidant activity were studied. The best result was found to be on ligand, with IC₅₀ = 22.51 μM, it is better than the substance Trolox, IC₅₀ = 33.33 μM used in medicine.

Keywords: antioxidant activity, thiosemicarbazones, IR spectroscopy, ligand.

AUTOPHAGY ROLE IN MODULATING INFLAMMATORY MARKERS RESPONSE IN KERATINOCYTES

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Autophagy is involved in a wide range of cellular physiological processes, including the response immune, tumorigenesis, differentiation, apoptosis, antimicrobial defense, etc. Impaired autophagy is associated with a wide range of human diseases, such as diseases neurodegenerative, cardiomyopathies, infectious diseases and cancers, and autophagy modulation was already considered a potential target for the therapy of these diseases.

Modulation of the autophagy mechanism could be a better strategy than inhibition of melanin synthesis or transfer for treating disorders of skin pigmentation. Moreover, inhibition of autophagy increased significantly accumulation of p62, expression cathelicidin/LL-37 and inflammatory cytokine production. The effects of autophagy on chronic skin disorders will be studied, the everyday factors that modulates the immune response in epithelial cells and possible therapeutic alternatives in expression of inflammatory markers in keratinocytes under the action of autophagy mechanisms, induced by fasting and pharmacological simulation.

Considering literature data indicating the anti-inflammatory action of some medicinal plants, the studying methods for this experimental study are:

1. Obtaining aqueous and hydroalcoholic plant extracts.
2. UV-Vis spectrophotometric analysis of the extracts and determination of the content in phenolic compounds.

Studying the role of autophagy in the human body and the mechanisms which it participates in, such as the evolution of tumors. The obtained results demonstrate that in the initial stage in the process of oncogenesis, autophagy has the role of tumor growth suppressor, however, in the late stage a oncogenesis, contributes to the development of the tumor process by providing the cells with a substrate for synthesis of ATP and macromolecules, inhibiting anoikis. Respectively, mutations in a number of genes from the reaction cascade of the autophagy involve expression products that are indispensable. Currently, have been discovered chemicals capable of activating/inhibiting autophagy activity.

Developing new positive modulators of autophagy opens a new perspective in making more effective remedies for fighting chronic skin diseases.

Keywords: autophagy, mechanism, modulators of autophagy, tumor growth suppressor.

OPTIMIZATION OF THE EXTRACTION PROCESS OF BIOACTIVE COMPOUNDS FROM PEACHES

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Peach fruits have long been promoted for their ability to prevent various diseases, due to phytochemicals that include carotenoids, phenolic acids, organic acids, micro- and macroelements. In the Republic of Moldova, peach fruits are widely used both fresh and for processing in order to obtain a wide range of canned products. Pomace obtained as a result of peach processing, is rich in polyphenols, tannins, aromatic substances, etc., is of interest for subsequent exploitation in the food industry. The aim of this paper is to optimize the extraction process of bioactive substances from peach pomace in order to obtain the extract for use in the formulation of novel foods with high biological value.

Peach pulp dried at a temperature of 60 ± 1 °C and ground to a granularity of 70μ was used for research. To determine the optimal volume of solvent for the extraction of water-soluble compounds, distilled water, ethyl alcohol 96 % (v/v) and 50 % (v/v) hydroalcoholic solution were used. The extraction was performed at a temperature of 70 ± 2 °C with different volumes: 8, 12, 16, 20, 24 mL. The water-soluble substance content of the extract was determined by the refractometric method. The extraction of water-soluble substances was performed until the equilibrium concentration was reached. In the extracts obtained at optimal volumes of solvent, the total polyphenol content (TPC) was determined by the Folin Ciocalteu method, and the antioxidant activity (AA) of the extracts was assessed by reaction with the radical DPPH.

The results of the research showed that the best extraction in water took place when applying the solvent volume of 12 mL, in the case of the hydroalcoholic extractant of 50% (v/v) the most effective was the volume of 20 mL, and in the alcoholic solvent of 96% (v/v) - the volume of 16 mL was the most efficient. The study of the extraction kinetics of water-soluble substances from peach pomace showed that the degree of extraction is higher at first, later showing a slowing trend. Distilled water has been shown to contribute to better tissue separation and rupture of cell walls of peach pulp, facilitating the diffusion of water-soluble compounds. It was found that solvents have a major influence on the TPC, as follows: the lowest polyphenol content was recorded in the case of extraction in ethyl alcohol 96% (v/v), with a value of 13.84 mg GAE/g plant. In the case of 50% hydroalcoholic solution and distilled water, TPC increased approximately 6.4 times. It is known that polyphenols are responsible for the antioxidant activity in plant extracts. It has been shown that the extracts with the highest TPC are characterized by the highest value of AA. In the case of our extracts the AA varies between 63.69 % (ethyl alcohol of 96% (v/v)) and 71.13% - 50% (v/v) hydroalcoholic solution. Thus, it has been shown that the type of solvent can influence the extraction yield of polyphenolic compounds and antioxidant activity.

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Keywords: bioactive substances, extraction process, food industry, peaches.

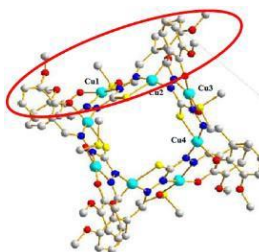
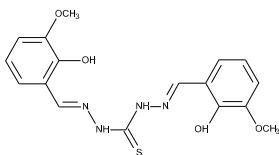
Cu(II) COMPLEXES WITH 1,5-BIS(2-HYDROXY-3-METHOXYBENZYLIDENE) CARBOHYDRAZIDE

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This communication shows that the nature of anions of the corresponding copper salts, as well as their coordination properties are essential in determining the final structures of the complexes with the ligand 1,5-bis(2-hydroxy-3-methoxybenzylidene)carbohydrazone (H_4L). Thus, by its interaction with copper(II) chloride, binuclear compound $[Cu_2(H_2L_{cycl})(CH_3OH)_2Cl_2] \cdot CH_3OH$ (**1**), was obtained, where cyclization of the thiocarbohydrazone ligand occurs with the formation of a 1,3,4-thiadiazole ring. It should be mentioned that the methoxy group plays the role of a bridge between copper ions [1]. By the interaction of the same ligand with copper(II) acetate in DMSO, octanuclear complex $[Cu_8L(DMSO)_8]$ (**2**) was prepared. The organic ligand keeps its original form, but it is totally deprotonated, behaving as a tetrabasic acid, and the methoxy group is not involved into coordination. The composition and the structure of compound **2** was determined using elemental analysis, IR spectroscopy and single crystal X-ray diffraction study. In the complex **2** it can be highlighted the neutral binuclear unit Cu_2L composed of the bicompartamental ligand that hosts two copper ions. A nitrogen atom of this fragment is located in the coordination sphere of another copper atom of the adjacent dimeric fragment. As a result, all 7 donor atoms of the organic reagent are involved in coordination, forming an octanuclear metalomacrocyclic compound. Among four independent copper(II) ions, two from the ONS donor atoms set have a square planar geometry, while the coordination polyhedra of another two from the ONN donor atoms set can be described as a distorted trigonal bipyramid.



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Keywords: copper salts, crystal X-ray diffraction, methoxy group, 1,5-bis(2-hydroxy-3-methoxybenzylidene), IR spectroscopy.

CU(II) COMPLEXES WITH 4-ALLYLTHIOSEMICARBAZONE AS POSSIBLE ANTIOXIDANT AGENTS

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Oxidation is an essential part of aerobic metabolism in living organisms. In this process, free radicals are constantly generated. It is widely known that free radicals play a dual role *in vivo* as both beneficial and adverse compounds. In order to maintain the balance of the activity of the free radicals in living cells, the organism has evolved antioxidant defense systems, protecting against free radical damage. However, when our endogenous defense system has incomplete efficiency, or a rise in free radicals under special conditions like smoking, chemical air pollutants, radiation, and inflammation, the imbalance between free radicals and antioxidants results in oxidative stress, which is now believed to be one of the important factors in the occurrence of certain human diseases including atherosclerosis, rheumatoid arthritis, cancer, and neurodegenerative diseases. So, the search of possible antioxidants agents is of interest, because they protect the body against damage by harmful free radicals. For this purpose two copper(II) coordination compounds of 1-(morpholin-4-yl) propane-1,2-dione 4-allylthiosemicarbazone (HL) with 1,10-phenantroline (1,10-Phen) and 2,2'-bipyridyl (2,2'-Bpy) were synthesized. On the first stage of the experiment the ethanolic solution of $\text{Cu}(\text{NO}_3)_2 \cdot 3\text{H}_2\text{O}$ reacts with 4-allylthiosemicarbazone HL and on the second stage the ethanolic solution of the prepared precursor $\text{Cu}(\text{L})\text{NO}_3$ reacts with bidentate amines forming crystalline coordination compounds. The composition of these compounds was determined using

elemental analysis for copper: $\text{Cu}(1,10\text{-Phen})(\text{L})\text{NO}_3$, $\text{Cu}(2,2'\text{-Bpy})(\text{L})\text{NO}_3$. The molar conductivity values of the synthesized coordination compounds are in the range $82\text{-}98 \Omega^{-1}\cdot\text{cm}^2\cdot\text{mol}^{-1}$ that indicates that these complexes are 1:1 electrolytes. A standard ABTS⁺ method has been used to determine the antioxidant properties of synthesized compounds and trolox was used as a reference. The pro-ligand HL is less active than trolox, its coordination to copper(II) atom decreases antioxidant activity. The introduction of bidentate amines into the internal sphere of precursor increases the activity of complexes, that become 4 times more active than pro-ligand HL, 6 times more active than precursor, and also more active than trolox.

Table 1. IC₅₀ values of the synthesized substances toward ABTS⁺ radical cation.

Compound	IC ₅₀ , μM
HL	94.44
Cu(L)NO ₃	140.30
Cu(1,10-Phen)(L)NO ₃	23.04
Cu(2,2'-Bpy)(L)NO ₃	22.31
Trolox	33.00

Acknowledgments: This work was fulfilled with the financial support of the ANCD projects 20.80009.5007

Keywords: antioxidants agents, Copper(II) coordination compounds, harmful free radicals.

**SYNTHESIS OF NEW *EPI*-MANOYLOXIDE DERIVATIVES WITH AZIDE
AND *gamma*-LACTAM FUNCTIONAL GROUPS**

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Manoyloxides have the carbon skeleton of the natural diterpenoid forskolin, which is a relevant secondary metabolite isolated from *Coleus forskohlii* with a myriad of therapeutic activities based on its ability to penetrate the cell membranes and stimulate the enzyme adenylyl cyclase. Other natural products belonging to the *ent*-series like ribenol and varodiol share the same tricyclic structure of manoyloxides and both have been reported in structure-activity relationship studies, providing derivatives with antimicrobial properties. These facts prompts following synthetic studies aimed to the generation of other manoyloxides derivatives with more and diverse functional groups.

We report in the current communication the free radical carboazidation of epimeric manoyloxide leading to azide-functionalized epimanoyloxide and converted successfully to a related *gamma*-lactame. The lactamization step occurred spontaneously on catalytic hydrogenation of carboazidation product. The structure of the title product was demonstrated by spectroscopic means and showed a flexible nitrogen-centered chirality, which is characteristic for such hybrid heterocycles.

The *gamma*-lactame is regarded as a surrogate of GABA derivatives and proved to be an efficient pharmacophore in our recent studies. Following investigations are in progress in order to reveal the cytotoxicity mechanism displayed by similar terpenic *gamma*-lactams.

Keywords: antimicrobial properties, *gamma*-lactame *Coleus forskohlii*, manoyloxides, spectroscopic methods,

PHENOLIC COMPOUNDS AND ANTIOXIDANT ACTIVITY OF SOME MOLDOVAN WHITE AND RED GRAPE VARIETIES AT DIFFERENT MATURATION PERIODS

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Viticulture, along with the winemaking, is the main food industry of Republic of Moldova, whose development is in a close relation with the country's economy [1]. Annually, hundreds of thousands of tons of grapes [2] are used to produce wines, juices, jams etc. – a job in which 25% of the working population of the country is involved [1].

Grapes are rich sources of different classes of polyphenols that possess antioxidant activity and are benefic for the humans' health. At different maturation periods, the content of the polyphenols in grapes varies within the grapes' tissues due to the biochemical processes that occur during the development of the berries [3].

In this study, the polyphenolic content and the antioxidant activity was determined in two white grape varieties of new selection, Viorica and Riton, and two red grape varieties, Feteasca Neagra and Copceac. All tests were accomplished at three different maturation periods: in unripe grapes, at the veraison and at the ripening. Various eco-friendly extraction methods were used in order to ensure the extraction of the grapes' polyphenols and antioxidants from the constituent parts of the berries: squeezing, ethanol – HCl extraction or thermal extraction. Spectral and analytical methods were applied for the qualitative determination of the particular groups of phenolic compounds. Thereby, the anthocyanins were analyzed by the pH differential method [4], the presence of the proanthocyanidins was established by using the DMACA reagent [5] and total phenolic content was determined through the Folin – Ciocalteu method [6]. The antioxidant assay was performed by utilizing the DPPH free radical [7], thanks to the accessibility and reproducibility of the method. A good correlation between the polyphenolic content and antioxidant activity of the grape extracts was established.

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Keywords: antioxidant activity, phenolic compounds, grape varieties, grape extracts.

COPPER COMPOUNDS AS STRESS FACTORS AND REGULATORS IN PHYCOBIOTECHNOLOGY

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Metals are part of the structural component of the active centers of key enzymes involved in the reactions on which life is based. In the course of evolution, living organisms have developed mechanisms that allow them to acquire metals from the environment. The vast majority of metals with a known biological value belong to the category of microelements. The existence of natural habitats with a high content of metals, and recently anthropogenic pollution of the environment with metals, have led to the emergence of mechanisms for protecting cells from the harmful effects of increased amounts of these chemical elements.

Microalgae and cyanobacteria, especially freshwater ones, in their natural environment are deficient in essential metals and have effective mechanisms for their capture and active transport into cells. Under technological conditions, the replenishment of the nutrient medium with metals in various forms is an effective method for controlling cellular biosynthetic processes. At the same time, metals, especially those with variable valence, are a source of free radicals that can threaten both the biotechnological process and the quality of biomass. The effect depends not only on the amount of metal, but also on the chemical form in which it is present.

In the case of biotechnologies involving the cyanobacterium *Arthrospira platensis* (spirulina), this can be demonstrated using copper as an example. Copper is an important component of more than metalloenzymes, including superoxide dismutase and cytochrome c oxidase. At the same time, being in the ionic form, even at a concentration of 2.5 mg/l, copper disrupts the growth of the culture, and at a concentration of 0.1 mg/l causes serious changes, expressed in the deterioration of the biomass quality, a decrease in the activity of antioxidant enzymes, and accumulation of products of oxidative degradation of biopolymers.

As a structural part of the coordinating compounds, copper is tolerated by the spirulina culture in much greater amounts, even providing a stimulatory effect, the extent of which is determined by both the amount of metal and the nature of the coordinating agent used in the synthesis of compounds. Thus, using copper in the composition of the compound in an amount of from 1 to 20 mg/l, it is possible to obtain a spirulina biomass with a modified content of pigments, lipids, carbohydrates and proteins.

Copper nanoparticles also have a high biological activity against spirulina, which is determined by a number of factors, such as the size of the nanoparticles, the method of their synthesis, and their quantity. The biological activity of copper nanoparticles becomes noticeable already at a concentration of 1 µg/L. Thus, depending on the form and applied concentration, copper can be both a major stressor and an effective biotechnological regulator.

Acknowledgement: The results presented in this work are part of the research project 20.80009.5007.05

Keywords: *Arthrospira platensis*, biotechnology, enzymes, nanoparticles.

IMPROVING SUNFLOWER CROP BIODIVERSITY, BY CREATING NEW, MORE PERFORMANT GENOTYPES

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Biodiversity is the diversity of life on earth, including plants, animals, fungi, and microorganisms. At the genetic level, biodiversity includes the diversity found between varieties and landraces of the same crop, but also extends to diversity present in crop wild relatives.

There are efforts of scientists exploring agrobiodiversity, to develop genetic tools critical for improving crop performance to support both resilience to climate stresses, and local adaptation to low-input agriculture. Oilseed sunflower accounts for up to 12% of the worldwide production of vegetable oils, ranking fourth after palm, soybean and canola oil. Compared to the other main temperate crops, cultivated sunflower is a recent crop. It experienced a domestication bottleneck that narrowed its genetic base but the large number of sunflower CWR makes it possible to mine a vast genetic pool for crop improvement. The use of CWR in the sunflower breeding is a long tradition in the breeding programs. A significant number of inbred lines have been created by interspecific crosses. These lines, together with the wild populations, represent a valuable resource of useful alleles that are abundantly used in the breeding program for increasing genetic variability. There have been introduced resistance genes to economically significant pathogens and parasitic plants, increasing tolerance to herbicides and altering the architecture of the plant.

Current research on CWR is focused on the discovery of new resistance genes to the more virulent populations of *Orobanche cumana*.

By using a very various and valuable germplasm, in different breeding programs there have been obtained valuable inbred lines, having very good characteristics.

It has been obtained an important genetic progress, regarding the productivity, also different agronomic and physiological traits and adaptation to the biotic and abiotic factors.

There have been studied some new sunflower hybrids – contribution of genetic diversity to increasing of productivity.

Keywords: biodiversity, sunflower, interspecific crosses, genetic variability, genetic progress.

AUTHOR INDEX

- Ababii A., 76
Adamciuc A., 26
Agapi I., 147
Anastas A., 148
Andriută C., 120
Andronic L., 49, 64
Antoci L., 64
Anton F. G., 20, 61
Aricu A., 209, 214
Arsene C., 188
Artiomov L., 77
Aslebagh R., 98
Babii C., 98
Babileva A., 121
Bacal S., 84, 149
Bacalov Iu., 122
Baciu A.Ja., 123
Bacu Gh., 79, 103
Bahsiev A., 21
Balacci S., 78, 79, 103
Balan G., 118
Balan I., 78, 79, 91, 101, 103
Balan P., 22, 23
Balici E., 24
Balmuş Z., 25, 29, 36
Banari E., 150
Banița C-D., 202
Barbă A., 203
Barbăroș M., 122
Barbulescu I.D., 202
Batiru Gr., 26
Bădărău S., 80
Begea M., 202
Bejan A., 151
Bejan I.G., 188
Belous S., 152
Belova V., 159
Beșliu A., 110
Bilici I., 23
Bivol A., 80, 178
Bivol C., 117
Bivol E., 80, 178
Bivol I., 40, 128
Bîrcă N., 203, 204
Bîrsa M., 82, 154
Bîrsan A., 122, 174
Blaja S., 209, 214
Blanita D., 124, 126, 142, 145
Blonschi V., 153, 175
Bogaciov E., 42
Bogdan N., 154, 155
Bogdan V., 83
Boian I., 156
Boiangiu R. Ș., 98, 129
Boiciuc C., 124, 145
Bolocan N., 205, 206
Borodaev R., 157
Borozaan E., 54
Botnaraș V., 25
Botnarenco P., 36
Botnari A., 158
Botnaru N., 116
Bounegru S., 26
Brindza J., 41, 213
Brinzaan A., 51
Brînză I., 125
Bulhac I., 86
Bulimaga C., 172, 173, 187
Burcovschi I., 28
Burduja D., 84
Burțeva S., 82
Bușmachiou G., 84, 112, 149
Butnarus V., 25, 29, 36
Buzan V., 78, 79, 103
Buză C., 66, 67
Бречко Е.В., 27
Calalb T., 207, 208
Caldari V., 106, 185
Calestru L., 159
Caraman N., 106, 160
Carrupt V., 209
Cassir P., 30, 31
Cazac V., 161
Cazacova Iu., 79
Cebotari V., 82
Celac M., 227
Cepoi L., 199, 230
Cernăuțeanu V., 154
Certan C., 187
Channaveerappa D., 98
Chihai O., 116, 163
Chiriac T., 199
Chirița E., 122
Chiselîța N., 110
Chiselîța O., 110
Chisnicean L., 76
Chitan R., 85
Ciloci A., 86
Ciobanu R., 32
Ciocarlan A., 209, 214

- Ciolacu T., 180
Ciorchina N., 85, 95, 208
Ciorchina P., 41
Ciomei L., 231
Ćiric A.I., 202
Cirimpei O., 204
Ciursin A., 210
Ćirilg N., 164, 168
Clapco S., 34, 86
Climenco O.A., 35
Cobzac V., 204
Codreanu L., 165
Cojocari D., 26
Cojocarui I., 154
Colibaba L., 166
Coliban Iu., 126
Coman D., 136
Comarova G., 26
Corcimaru, S., 167
Corlateanu L., 168
Coşcodan M., 87, 88, 89
Cotelea L., 25, 29, 36
Cotelea T., 204
Covaci E., 197
Covalciuc D., 76
Covaliov E., 225
Cozari S., 76, 106
Cozmic R., 54, 56
Cretu R., 79, 103
Creţescu I., 202
Cristea N., 37
Criucikov O., 178
Crivoi A., 122
Crucean Ş., 38
Cucicova C., 214
Curiev L., 90, 107
Curşunji D., 39
Darie C., 98
David T., 107
Demcenco B., 91, 101
Demchukova N.V., 172
Deseatnicov O., 225
Dibolscaia N., 185
Dillon T.J., 188
Dobrojan S., 180
Domenco R., 169
Doncila A., 64
Dorif A., 127, 141
Doroftei S.D., 172
Doroftei V., 76, 109
Dreglea M., 62
Druţa A., 122
Du L., 74
Duan R., 74, 211
Duan X., 74
Duca Gh., 197, 205, 206, 229
Duca M., 40, 128
Dudnicenco T., 170, 171
Dumitrache C., 202
Dupree E., 98
Dvornina E., 86
Efremova N., 110
El-Sabeh A., 129
Eldahshan O., 125
Elenciuc D., 117, 118
Elisovetcaia D., 41
Erhan D., 116
Erhan T., 42, 212
Eroshenkova V.A., 172, 173
Fedas V., 123
Frîncu M., 202
Frunză M., 174
Frunze N., 77, 130
Frunzete M. E., 92
Ganea A., 168
Garbuz O., 212, 223, 227
Garbuzneac A., 131
Gavzer S., 37
Ghendov V., 30, 31, 196
Ghendov-Moşanu A., 225
Gherasim E., 93
Ghereg M., 95
Gîrbu V., 228
Gîscă I., 50
Gladcaia A., 46
Gladchi V., 153, 175
Gladei M., 43
Gliga O., 116
Gonceariuc M., 29, 36
Gorgan L., 125
Grabco N., 187
Grajdieru C., 24, 44
Gramovici A., 91, 101
Graur V., 227
Gribincea V., 45
Grosul-Raileanu O., 132
Grozdeva S., 176
Gulea A., 210, 212, 223, 227
Guo X., 211
Gusan A., 46, 58, 68

- Guțu A., 164
Guțu N., 96
Guzun D., 98
Hadjiu S., 126
Hlistun V., 124
Honceriu I., 129
Hritcu L., 125
Indoitu D., 177
Iurcu-Străistaru E., 80, 164, 178
Ivanov I., 67
Ivanova R., 41, 213
Ivănescu L. C., 92, 217
Izverscaia T., 31
Jalba S., 42, 60, 69
Jian M., 204
Jigău Gh., 180
Joita-Pacureanu M., 20, 61
Kallabi F., 129
Kandić V., 53
Kravić N., 53
Kulciti V., 203, 204, 228
Labliuc S., 86
Larion A., 163, 185, 186
Leatamborg S., 168
Leferber D., 124
Leorda A., 75, 111, 154
Listopadova L., 123
Liu Z., 73, 211
Lukasheva N.V., 173
Lungu A., 38, 87, 97, 107, 115
Lungu L., 209, 214
Lupascu G., 37
Mager M., 80
Makovey M.D., 47, 48
Mandela Elorm A., 74
Mangul O., 133
Mapelli C., 188
Marareskul V.A., 182
Marculescu I.S., 202
Marii L., 49
Marteza R., 50
Medvedeva N.L., 173
Meleca A., 178
Melnic G., 163
Merenuic L., 167
Merenuic R., 167
Mereuta I., 79, 123, 134
Mihaila V., 51
Mihasan M., 98, 125, 129
Mîndru A., 215
Mînzat C., 183
Mocanu N., 76, 109
Moldovan C., 52, 104
Morarescu O., 204, 228
Moroz M., 79, 103
Moșin V., 139
Muntean A., 184
Mustea M., 99
Musteata L., 136
Musteata V., 136
Mutu A., 40, 50
Nacu V., 204
Nagacevschi T., 192
Neguța A., 216
Nikolić V., 53
Nistoreanu V., 163, 185, 186
Odobescu V., 42, 69
Olaru E.-I., 217
Olaru R.I., 188
Olaru Ș. M., 217
Oprea D., 61
Osadci N., 91, 101
Osipciuc G., 103
Pacureanu M.J., 231
Paladi V., 100
Perić V., 53
Petcu I., 97, 101
Pintea M., 54, 56
Poleacova L., 134, 137
Popa A., 46, 58, 68
Popescu G., 231
Popescu V., 209, 210
Popușoi A., 220
Popușoi Iu., 122
Port A., 59
Portarescu A., 187
Postovan R., 102
Prisacari S., 87
Racoviță S., 139
Raileanu N., 42, 60, 69
Raischi I., 222
Răileanu V., 195
Revenco A., 122
Revenco N., 126
Risnoveanu L., 20, 61, 231
Roman C., 188
Romanovich N.A., 189
Rosca N., 79, 103
Roșca F., 91, 101
Roșca I., 72

- Roșca T., 143
Rotari E., 26, 62
Rotaru L., 140
Rotaru T., 140
Rudi L., 199
Rudic V., 118
Rusnac A., 223
Rusnac R., 210
Rusu Iu., 63
Rusu Ș., 80, 116, 163, 178
Sacara V., 126, 127, 141, 142, 145
Saltanovici T., 64
Samoilova A., 65, 106
Sarban V., 66, 67
Sasco E., 190
Sava E., 61, 231
Savranschii D., 68
Scerbacova T., 38, 107
Scortesco F., 196
Scraliuc M., 224
Scurtul M., 145
Secu D., 142, 143
Secu Gh., 143
Shepitsky V., 131, 132, 133,
Shleahitici V., 69
Shova E., 191
Shova S., 223
Simić M., 53
Sitnic V., 106
Sitnic V., 106, 144
Șirbu T., 52, 104, 105, 114
Șitnic F., 167
Slanina V., 155
Smerea S., 49
Sochircă V., 192
Soreanu G., 202
Spinu A., 70
Sprincean M., 139
Srdić J., 53
Stache A., 125
Stadnic A., 180, 193
Stingaci A., 71, 107
Stratan L., 195
Stratulat T., 60
Sturza R., 225
Suhodol N., 225
Tabara M., 85, 196
Talmaci N., 226
Talpalaru D., 22, 23
Tanachi T., 72
Tasca C., 197
Teodorescu R.I., 202
Terentii P., 56
Timuș I., 104, 105, 108
Toderaș I., 80
Todiraș V., 87
Todiraș V., 58, 68
Tofan E., 110
Tolstenco D., 111
Tretiacova T., 46, 48
Trofim A., 118
Tsapcov V., 227
Turchin B., 180
Țiței V., 76, 109, 164
Țugulea A., 198
Țugulea C., 112
Țurcan O., 104, 105, 113, 114
Ulchina Ia., 227
Ureche D., 86
Usurelu D.-C., 145
Usurelu N., 124, 126, 142, 145
Vasiliev A., 228
Vicol C., 229
Volosciuc L., 71, 115
Vrinceanu C-R., 202
Yan N., 73
Yang J., 74
Yun X., 74
Yushin N., 199
Zainea P., 138
Zakharov D. S., 200
Zamfirache M. M., 92, 217
Zamornea M., 116, 163
Zamorzaeva I., 21
Zavtoni P., 107
Zhang J., 73, 74, 211
Zhang W., 74, 211
Zhang Z., 211
Zhao J., 73, 74, 211
Žilić S., 53
Zinicovscaia I., 799, 209
Zosim L., 117, 118