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## GENETIC, CHEMICAL, AND PHYSICAL PREDISPOSITIONS OF NEW MAIZE INBRED LINES AND HYBRIDS WITH EFFICIENT PHOTOSYNTHESIS\*\*

**Abstract:** This study confirmed our hypothesis that new maize inbred lines and hybrids derived from them had a dominant property of an efficient photosynthetic model. This property is successfully used in breeding programmes, modern technologies of the seed, and commercial maize production. This statement is supported by the results displayed on the erect position of the top leaves of new maize inbred lines and photosynthetic and florescence parameters: the change of the delayed chlorophyll fluorescence intensity during its course and dynamics, the Arrhenius criterion for the determination of critical temperatures (phase transition temperatures) and the activation energies, as a measure of conformational changes in chloroplasts and the thylakoid membrane. Furthermore, a grain structure including its physical and chemical parameters of new maize inbred lines and hybrids was analysed in the present study. In addition, relevant breeding, seed production and technological traits, properties and parameters of new maize inbred lines and maize hybrids were observed in the present study. The overall presented results show that properties of new inbred lines and maize hybrids are based on the nature of conformational and functional changes that occur in their chloroplasts and thylakoid membranes, as well as, on progressive effects in modern breeding, contemporary hybrid seed production, and the commercial maize production.

**KEY WORDS:** delayed chlorophyll fluorescence, grain, hybrid, inbred, leaf, photosynthetic model, thylakoid membrane, *Zea mays* L.

### INTRODUCTION

The complex and interdependent processes in fundamental, multidisciplinary, and applied sciences are frequently interrelated. This manuscript presents

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\*\* Authors devote this manuscript to the memory of Professor Dr. Milorad Piper, the first and a long-time Director of the Maize Research Institute. This is the 36<sup>th</sup> anniversary of his death. This manuscript presents implementation of his ideas on activities related to maize

the bonds of interrelated studies carried out within breeding, photosynthesis, fluorescence, biophysical chemistry and seed production in new maize inbred lines and hybrids with efficient photosynthetic functions. Maize breeding and seed production have been intensively developing for the last 65 years. As a result of such activity, over 1400 grain and silage hybrids have been derived. Modern equipment and technical and technological prerequisites were provided for carrying out the process of breeding and hybrid and commercial maize seed production (D u v i c k, 1977, 1984; S p r a g u e, 1984; T r i f u n o v i ć, 1986; D u m a n o v i ć, 1986; H a l l a u e r, 1988; I v a n o v i ć et al., 1995; R a d e n o v i ć et al., 2000).

Since 1978, the number of plants per area unit (plant density) has been significantly increasing, which resulted in the significant increase in grain yields of both, maize hybrids and commercial maize (R a d e n o v i ć et al., 1978, 2001 a, b; K o j i ć and I v a n o v i ć, 1986). At the same time, a programme on breeding and the seed production of maize hybrids that included inbreds with erect top leaves has been developed (R a d e n o v i ć et al., 1978, 2003, b, 2004a, b, 2007, 2008; F e l n e r et al., 2006). According to our hypothesis, new maize inbred lines with erect leaves are the closest to the assumptive maize photosynthetic model (R a d e n o v i ć and G r o d z i n s k i j, 1998).

The studies on maize photosynthesis carried out in the previous period did not have a more important application in breeding and the production of maize hybrid seed. It was almost impossible to present practical results and a clear and direct interrelationship among photosynthesis, breeding and the production of maize hybrid seed by an old and traditional approach. The way out was found in the functional connection of interdependence of photosynthetic functions and fluorescence (R a d e n o v i ć et al., 2000, 2001a, b, 2004a, b, 2007, 2010).

During the last 40 years, new and significant studies within the field of bioluminescence and fluorescence phenomena and processes within the plant systems, including maize, have been carried out (B a r b e r and N e u m a n n, 1974; B u k h o v et al., 1989; D z h i b l a d z e et al., 1988; G o v i n d j e e and P a p a g e o r g i o u, 1971; G o v i n d j e e et al., 1990; H a v e m a n and L a v o r e l, 1975; H i p k i n s and B a r b e r, 1974; H o l z a p f e l and H a u g, 1974; J u r i s n i c, 1986; J u r i s n i c and G o v i n d j e e, 1982; K r a u s e and W e i s, 1991; L i c h t e n t h a l e r and R i n d e r l e, 1988; M c c a u l e y and R u b b y, 1981; P a p a g e o r g i o u, 1975; V e s e l o v s k i and V e s e l o v a, 1990; M a r k o v i ć et al., 1987, 1993, 1999; R a d e n o v i ć, 1994; R a d e n o v i ć et al., 1994 a, b; R a d e n o v i ć and J e r e m i ć, 1996). The direct dependence of the delayed chlorophyll fluorescence (DF) intensity on changes of photosynthetic processes in chloroplasts and thylakoid membranes of maize intact leaves was determined (R a d e n o v i ć, 1994 a, b; R a d e n o v i ć and J e r e m i ć, 1996). S Conditions that provided monitoring of complex photosynthetic processes in the intact leaf of maize inbreds by parameters of a photosynthetic and fluorescence model in the form of chlorophyll DF were developed (R a d e n o v i ć et al., 2000, 2001a, b, 2010).

Research methods within the field of biophysical chemistry contributed to diversified binding of studies on photosynthetic and transport processes in

chloroplasts and the thylakoid membrane and different chemical structures of grain with processes of fluorescence spectroscopy and chemical kinetics (R a d e n o v i ć, 1994; R a d e n o v i ć et al., 2007, 2008, 2010; R u b i n et al., 1988).

The objective of the present study was to show that new inbred lines that are included into new high yielding maize hybrids could be an efficient photosynthetic model and could contribute to the functional connection of breeding, photosynthesis and florescence, and thereby to a greater extent to progress of maize breeding and the modern production of hybrid seed and commercial maize.

## MATERIAL AND METHODS

**Plant material** – The studies were performed with the following two new maize inbred lines: ZPPL 218 and ZPPL 318 and the hybrids developed from them: ZP 600, ZP 606, and ZP 666. The observed inbreds and maize hybrids belong to the collection of the Maize Research Institute, Zemun Polje, Belgrade, Serbia.

As these are new inbred lines and maize hybrids, their traits will be presented in this manuscript. Figure 1 shows the actual appearance of new maize inbred lines with erect top leaves: ZPPL 218 and ZPPL 318 and prospective maize hybrids: ZP 600, ZP 606, and ZP 666 with their ears.

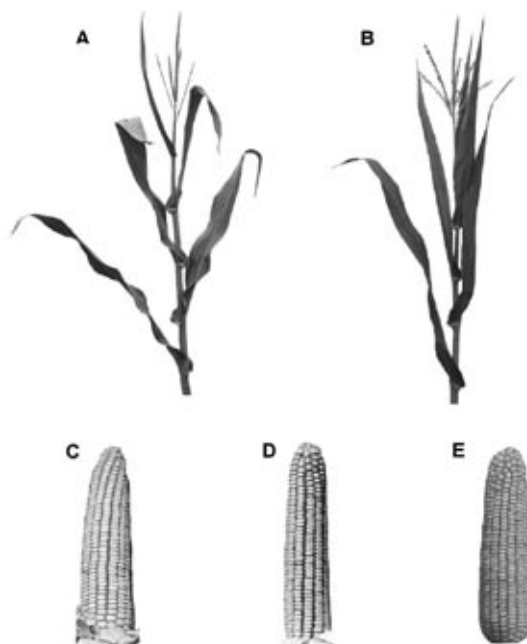


Fig. 1. – Actual appearance of new inbred lines with erect top leaves: ZPPL 218 (A), ZPPL 318 (B), and of prospective hybrids with their ears: ZP 600 (C), ZP 606 (D) and ZP 666 (E)

**Methods** – Overall studies of the stated new inbred lines and prospective maize hybrids developed from them with erect top leaves encompassed several series of experiments in which new and conventional methodological procedures were applied.

1. *The measure of an angle and leaf area* – The first series of experiments was related to studying the erect position of top leaves. A specially designed protractor was used to measure the angle between lines of the position of the above-ear leaf and the position of the plant stalk of new maize inbred lines. The leaf area was measured by using the portable area meter (model LI-3000). Measures of the angle between the above-ear leaf and the stalk and the leaf areas were carried out on 218 plants for each inbred line during the three-year period. These methodical procedures were described in previously published papers (R a d e n o v i ć et al., 2003, 2004a, b, 2007).

2. *Photosynthetic and fluorescence measurements* – The second series of the experiments was related to photosynthetic-fluorescence measurements, including thermal processes of DF, critical phase transition temperatures and activation energies. The test maize inbreds grown in the experimental field of the Maize Research Institute, Zemun Polje, were brought to the laboratory between 7 a.m. and 8 a.m. Plants sampled in the field were transversally cut in the ground internode. In the laboratory, plants were internode lengthwise placed in water. Prior to the fluorescence experiment, all plants were kept under the black ball glass for two hours. Segments of intact above ear leaves were taken from such plants and placed into a chamber of the phosphoroscope. The intact leaf segments were kept in the chamber (in the dark) for at least 15 minutes, and then thermal processes of delayed chlorophyll fluorescence were measured. These tests were performed on 168 plants of each maize inbred line.

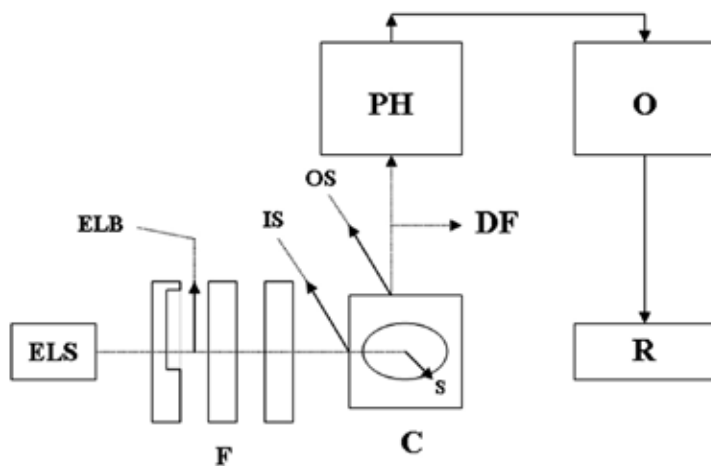


Fig. 2. – Experimental setup of the photosynthetic fluorescence method and the measuring equipment for delayed chlorophyll fluorescence: C – dark chamber with a sample stand; S – sample (intact leaf segment), ELS – excitation light source, PH – photo-multiplier; O – oscilloscope, R – printer, ELB – excitation light beam, DF – luminescent light (delayed fluorescence), IS – input dark chamber slot, OS – output dark chamber slot, F – filters

The improved non-invasive photosynthetic-fluorescence method used to measure DF is schematically presented in Figure 2. This method, developed at the Maize Research Institute, Zemun Polje, has been improved several times. Photosynthetic-fluorescence measurements were performed after a method that had been described in previously published papers (R a d e n o v i ć, 1994; M a r k o v i ć et al., 1996; R a d e n o v i ć et al., 2001a, b, 2002, 2004a, b, 2007, 2008, 2010).

3. *Functional dependence of the yield of prospective maize hybrids for various locations in Serbia* – Functional dependence of the yield of new and prospective maize hybrids ZP 600, ZP 606 and ZP 666 was observed in eight different locations in Serbia with the application of standard methods for a contemporary maize production (R a d e n o v i ć et al., 2010).

4. *Broader presentation of breeding and seed production properties of new inbred lines and maize hybrids with erect top leaves* – As new maize inbred lines, with erect top leaves, and prospective hybrids were observed a broader presentation of relevant breeding, seed production and technological traits, properties and parameters gained by use of standard methods is given.

5. *Chemical composition, physical properties and a structure of grain of prospective maize hybrids with efficient photosynthetic functions* – Methods used for the determination of the chemical composition, physical properties and a structure of grain of prospective maize hybrids with erect top leaves were fully described in previously published papers (R a d o s a v l j e v i ć et al., 2000; R a d e n o v i ć et al., 2010).

## RESULTS

**1. The measure of the angle and the area of the above-ear leaf** – Results on the measures of angles between the above-ear leaf and the stalk and the average leaf areas are presented in Table 1. Based on obtained results on the measures of angles it can be stated that the observed new maize inbred lines belong to the group of 10-15 recently developed inbred lines with erect top leaves.

Tab. 1. – The angle of the above-ear leaf and the leaf area of new maize inbred lines with efficient photosynthesis

Inbred line	FAO maturity group	Heterotic origin of the inbred*	Angle of the above-ear leaf in degrees		Area of the above-ear leaf (x 10 <sup>3</sup> cm <sup>2</sup> )	
			$\bar{x}$	$\sigma$	$\bar{x}$	$\Sigma$
ZPPL 218	650	Zemun Polje – Lancaster	22.1°	1.36	3.91	0.41
ZPPL 318	600	Zemun Polje – BSSS	21.2°	1.15	3.58	0.39

\*Studied new maize inbred lines represent good heterotic pairs, they are characterised as good general combiners for grain yield, they increase well and they are high yielding inbreds

**2. Empirical procedure for photosynthetic fluorescence studies on the above-ear leaf** – The detailed studies on thermal processes of DF of observed new maize inbred lines with erect top leaves were performed. The thermal curve is a curve that shows the dynamics of changes in the stationary DF level intensity in dependence on a temperature. The trend of its establishment is most often analogous to changes in the duration in seconds of segments marked with **a**, **b**, **c**, **d**, **e**, **f**, and **g**, Figure 3, which was determined by the empirical procedure (Radonvić et al., 2008, 2009, 2010). Monitoring the course of the thermal curve and the analysis of the duration of certain segments provided data on the existence of a greater number of critical temperatures (phase transition temperatures) at which greater or smaller structural and functional changes occurred in chloroplasts and the thylakoid membrane of observed new maize inbred lines with erect top leaves.

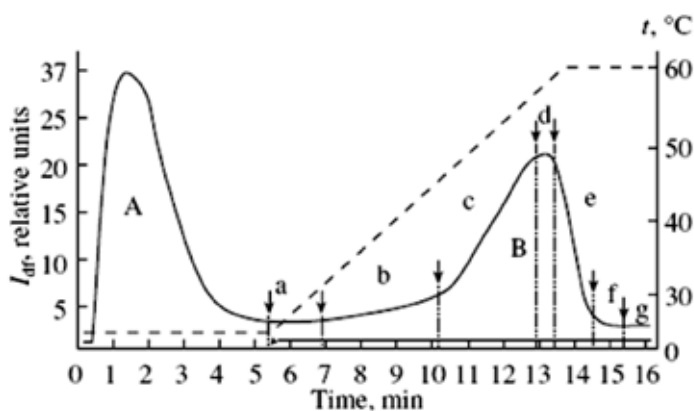


Fig. 3. – Schematic presentation of the empirical procedure for typical changes in DF intensities ( $I_{dr}$ ) on the intact above-ear leaf of the observed new maize inbred lines (solid line) and changes in temperatures (dashed line): Curve **A** indicates induction processes of DF, while curve **B** encompasses photosynthetic fluorescence thermal processes of DF. Typical temporal segments (**a**, **b**, **c**, **d**, **e**, **f** and **g**) on the thermal curve **B** correspond to dynamics of  $I_{dr}$  changes at the time of a DF formation. Conformational and functional changes in chloroplasts and the thylakoid membrane of observed new maize inbred lines with erect top leaves occur at the interception points of typical temporal segments

**3. The exact temperature dependence of the delayed chlorophyll fluorescence intensity for the thylakoid membrane of new maize inbred lines with erect top leaves** – The experimental measures of changes in the stationary DF level in dependence on the temperature, ranging from 25 to 60 °C, were performed. The dynamics of temperature dependence for observed new maize inbred lines with erect top leaves is presented in Figure 4A and B.

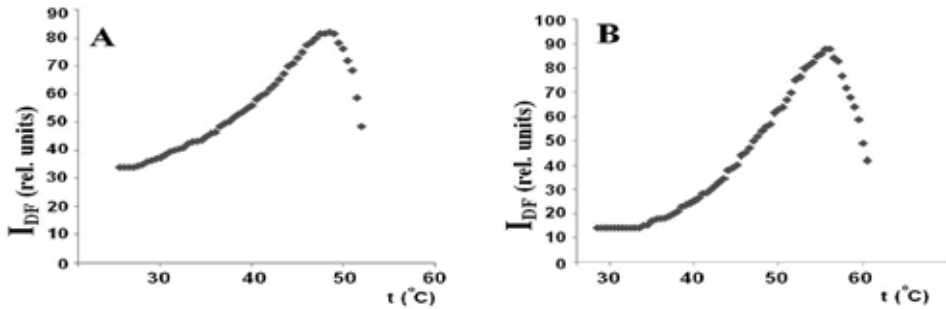


Fig. 4. – A, B. Changes in the intensity of the delayed chlorophyll fluorescence ( $I_{DF}$ ) of thermal processes in dependence on the effects of temperatures in chloroplasts and the thylakoid membrane of the intact above-ear leaf of new maize inbred lines with erect top leaves: ZPPL 218 (A), and ZPPL 318 (B)

**4. The Arrhenius plot for the determination of critical temperatures and conformational changes in chloroplasts and the thylakoid membrane of the new maize inbred lines with erect top leaves** – The Arrhenius plot is based on the linearization of the exact DF temperature dependence of observed new maize inbred lines. Critical temperatures (phase transition temperatures) at which conformational changes occur in chloroplasts and the thylakoid membrane are determined by the application of the Arrhenius plot. Results of the Arrhenius plot application to new maize inbred lines with erect top leaves are presented in Figure 5A, B.

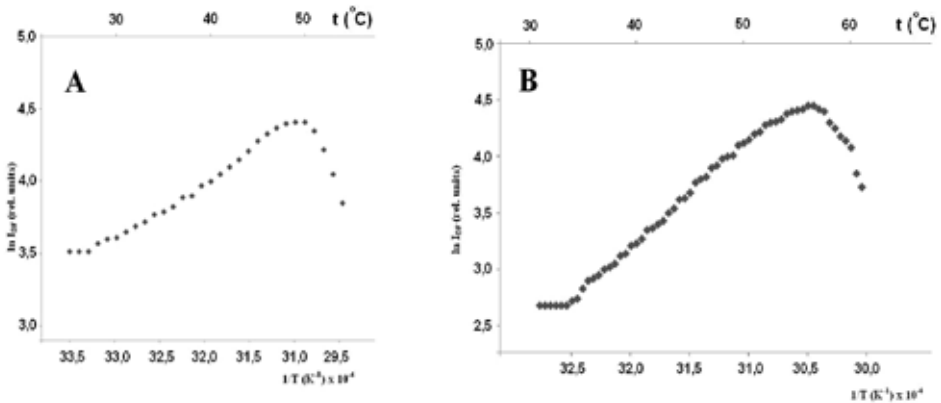


Fig. 5. – A, B. The Arrhenius plot for the determination of critical temperatures ( $T$ , °K) and conformational changes in chloroplasts and the thylakoid membrane of the above-ear leaf of observed new maize inbred lines with erect top leaves: ZPPL 218 (A) and ZPPL 318 (B)

**5. Activation energy and critical temperatures in the thylakoid membrane of the observed new maize inbred lines with erect top leaves** – Detailed studies on the thermal processes of DF, and especially on the analysis

of experimental thermal curve, encompassed not only the temperature dependence and the Arrhenius plot, but also the estimation of values of activation energies (Ea) for critical temperatures (phase transition temperatures) in chloroplasts and the thylakoid membranes of the observed new maize inbreds with erect top leaves: ZPPL 218 and ZPPL 318. Obtained results are shown in Table 2.

Tab. 2. – Changes in activation energies (Ea) and critical temperatures (t°C) in the course of thermal processes in the thylakoid membrane of the intact above-ear leaf of studied new maize inbred lines with erect top leaves

ZPPL 218		ZPPL 318	
Ea, kJ/mol	t, °C	Ea, kJ/mol	t, °C
–	27.0	–	33.5
43.1	29.0	40	38
27.3	36.9	77.23	53.5
37.0	43.5	26.09	56.5
42.5	47.8	50.51	59.3
51.1	49.9	227.52	–

**6. Functional dependence of the yield of new maize hybrids for different locations in Serbia** – New and prospective maize hybrids: ZP 600, ZP 606 and ZP 666, are mainly indented for the cultivation in Banat, Srem, Bačka, Mačva, and alongside riverbanks in Serbia. The preliminary results on yields of the stated maize hybrids are presented in Table 3.

Tab. 3. – Yields of new and prospective maize hybrids (t ha<sup>-1</sup>) with efficient photosynthesis in eight locations of Serbia\*

Hybrid	Locations in Serbia**								Average $\bar{x}$
	1 $\bar{x}$	2 $\bar{x}$	3 $\bar{x}$	4 $\bar{x}$	5 $\bar{x}$	6 $\bar{x}$	7 $\bar{x}$	8 $\bar{x}$	
ZP 600	12.8	11.0	11.2	12.9	11.8	10.6	10.8	13.5	11.8
ZP 606	12.9	11.6	10.5	12.5	10.9	10.7	10.4	12.2	11.6
ZP 666	12.7	10.9	9.5	11.6	10.6	10.0	10.3	12.5	11.0

\*Results were obtained during 2009, 2010 and 2011

\*\*Locations in Serbia by the ordinal number: 1 – Loznica, western Serbia; 2 – Sakule, southern Banat; 3 – Smederevo, the Danube region; 4 – Zmajev, southern Bačka; 5 – Žarkovac, eastern Srem; 6 – Batoš, mid Banat; 7 – Divoš, northern Srem; 8 – Bečej, eastern Bačka

**7. Brief survey of breeding and seed production traits of new maize inbred lines and maize hybrids with efficient photosynthetic functions** – Observed new maize inbred lines ZPPL 218 and ZPPL 318 have been included in breeding for the last 2-3 years. Due to it, relevant observations of their total traits, performances, and parameters are presented in Table 4.



Tab. 4. – Relevant breeding and seed production traits of new maize inbred lines with efficient photosynthesis

Ord. no.	Name and defining of traits	Brief description of breeding, seed production and technological traits of new maize inbred lines	
		ZPPL 218	ZPPL 318
1	Heterotic origin	Zemun Polje – Lancaster	Zemun Polje – BSSS
2	FAO maturity group	650	600
	Grain yield ha <sup>-1</sup> in kg at 14% moisture		
3	a) dry land farming	3220±204	4056±265
	b) irrigation	4186±255	6045±330
	Number of plants ha <sup>-1</sup> at harvest		
4	a) dry land farming	65000	71500
	b) irrigation	71500	79400
	Inbred tolerance to stress factors such as drought, high temperature and the like	Inbred has a good tolerance to drought and high temperatures	Inbred has a good tolerance to drought and high temperatures
5			
6	Grain properties	Kernel of this inbred belongs to dent type and has plenty of anthocyanins on its flanks	Yellow-orange kernel of this inbred belongs to semi-dent type
7	% moisture in grain at harvest	Inbred is harvested at 18.00%	Inbred is harvested at 22.00%
			Inbred has high quality grain and hybrids developed from this inbred have also high quality grain suitable for nutrition of ruminants and nonruminants.
8	Is grain suitable for nutrition of ruminants and nonruminants	Inbred has high quality grain and hybrids developed from this inbred have also high quality grain suitable for nutrition of ruminants and nonruminants.	Inbred has high quality grain and hybrids developed from this inbred have also high quality grain suitable for nutrition of ruminants and nonruminants.
9	Is the inbred suitable for the development of silage hybrids?	Inbred is very suitable for the development of silage hybrids	Inbred is very suitable for the development of silage hybrids
10	Grain digestibility (%)	81.6	80.44

In the same way, new and prospective maize hybrids ZP 600, ZP 606, and ZP 666 have already caught attention of experts; hence, it is necessary to study their overall traits. Results on studied traits of observed new and prospective hybrids are presented in Table 5.

**8. Chemical composition and physical properties of new inbred lines and prospective maize hybrids with efficient photosynthetic functions** – Results on studies of chemical composition and physical properties of new inbred lines (ZPPL 218 and ZPPL 318) and prospective maize hybrids with erect top leaves (ZP 600, ZP 606 and ZP 666) are presented in Tables 6 and 7.

Tab. 5. – Relevant breeding and seed production traits of prospective maize hybrids with efficient photosynthesis

Ord. no.	Name and defining of traits	Brief description of breeding, seed production and technological traits of prospective maize hybrids		
		ZP 600	ZP 606	ZP 666
1	Grain yield ha <sup>-1</sup> in kg at 14% moisture	11930	11830	11750
2	Optimum sowing density and affinity of hybrids to densities (ha)	58-62000	58-62000	60-65000
3	Regional distribution of hybrids according to agroecological characteristics of the region	Very adaptable, tolerant to drought and various growing conditions of the region	Very tolerant to drought and high temperatures. Hybrid has high yields under conditions of Banat.	Very adaptable, resistant to precipitation distribution and high temperatures
4	FAO maturity group	FAO 580-600	FAO 640-660	FAO 580-600
5	Description of the essential hybrid stalk traits	Medium tall, slender, elastic and very firm	Medium tall, slender, elastic and very firm	Medium short, slender, elastic and very firm
6	General data on the type and quality of grain	Yellow kernel belongs to a dent type, is of high quality. 1000-kernel mass is 488.7 g	Orange kernel belongs to a dent type. 1000-kernel weight is 474.6 g. Protein content up to 12%.	Orange kernel is deeply set. 1000-kernel weight is 356.2 g. Oil content up to 6%.
7	Dates of sowing and data on emergence and early growth	Tolerates early sowing, has good emergence and early growth	Tolerates early sowing. Emerges excellently under conditions of positive temperatures. Has a good early growth.	Tolerates early sowing. It prefers a slightly deeper sowing. Has a good emergence and early growth.
8	Tolerance, resistance and adaptability	Very adaptable to soil and conditions of cropping practices. Tolerant to drought and high temperatures.	It prefers more fertile soils and intensive cropping practices. Extremely tolerant to drought.	Very tolerant and adaptable to growing conditions. Tolerant to drought.
9	Stay green and suitability for silage	Grain filling period is long and dry down is good. Expresses stay green trait. It is excellent for silage production.	It has grain of high quality with protein content up to 12%. It is suitable for ruminants and nonruminants.	It has extremely pronounced stay green trait. Stalk is short and slender hence silage mass yield is low.
10	Grain suitability for the nutrition of domestic animals	Proportion of horny flourey endosperm is greater. Grain is healthy and of high quality. It suitable for ruminants and nonruminants.	It has grain of high quality with protein content up to 12%. It is suitable for ruminants and nonruminants.	Grain protein, i.e. oil content amount to 10-11%, i.e. 6%, respectively. It is suitable for ruminants and nonruminants.
11	Grain digestibility (%)	92.47	92.08	96.65

Tab. 6. – Chemical composition of new maize inbred lines and hybrids with efficient photosynthesis

Inbred	Starch (%)	Proteins (%)	Oil (%)	Crude fibre (%)
ZPPL 218	69.10	9.60	5.79	2.18
ZPPL 318	71.27	10.31	4.91	2.39
Hybrid				
ZP 600	73.01	7.76	5.76	1.91
ZP 606	73.48	9.84	5.06	2.23
ZP 666	74.32	9.61	6.10	2.42

Tab. 7. – Physical properties of grain of new maize inbred lines and hybrids with efficient photosynthesis

Inbreds	TKW*	TW	D	FI	MR	HEF	SEF
ZPPL 218	341.5	844.1	1.29	23.28	10.5	58.1	41.9
ZPPL 318	316	811.7	1.28	24.31	12.2	62.3	37.7
Hybrids							
ZP 600	488.0	788.2	1.27	34.3	12.7	55.8	44.2
ZP 606	474.0	777.0	1.26	48.2	11.3	57.9	59.1
ZP 666	356.0	806.2	1.28	24.5	10.9	42.7	40.0

\* TKW = 1000-kernel weight (g), TW = test weight (kg m<sup>-3</sup>), D – density (g cm<sup>-3</sup>), FI – floatation index (%), MR – milling response (s), HEF – hard endosperm fraction, (%), SEF – soft endosperm fraction (%)

## DISCUSSION

The second half of the 20<sup>th</sup> and the first decade of the 21<sup>st</sup> century are characterised by a great success achieved in maize breeding and the production of fundamental maize seed, hybrid maize seed of high quality and of commercial maize. The number of plants per area unit has been increasing since 1978. This programme was referred to as a “plant density” programme and it further directly affected the yield increase of high quality fundamental and hybrid maize seed (Radenović et al., 1978, 2001 a, b). In addition, a programme on the development of maize inbred lines with erect top leaves was established at the same time as the “plant density” programme. It was considered that inbreds with the erect top leaves were the closest to the proposed efficient photosynthetic model (Radenović et al., 1978; Radenović and Grodzinski, 1998; Radenović et al. 2000, 2001a, c, 2003, 2004a). The complementary and mass implementation of these programmes led to very important results in both, maize breeding and the hybrid seed production (Ivanović et al., 1995; Trifunović, 1986; Trifunović et al., 2000; Dumanović, 1986; Kojić and Ivanović, 1986). New and numerous hybrids for grain and silage were developed and grown on large areas due to their high yielding potential and the appropriate quality of the plant and the grain (Duvick, 1984; Russell, 1986; Dumanović, 1986; Hallauer, 1988; Kojić and Ivanović, 1986; Ivanović et al., 1995).

The special contemporary breeding studies have been performed on top leaves of maize inbred lines. In recent times, the ear leaves have been particularly observed, but also other top leaves up to the tassel. The most efficient and the longest photosynthetic processes necessary for the maize plant have been achieved by these leaves (R a d e n o v i ć and G r o d z i n s k i, 1998). According to the stated, a hypothesis that top leaves (above-ear leaves) achieving the efficient photosynthesis has been proposed.

This study was an attempt to answer the following questions by using different interdependent studies and analyses: (1) were there reliable and dominant traits of maize inbred lines with erect top leaves by which planned and satisfactory progress in maize breeding and the high-quality hybrid seed maize production could be achieved? and (2) which traits should such maize inbred lines have?

The gained results of experimental studies can offer at least a partial answer to asked questions. The first series of experiments included the measure of the angle and the leaf area of observed new maize inbred lines with erect top leaves. The results obtained on these traits (Table 1) classify them into important breeding and seed production traits (R a d e n o v i ć et al., 2003, 2004a, b, 2007, 2008, 2010). The second series of experiments encompassed photosynthetic fluorescence studies on conformational and functional changes in chloroplasts and the thylakoid membrane of the intact above-ear leaf of new maize inbred lines. The temperature dependence of thermal processes of DF for the studied maize inbred lines is presented in a form of the empirical procedure (Figure 3). However, the exact results of the temperature dependence of DF for all new maize inbred lines with erect top leaves are presented in Figure 4A, B. The presented results show that the temperature dependence of DF in each of new maize inbred lines with erect top leaves is characterised by typical intersection points of two segments on the thermal curve (Figures 3 and 4A, B). The first typical point occurred on the intersection of the segment **a** and the segment **b** and it represented the lowest critical temperature at which the initial change in the DF intensity was observed. The second typical point occurred on the intersection of the segment **b** and the segment **c** and it was related to a linear monotony with the angle of the increasing part of the DF intensity curve. Evident changes in the structure of the thylakoid membrane occurred in this region. The third typical point reflected a smaller or a greater rotundity of DF intensity peaks. The breaking “conformational” changes occurred in two intersection points of the segments **c** and **d** and the segments **d** and **e**. The fourth typical point was related to the linear monotony and the inclination angle of the declining part of the DF intensity curve. This segment of the thermal curve bore the last conformational changes that had occurred in chloroplasts and the thylakoid membrane. These changes can hardly be described as characters of functioning of a living leaf. The typical intersection points designated as **f** and **g** almost had no physiological role. The analysed typical intersection points, Figures 3 and 4A, B, can be considered the points characterising new maize inbred lines with erect top leaves, as these points are precisely the points of conformational and functional changes in the thylakoid membrane (R a d e n o v i ć et al., 2003, b, 2004a, b, 2007, 2008, 2010).

All critical temperatures (phase transition temperatures) at which even the slightest conformational changes had occurred in chloroplasts and the thylakoid membranes of new maize inbred lines with erect top leaves were determined by the Arrhenius criterion and the linearisation of the DF temperature dependence. The values of critical temperatures in °C, their frequency and intermediate distance characterise observed new maize inbred lines with erect top leaves in relation to their tolerance, resistance, flexibility and adaptability not only to increased and high temperatures, but also to drought (R a d e n o v i ć et al., 2001a, b, c, 2002, 2003). The Arrhenius criterion is based on the existence of straight lines. Each Arrhenius straight line represents its activation energy ( $E_a$ ). The intersection point of two straight lines is designated by a critical temperature. Results of the  $E_a$  values in the inclining and declining part of the thermal curve are explained by lesser or greater conformational changes that occur in the molecules of pigments (chlorophyll) in the thylakoid membrane with the temperature increase. Due to such changes, these molecules become more reactive and thereby gain the additional energy that is used in the recombining process of the DF occurrence (Table 2) (R a d e n o v i ć, 1994; R a d e n o v i ć et al., 2001c, 2004a, b).

Presented photosynthetic fluorescence traits of studied new maize inbred lines with erect top leaves can contribute to more exact, rational and expeditious proceedings of breeding processes and the production of high-quality hybrid maize seed and commercial maize, which makes these maize inbred lines exceptionally important.

Achieved results on yields of new and prospective maize hybrids (Table 3) should be considered as preliminary ones. According to the description of breeding, seed production and technological traits, properties and parameters (Table 5), it is obvious that these are stable hybrids with high quality grains. However, it is necessary to find appropriate locations for such hybrids (Banat, Bačka, Srem, Mačva, river valleys...) in which their full genetic potential of the yield can be used.

Gained results (Tables 6 and 7) present physical traits and the chemical composition that especially indicate grain quality of new maize inbred lines and prospective hybrids with efficient photosynthetic functions (R a d o s a v l j e v i ć et al., 2000; R a d e n o v i ć et al., 2010).

A brief survey of breeding, seed production and technological traits of new inbred lines and prospective maize hybrids with efficient photosynthetic functions (Tables 4 and 5) completes above presented results and contributes to the improvement of modern programmes of both, breeding and current hybrid seed and commercial maize productions.

## CONCLUSION

According to obtained results, it can be concluded that the non-invasive photosynthetic fluorescence method can be used in breeding and the production of maize hybrid seed and thereby the estimation of new maize inbred

lines for tolerance, resistance, flexibility and adaptability to increased and high temperatures, as well as, to drought can be performed. The application of the stated non-invasive method resulted in the determination of numerous traits, properties and parameters of the photosynthetic apparatus of new inbred lines and maize hybrids with efficient photosynthetic functions, such as:

- Temperature dependence observed within the range of 24°C to 60°C,
- The critical temperatures at which greater or smaller structural and functional changes occur in chloroplasts the thylakoid membrane were determined,
- Values of activation energy ( $E_a$ , kJ/mol ) alongside straight lines before and after occurrence of critical temperatures in the thermal chlorophyll DF process were determined,
- Different monotonies of the intensity in the inclining part of the thermal curve was present; these monotonies point out to unequal tolerance, resistance, flexibility, stability and adaptability of new maize inbred lines to increased and high temperatures, as well as, to drought,
- It was shown that observed, new inbred lines maize hybrids have a dominant property of efficient photosynthetic functions,
- A numerous relevant breeding, seed production and technological traits of new maize inbred lines and hybrids with efficient photosynthetic functions were presented,
- A functional dependence of yields of prospective maize hybrids for eight locations in Serbia was established,
- It was established that commercial maize of studied new and prospective hybrids is of high quality that provides its diverse utilisation.

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## ГЕНЕТИЧКЕ И ХЕМИЈСКО-ФИЗИЧКЕ ПРЕДИСПОЗИЦИЈЕ НОВИХ ЛИНИЈА И ХИБРИДА КУКУРУЗА СА ЕФИКАСНИМ ФОТОСИНТЕЗОМ

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### Резиме

Проучаване су две нове инбред линије кукуруза: ZPPL 218 i ZPPL 318 и са њима створени перспективни хибриди ZP 600, ZP 606 и ZP 666 за које је доказано да поседују доминантно својство ефикасног фотосинтетичног модела што се успешно користи у оплемењавању, савременим технологијама за производњу хибридног семена и меркантилног кукуруза. Овој констатацији иду у прилог изложени резултати о исправном положају вршних листова нових инбред линија кукуруза и о фотосинтетично-флуоресцентним показатељима: промени интензитета закаснеле флуоресценције хлорофила у њеном току и динамици, Аренијусовом критеријуму за одређивање критичних температура (температуре фазних прелаза) и о енергији активације као мери структурних промена у хлоропластима и тилакоидној мембрани. У раду се анализира структура зрна укључујући и његове физичке и хемијске показатеље нових инбред линија и хибрида кукуруза. Исто тако, у раду се разматрају релевантна селекционарска, семенарска и технолошка својства, карактеристике и параметри нових инбред линија и хибрида кукуруза. Укупно изложени резултати показују да су својства нових инбред линија и хибрида кукуруза заснована на природи структурних и функционалних промена, које се одигравају у хлоропластима и тилакоидној мембрани као и на прогресивним ефектима у модерном оплемењавању, савременој производњи хибридног семена и меркантилног кукуруза.

КЉУЧНЕ РЕЧИ: закаснела флуоресценција хлорофила, *Zea mays* L., зрно, линија, лист, тилакоидна мембрана, фотосинтетични модел, хибрид

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