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Traits of New Maize Inbred Lines and Hybrids with Efficient Photosynthetic Functions

- Original scientific paper -

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Abstract: This study confirms the hypothesis that there are new elite maize inbred lines and prospective hybrids, which have a dominant property of an efficient photosynthetic model that is successfully used in breeding programmes, modern technologies of the seed and commercial maize production. This statement is supported by the displayed results 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 the thylakoid membrane. Furthermore, a grain structure, including its physical and chemical parameters, of new maize inbred lines was analysed in the present study. Also, relevant breeding, seed production and technological traits, properties and parameters of new inbred lines and prospective maize hybrids were observed in the present study. The presented results show that properties of new inbred lines and prospective 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 maize breeding, modern hybrid seed production and the commercial maize production.

Key words:. Delayed chlorophyll fluorescence, grain, inbred, hybrid, leaf, photosynthetic model, thylakoid membrane, *Zea mays* L.

Introduction

The connection of complex and interrelated processes in fundamental, multidisciplinary and applied sciences is necessary in current studies. This manuscript presents the connection 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 and because of such activity more than 1350 grain and silage hybrids were derived. Modern equipment and technical and technological prerequisites were provided for carrying out the process of breeding and hybrid maize seed production, ***Duvick***, 1977, 1984, ***Sprague***, 1984, ***Trifunović***, 1986, ***Dumanović***, 1986, ***Hallauer***, 1988, ***Ivanović et al***., 1995, ***Radenović et al.,*** 2000.

Since 1978, the number of plants per area unit (plant density) has been significantly increasing, which mostly affected the increase in grain yields of both, maize hybrids and commercial maize, ***Radenović et al***., 1978, ***Kojić*** and ***Ivanović***, 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 performed, ***Radenović et al***., 1978, 2003a, b, 2004a, b, 2007, 2008, ***Felner et al***., 2006. According to our hypothesis, maize inbred lines with erect leaves are the closest to the assumptive maize photosynthetic model, ***Radenović*** and ***Grodzinskij***, 1998.

The studies on maize photosynthesis carried out in the previous period did not have a more important application in breeding and the maize hybrid seed production. 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 photosynthetic functions with fluorescence, ***Radenović 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, ***Barber*** and ***Neumann***, 1974, ***Bukhov et al***., 1989, ***Dzhibladze et al***., 1988, ***Govindjee*** and ***Papageorgiou***, 1971, ***Govindjee et al***., 1990, ***Haveman*** and ***Lavorel***, 1975, ***Hipkins*** **and** ***Barber***, 1974, ***Holzapfel*** and ***Haug***, 1974, ***Jurisnic***, 1986, ***Jurisnic*** and ***Govindjee***, 1982, ***Krause*** and ***Weis***, 1991, ***Lichtenthaler*** and ***Rinderle***, 1988, ***McCauley*** and ***Rubby,*** 1981, ***Papageorgiou***, 1975, ***Veselovski*** and ***Veselova***, 1990, ***Marković et al***., 1987, 1993, 1999, ***Radenović***, 1994, ***Radenović et al.,*** 1994a, b**,** ***Radenović*** and ***Jeremić***, 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, ***Radenović***, 1994, ***Radenović*** and ***Jeremić***, 1996. Conditions that provided monitoring of complex photosynthetic processes in the intact leaf of maize inbreds by a photosynthetic and fluorescence model in the form of chlorophyll DF were developed, ***Radenović et al***., 2000, 2001a, b, 2010.

Research methods within the filed of biophysical chemistry contributed to diversified connections 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, ***Radenović***, 1994, ***Radenović et al***., 2007, 2008, 2010, ***Rubin 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 the total 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 prospective maize hybrids belong to the collection of the Maize Research Institute, Zemun Polje, Belgrade.

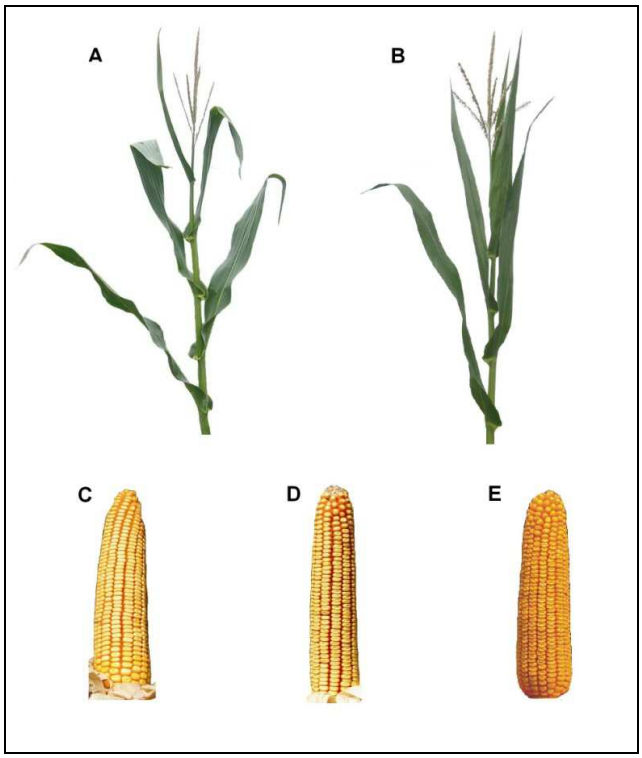

Figure 1. Actual appearance of new maize inbred lines with erect top leaves: ZPPL 218 (A), ZPPL 318 (B), and ears of their prospective hybrids ZP 600 (C), ZP 606 (D) i ZP 666 (E)
Stvarni izgled novih elitnih inbred linija, sa uspravnim položajem vršnih listova: ZPPL 218 (A), ZPPL 318 (B) i perspektivnih hibrida, sa njihovim klipovima: ZP 600 (C), ZP 606 (D) i ZP 666 (E)

As these are new elite inbred lines and prospective maize hybrids their traits will be separately 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.

Methods. - Overall studies of the stated new inbred lines and hybrids developed from them with erect top leaves encompassed several series of experiments in which standard and other appropriate 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 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, ***Radenović et al***., 2003a, 2004a, b, 2007.

2. Photosynthetic 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. A segment of intact above ear leaves was 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 DF were measured. These tests were performed on 168 plants of each inbred line.

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, ***Radenović***, 1994, ***Marković et al***., 1996, ***Radenović et al***., 2001a, b, 2002, 2004a, b, 2007, 2008, 2010.

3. *Functional dependence of the yield of new and prospective maize hybrids for various locations in Serbia.* - Functional dependence of the yield of prospective 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, ***Radenović et al.***, 2010.

4. *Broader presentation of breeding and seed production properties of new inbred lines and prospective maize hybrids with erect top leaves.* - As new elite inbred lines with erect top leaves and new maize 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.

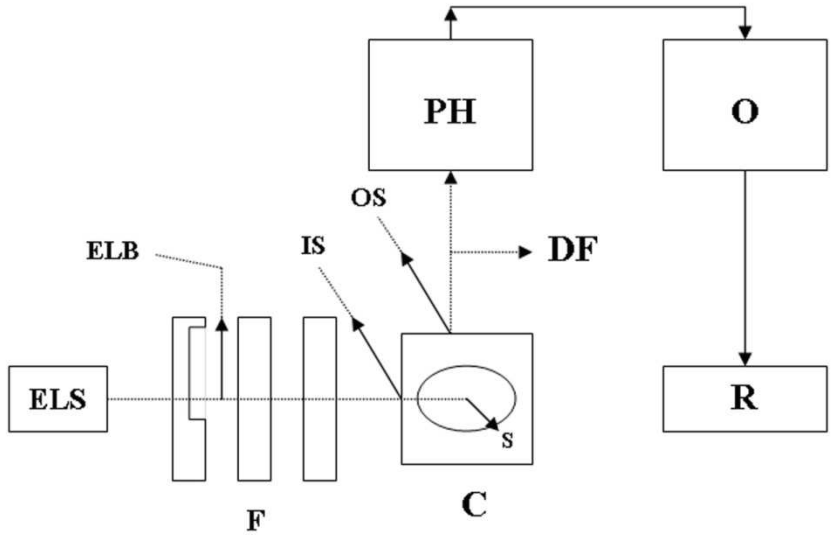

Figure 2. Experimental setup of the photosynthetic fluorescence method and the measuring equipment for delayed chlorophyll fluorescence: C - dark chamber with a sample stand; S - sample (intact leaf segment), ELS - excitation light source, PH - photo-multiplier; O - oscilloscope, R - printer, ELB - excitation light beam, DF - luminescent light (delayed fluorescence), IS - input dark chamber slot, OS - output dark chamber slot, F - filters
Principijelna shema fotosintetično-fluorescentnog metoda i aparature za merenje zakasnele fluorescencije hlorofila: C - mračna komora sa postoljem za uzorke; S - uzorak (segment intaktnog lista) ELS - izvor ekscitirajuće svetlosti, PH - fotomultiplikator; O - osciloskop, R - pisač, ELB - ekscitirajući zrak, DF - lumuniscentna svetlost (zakasnela fluorescencija), IS - ulazni prorez u mračnu komoru, OS - izlazni prorez iz komore, F-filtri

5. *Grain chemical composition, physical properties and structure of new elite maize inbred lines with efficient photosynthetic functions.* - Methods used for the determination of the chemical composition, physical properties and the grain structure of new maize inbred lines with erect top leaves were fully described in previously published papers, ***Radosavljević et al***., 2000, ***Radenović et al.***, 2010.

Results and Discussion

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, as well as, 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 elite maize inbred lines belong to the group of 10-15 recently developed inbred lines with erect top leaves.

Table 1. The Angle of the Above-ear Leaf and the Leaf Area of New Maize Inbred Lines with Efficient Photosynthetic Functions
Veličina ugla prvog lista iznad klipa i lisne površine novih inbred linija kukuruza sa efikasnim fotosintetičnim funkcijama

Inbred line Inbred linija	FAO maturity group FAO grupa zrenja	Heterotic origin of the inbred* Heterotično poreklo linije*	Angle of the above-ear leaf in degrees Ugao prvog lista iznad klipa u stepenima		Area of the above-ear leaf ($\times 10^3$ cm^2) Lisna površina prvog lista iznad klipa	
			$\bar{x}$	σ	$\bar{x}$	Σ
ZPPL 218	650	Zemun Polje - Lankaster	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.
*Proučavane nove inbred linije kukuruza predstavljaju dobre heterotične parove, odlikuju se dobrim kombinacionim sposobnostima za prinos zrna, dobro se umnožavaju i rodne su.

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 usually 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, ***Radenović 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 the thylakoid membrane of observed new maize inbred lines with erect top leaves.

3. The 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 a 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.

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 linearisation of the DF temperature dependence of observed 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.

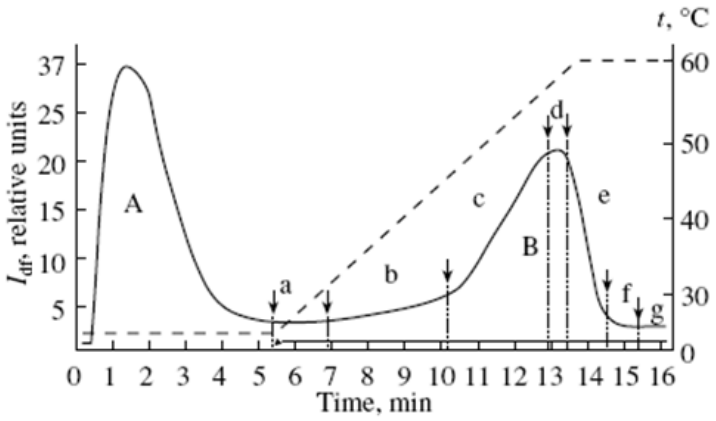

*Figure 3. Schematic presentation of the empirical procedure for typical changes in DF intensities (I_{df}) on the intact above-ear leaf of the observed new maize inbred lines (solid line) and changes in temperatures (dashed line): Curve **A** indicates induction processes of DF, while curve **B** encompasses 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_{df} 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*

*Shematski prikaz empirijskog postupka tipičnih promena intenziteta zakasnele fluorescencije hlorofila (I_{ZF}) na intaktnom prvom listu iznad klipa proučavanih novih inbred linija kukuruza (puna linija) i promene temperature (isprekidana linija): Kriva **A** označava indukcione procese ZF hlorofila, a kriva **B** obuhvata fotosintetično-fluorescentne termalne procese ZF hlorofila. Tipični vremenski segmenti (**a**, **b**, **c**, **d**, **e**, **f** i **g**) na termalnoj krivoj **B** odgovaraju dinamici promene I_{ZF} u vremenu uspostavljanja ZF hlorofila. Na mestu dodira tipičnih vremenskih segmenata dolazi do strukturnih i funkcionalnih promena u hloroplastima i tilakoidnoj membrani proučavanih novih inbred linija kukuruza sa uspravnim položajem vršnih listova.*

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 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.

6. Functional dependence of the yield of new and prospective maize hybrids for different locations in Serbia. - New and prospective maize hybrids: ZP600, ZP 606 and ZP 666, are mainly indented for the cultivation in Banat, Srem, Bačka, Mačva and alongside river banks in Serbia. The preliminary results on yields of the stated maize hybrids are presented in Table 3.

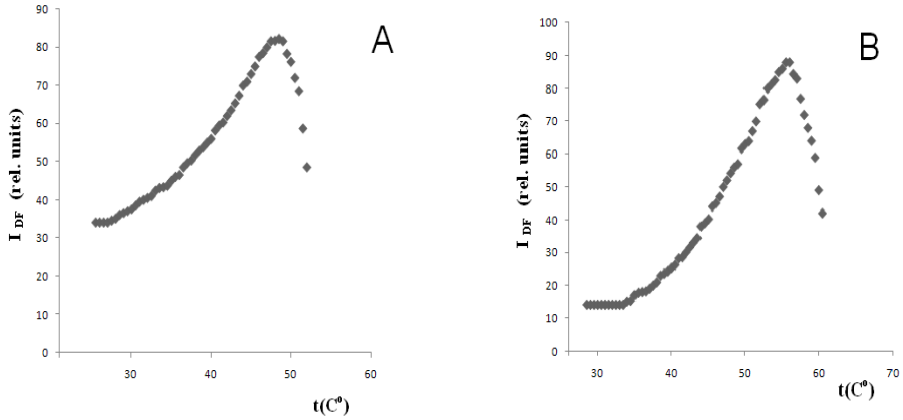

Figure 4 A, B *Changes in the intensity of the delayed chlorophyll fluorescence (I_{DF}) of thermal processes in dependence on the effects of temperatures in the thylakoid membrane of the intact above-ear leaf of new maize inbred lines with erect top leaves: ZPPL 218 (A), and ZPPL 318 (B)*
Dinamika promene intenziteta zakasnele fluorescencije hlorofila (I_{ZF}) termalnih procesa u zavisnosti od delovanja temperature u hloroplastima i tilakoidnoj membrani intaktnog prvog lista iznad klipa novih inbred linija kukuruza sa uspravnim položajem vršnih listova: ZPPL 218 (A), ZPPL 318 (B)

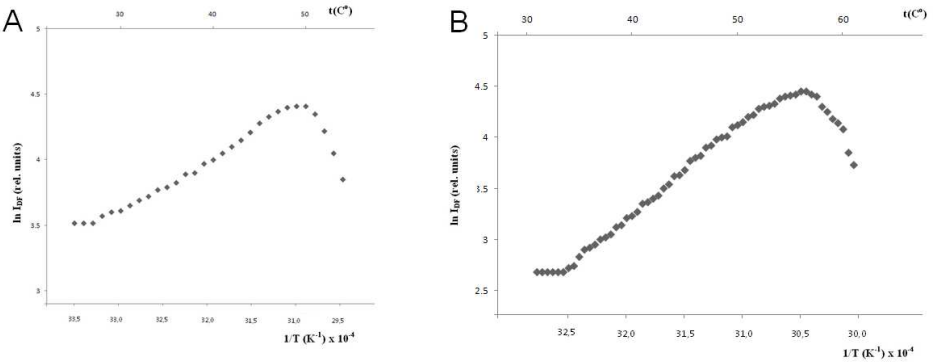

Figure 5 A, B *The Arrhenius plot for the determination of critical temperatures(T, °K) and conformational changes in chlroplasts and the thylakoid membrane of the above-ear leaf of observed new maize inbred lines with erect top leaves: ZPPL 218 (A) and ZPPL 318 (B)*
Arenijusov kriterijum za određivanje kritičnih temperatura (T, °K) i strukturnih promena u hloroplastima i tilakoidnoj membrani intaktnog prvog lista iznad klipa proučavanih novih inbred linija kukuruza sa uspravnim položajem vršnih listova: ZPPL 218 (A) i ZPPL 318 (B).

Table 2. Changes in Activation Energies (Ea) and Critical Temperatures (tºC) in the Course of Thermal Processes in the Thylakoid Membrane of the Intact Above-ear Leaf of Studied New Maize Iinbred Lines with Erect Top Leaves
Promena energija aktivacije (Ea) i kritičnih temperatura (tºC) za vreme termalnih procesa u tilakoidnoj membrani intaktnog prvog lista iznad klipa proučavanih novih inbred linija kukuruza sa uspravnim položajem vršnih listova

ZPPL 218		ZPPL 318	
Ea, kJ/mol	t, ºC	Ea, kJ/mol	t, ºC
-	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	-

Table 3. Yields of New and Prospective Maize Hybrids (t ha^{-1}) with Efficient Photosynthetic Functions in Eight Locations of Serbia
Prinosi novih i perspektivnih hibrida kukuruza (t/ha) sa efikasnim fotosintetičnim funkcijama u osam lokacija Srbije

Hybrid Hibrid	Location in Serbia - Lokacije u Srbiji*								
	1 $\bar{x}$	2 $\bar{x}$	3 $\bar{x}$	4 $\bar{x}$	5 $\bar{x}$	6 $\bar{x}$	7 $\bar{x}$	8 $\bar{x}$	Prosek $\bar{x}$
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

*Location in Serbia by the ordinal number: **1 -** Loznica, western Serbia; **2 -** Sakule, southern Banat; **3 -** Smederevo, the Danube region; **4 -** Zmajevo, southern Bačka; **5 -** Žarkovac, eastern Srem; **6 -** Batoš, mid Banat; **7 -** Divoš, northern Srem; **8** - Bečej, eastern Bačka
*Lokacije u Srbiji prema rednom broju: **1 -** Loznica, zapadna Srbija; **2 -** Sakule, južni Banat; **3 -** Smederevo, Podunavlje; **4 -** Zmajevo, južna Bačka; **5 -** Žarkovac, istočni Srem; **6 -** Batoš, srednji Banat; **7 -** Divoš, severni Srem; **8** - Bečej, istočna Bačka
**Results gained in 2009 and 2010 - Rezultati dobijeni za 2009 i 2010 godinu

7. Broad survey of breeding and seed production traits of new maize inbred lines and prospective maize hybrids with efficient photosynthetic functions. - Observed new inbred lines ZPPL 218 and ZPPL 318 have been included in breeding for the last 3-5 years. Due to it, Table 4 presents relevant observations of their total traits, performances and parameters.

As new and prospective maize hybrids: ZP 600, ZP 606 and ZP 666 have already caught attention of experts it is necessary to study their total traits. Results on studied traits of observed prospective hybrids are presented in Table 5.

Table 4. Relevant Breeding and Seed Production Traits of New Maize Inbred Lines with Efficient Photosynthetic Functions
Relevantna selekcionarska i semenarska svojstva novih inbred linija kukuruza sa efikasnim fotosintetičnim funkcijama

Ord. no. Red. br.	Name and defining of traits Naziv i definisanje svojstava	Brief description of breeding, seed production and technological traits of new maize inbred lines Kraći opis selekcionarskih, semenarskih i tehnoloških svojstava novih inbred linija kukuruza	
		ZPPL 218	ZPPL 318
1	Heterotic origin Heterotično poreklo	Zemun Polje - Lancaster	Zemun Polje - BSSS
2	FAO maturity group FAO grupa zrenja	650	600
3	Grain yield ha^{-1} in kg at 14% moisture Prinos zrna sa 14 % vode, kg/ha a) dry land farming a) u suvom ratarenju b) irrigation b) u navodnjavanju	 3220±204 4186±255	 4056±265 6045±330
4	Number of plants ha^{-1} at harvest a) dry land farming a) u suvom ratarenju b) irrigation b) u navodnjavanju	 65000 71500	 71500 79400
5	Stalk properties Karakteristike stabljike	Inbred has a modern stalk that is medium high, slender and elastic Linija ima modernu stabljiku koja je srednje visoka, tanka i elastična	Inbred has a modern stalk that is lower, slender and elastic Linija ima modernu stabljiku koja je niža, tanka, čvrsta i elastična
6	Stalk resistance to lodging Otpornost stabljike na poleganje	Inbred is resistant to lodging, which makes it suitable for growing in high densities Linija je otporna na poleganje i pogodna za gajenje u velikim gustinama	Inbred is very resistant to lodging, which makes it suitable for growing in high densities Linija je veoma otporna na poleganje i pogodna za gajenje u velikim gustinama
7	Erect position of ear leaf Uspravni položaj lista na klipu	Inbred has an erect top leaves Linija ima uspravan položaj vršnih listova	Inbred has very erect top leaves Linija ima veoma uspravan položaj vršnih listova

8	Stay green Da li je list do berbe ostao zelen?	Stay green trait is medium pronounced. Leaves above ear remain green until waxy ripeness Linija je sa srednje izraženim svojstvom *stay greena*. Listovi iznad klipa ostaju zeleni sve do voštane zrelosti zrna.	Stay green trait is very pronounced. Leaves above ear remain green until waxy ripeness Linija ima veoma izraženo svojstvo *say greena*. Listovi iznad klipa ostaju zeleni sve do voštane zrelosti
9	Inbred tolerance to stress factors such as drought, high temperature and the like Kakva je tolerantnost linije prema stresnim faktorima: suša, visoke temperature i sl.?	Inbred has good tolerance to drought and increased and high temperatures Linija dobro podnosu sušu, više i visoke temperature	Inbred has very good tolerance to drought and increased and high temperatures Linija veoma dobro podnosu sušu, više i visoke temperature
10	Grain properties Karakteristike zrna	Kernel of this inbred belongs to dent type and has plenty of anthocyanins on its flanks Linija ima zrno tipa zubana sa dosta antocijana po bokovima zrna	Orange-yellow kernel of this inbred belongs to semi-dent type Linija ima zrno tipa polutvrdunca narandžasto žute boje
11	% moisture in grain at harvest % vode u zrnu u berbi	Inbred is harvested at 18.00% Berba linije se vrši sa 18,00%	Inbred is harvested at 22.00% Berba linije se vrši sa 22,00%
12	Grain water status in the maturation period Stanje vode u zrnu u periodu sazrevanja	Daily dry down at maturation is 0.74% Dnevno otpuštanje vode u periodu sazrevanja zrna, je 0,74%	Daily dry down at maturation is 0. 56% Dnevno otpuštanje vode u periodu sazrevanja zrna je 0,56%
13	Is harvest of the inbred easy? Da li je kod linije berba laka?	Harvest of the inbred is easy Linija se lako bere	Harvest of the inbred is easy Linija se lako bere
14	What does inbred emergence look like? Kakvo je nicanje linije?	Inbred emerges very well Linija ima vrlo dobro nicanje	Inbred emerges very well Linija ima vrlo dobro nicanje
15	What does initial growth of the inbred look like? Kakav je rani porast kod linije?	Inbred initial growth is good Linija ima dobar rani porast	Inbred initial growth is good Linija ima dobar rani porast

16	Is grain suitable for nutrition of ruminants and nonruminants? Da li je zrno linije pogodno za ishranu preživara i nepreživara?	Inbred has high quality grain and hybrids developed from this inbred have also high quality grain suitable for nutrition of ruminants and nonruminants. Linija ima veoma kvalitetno zrno i daje hibride sa kvalitetnim zrnom pogodnim za ishranu preživara i nepreživara.	Inbred has high quality grain and hybrids developed from this inbred have also high quality grain suitable for nutrition of ruminants and nonruminants. Linija ima veoma kvalitetno zrno i daje hibride sa kvalitetnim zrnom pogodnim za ishranu preživara i nepreživara.
17	What is a content of carotene in inbred grain? Kakav je sadržaj karotena u zrnu linije?	Grain of this inbred has a common content of carotene and hybrids developed from this inbred are very suitable for nutrition of pigs and ruminants Zrno linije ima uobičajeni sadržaj karotena i njeni hibridi su vrlo pogodni za ishranu svinja i preživara	Grain of this inbred has a higher content of carotene and hybrids developed from this inbred are very suitable for nutrition of laying hens Zrno linije ima veći sadržaj karotena i njeni hibridi su vrlo pogodni za ishranu koka nosilja
18	Is the inbred suitable for the development of silage hybrids? Da li je linija pogodna za kreiranje hibrida kukuruza za silažu?	Inbred is very suitable for the development of silage hybrids Linija je veoma pogodna za kreiranje hibrida kukuruza za silažu	Inbred is very suitable for the development of silage hybrids Linija je veoma pogodna za kreiranje hibrida kukuruza za silažu
19	What is a shelling percentage of the inbred at milling? Kakav randman ima linija pri meljavi?	Inbred has a floury endosperm and due to it milling is good and easy Linija ima brašnjavi endosperm i dobro i lako se melje	Inbred has a harder endosperm but milling is good Linija ima tvrđi, endosperm, ali se dobro melje
20	Digestibility (%) Svarljivost zrna (%)	81.6	80.44

Table 5. Relevant Breeding and Seed Production Traits of New Maize Hybrids with Efficient Photosynthetic Functions
Relevantne karakteristike perspektivnih hibrida kukuruza sa efikasnim fotosintetičnim funkcijama

Ord. no. Red. br.	Name and defining of traits Naziv i defisanje svojstava	Brief description of breeding, seed production and technological traits of prospective maize hybrids Kraći opis selekcionarskih, semenarskih i tehnoloških svojstava perspektivnih hibrida kukuruza		
		ZP 600	ZP 606	ZP 666
1	Average grain yield ha^{-1} in kg at 14% moisture Prosečan prinos zrna sa 14 % vode (t/ha)	11.93	11.83	11.75
2	Optimum sowing density and affinity of hybrids to densities (plants ha^{-1}) Optimalna gustina i sklonost hibrida prema gustini (bilj/ha)	58-62000	58-62000	60-65000
3	Regional distribution of hybrids according to agroecological characteristics of the region Rejonizacija hibrida prema agroekološkim karakteristikama rejona	Very adaptable, tolerant to drought and various growing conditions of the region Vrlo adaptibilan, dobro podnosi sušu i različite uslove gajenja rejona.	Very tolerant to drought and high temperatures. Hybrid has high yields under conditions of Banat. Veoma tolerantan na sušu i visoke temperature. Dobre prinose daje u uslovima Banata.	Very flexible and resistant to precipitation distribution and temperatures Veoma plastičan i otporan prema rasporedu padavina i temperaturi
4	FAO maturity group FAO grupa zrenja	FAO 580-600	FAO 640-660	FAO 580-600
5	Description of the essential hybrid stalk traits Opis bitnih karakteristika stabljike hibrida	Medium tall, slender, elastic and very firm Srednje visoka, tanka, elastična i veoma čvrsta	Medium tall, slender, elastic and very firm Srednje visoka, tanka, elastična i veoma čvrsta	Medium short, slender, firm and very elastic Srednje niska, tanka, čvrsta i veoma elastična

6	General data on the type and quality of grain Opšti podaci o tipu i kvalitetu zrna	Yellow kernel belongs to a dent type, is of high quality. 1000-kernel weight is 488.7 g Zrno žute boje u tipu zubana, odličnog kvaliteta. Njegova apsolutna masa je do 488,7 g	Orange kernel belongs to a dent type. 1000-kernel weight is 474.6 g. Protein content up to 12%. Zrno narandžaste boje u tipu zubana. Njegova apsolutna masa je do 474,6 g. Sadržaj proteina do 12%	Orange kernel is deeply set. 1000-kernel weight is 356.2 g. Oil content up to 6%. Zrno narandžaste boje, jako duboko postavljeno. Njegova apsolutna masa je do 356,2 g. Sadržaj ulja do 6%.
7	Dates of sowing and data on emergence and early growth Rokovi setve i podaci o nicanju i ranom porastu	Tolerates early sowing, has good emergence and early growth Dobro podnosi ranu setvu, ima dobro nicanje i dobar rani porast	Tolerates early sowing. Emerges excellently under conditions of low positive temperatures. Has a good early growth. Dobro podnosi ranu setvu. Odlično niče u uslovima nižih pozitivnih temperatura. Ima veoma dobar rani porast	Tolerates early sowing. It prefers a slightly deeper sowing. Has a good emergence and early growth. Dobro podnosi ranu setvu. Pogoduje mu malo dublja setva. Ima odlično nicanje i izuzetan rani porast.
8	Tolerance, resistance and flexibility Tolerantnost, otpornost i plastičnost	Very flexible to soil and conditions of cropping practices. Tolerant to drought and high temperatures. Vrlo plastičan prema zemljištu i uslovima agrotehnike. Tolerantan na sušu i visoke temperature.	It prefers more fertile soils and intensive cropping practices. Extremely tolerant to drought. Pogoduju mu plodnija zemljišta i intezivnija agrotehnika. Izuzetno tolerantan na sušu.	Very tolerant and flexible to growing conditions. Tolerant to drought. Veoma tolerantan i plastičan na uslove gajenja. Dobro podnosi sušu.
9	Stay green and suitability for silage Da li je list do berbe ostao zelen (*atay green*)? Pogodnost za silažu	Grain filling period is long and dry down is good. Expresses stay green trait. It is excellent for silage production. Ima dug period nalivanja zrna i dobro otpušta vlagu. Ispoljava stay green svojstvo. Odličan za proizvodnju silaže	It has grain of high quality with protein content up to 12%. It is suitable for ruminants and nonruminants. Ima veoma kvalitetno zrno sa sadržajem proteina do 12%. Pogodan je i za preživare i za nepreživare.	It has extremely pronounced stay green. Stalk is low and slender hence silage mass yield is low. Ima izraženo *stay green* svojstvo. Stabljika je niska i tanka te daje mali prinos silažne mase.

10	Suitability for the nutrition of domestic animals Pogodnost zrna za ishranu domaćih životinja	Proportion of horny floury endosperm is greater. Grain is healthy and of high quality. It is suitable for ruminants and nonruminants. Ima veći udeo rožastog endosperma. Zrno mu je veoma zdravo i kvalitetno. Pogodan je za preživare i za nepreživare.	It has grain of high quality with protein content up to 12%. It is suitable for ruminants and nonruminants. Ima veoma kvalitetno zrno sa sadržajem proteina do 12%. Pogodan je za preživare i za nepreživare.	Grain protein, i.e. oil content amount to 10-11%, i.e. 6%, respectively. It is suitable for ruminants and nonruminants. Sadržaj proteina u zrnu je 10-11%, a sadržaj ulja preko 6%. Pogodan je za preživare i za nepreživare.
11	Grain digestibility (%) Svarljivost zrna (%)	92.47	92.08	96.65

7. Chemical composition, physical properties and grain structure of new maize inbred lines with efficient photosynthetic functions. - Results on studies on grain structure, physical properties and chemical composition of new maize inbred lines ZPPL 218 and ZPPL 318 with erect top leaves are presented in Tables 6, 7 and 8.

Table 6. Grain Structure of New Maize Inbred Lines with Efficient Photosynthetic Functions
Struktura zrna novih inbred linija kukuruza sa efikasnim fotosintetičnim fukcijama

Inbred Linija	Pericarp (%) Perikarp (%)	Germ (%) Klica (%)	Endosperm (%) Endosperm (%)
ZPPL 218	5.76	11.53	82.71
ZPPL 318	7.47	11.40	81.14

Table 7. Physical Properties of Grain of New Maize Inbred Lines with Efficient Photosynthetic Functions
Fizička svojstva zrna novih inbred linija kukuruza sa efikasnim fotosintetičkim funkcijama

Inbred Linija	TKW* AM*	TW HM	D G	FI IF	MR OM	HEF TF	SEF MF
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

* TKW = 1000-kernel weight (g), TW = test weight (kg m^{-3}), D - density (g cm^{-3}), FI - floatation index (%), MR - milling response (s), HEF - hard endosperm fraction, (%), SEF - soft endosperm fraction (%)

*AM-apsolutna masa (g), HM-hektolitarska masa (kgm^{-3}), G-gustina (gcm^{-3}), IF-indeks flotacije (%), OM-otpornost na mlevenje (%), TF-tvrda frakcija endosperma (%), meka frakcija endosperma (%)

Table 8. Grain Chemical Composition of New Maize Inbred Lines with Erect Top Leaves
Hemijski sastav novih inbred linija kukuruza sa uspravnim položajem vršnih listova

Inbred Linija	Moisture (%) Vlaga (%)	Starch (%) Skrob (%)	Proteins (%) Proteini (%)	Oil (%) Ulje (%)	Crude fibre (%) Celuloza (%)
ZPPL 218	9.4	69.1	9.6	5.79	2.18
ZPPL 318	9.3	71.27	10.31	4.91	2.39

The second half of the 20th and the first decade of the 21st century are characterised by a great success achieved in maize breeding and the production of fundamental and hybrid maize seed of high quality and commercial maize. The number of plants per area unit has been growing 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. 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, 2003a, 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 with high grain and silage yields 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 maize leaves. 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, ***Radenović*** and ***Grodzinski***, 1998. According to the stated, a hypothesis, that top leaves (above-ear leaves) in particular achieved the efficient photosynthesis, has been proposed.

This study was an attempt to answer the following questions by using different tests 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, ***Radenović et al***., 2003a, 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 with 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 inbred lines with erect top leaves, as these points are precisely the points of conformational and functional changes in the thylakoid membrane, ***Radenović et al***., 2003a, b, 2004a, b, 2007, 2008, 2010.

All critical temperatures (phase transition temperatures) at which even the slightest conformational changes had occurred in 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 ^{o}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, ***Radenović et al***., 2001a, b, 2002, 2003a. The Arrhenius criterion is based on the existence of straight lines. Each Arrhenius straight line represents its activation energy (Ea). The intersection point of two straight lines is designated by a critical temperature. Results of the Ea 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), ***Radenović***, 1994, ***Radenović et al***., 2003a, b, 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, 7 and 8) point out to structural grain parameters (pericarp, germ and endosperm), physical traits and the chemical composition that indicate exceptional quality of grain of new maize inbred lines with efficient photosynthetic functions, ***Radosavljević et al***., 2000, ***Radenović et al***., 2010.

A broad survey of breeding, seed production and technological traits of new maize inbred lines with efficient photosynthetic functions (Table 4) completes above presented observations 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 maize hybrid seed production 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 prospective maize hybrids with efficient photosynthetic functions, such as:

Temperature dependence observed within the range of 24-60^{o}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 (Ea, kJ/mol) alongside straight lines 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 were 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 and prospective maize hybrids have a dominant property of efficient photosynthetic functions;

A numerous relevant breeding, seed production and technological traits of new maize inbred lines with efficient photosynthetic functions were presented;

A functional dependence of yields of prospective maize hybrids for eight locations in Serbia was established;

Relevant traits of prospective maize hybrids with efficient photosynthetic functions were presented;

It was established that commercial maize of studied prospective hybrids is of high quality that provides its diverse utilisation.

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Svojstva novih inbred linija i hibrida kukuruza sa efikasnim fotosintetičkim funkcijama

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R e z i m e

U ovom radu vršena su proučavanja novih elitnih inbred linija kukuruza: ZPPL 218 i ZPPL 318 i perspektivnih hibrida kukuruza ZP 600, ZP 606 i ZP 666 stvorenih iz njih. Pokazano je da se potvrđuje naša hipoteza da nove elitne inbred linije i perspektivni hibridi kukuruza poseduju dominantno svojstvo efikasnog fotosintetičnog modela što se uspešno koristi u oplemenjivanju, savremenim tehnologijama proizvodnje hibridnog semena i merkantilnog kukuruza. Ovoj konstataciji idu u prilog izloženi rezultati o uspravnom položaju vršnih listova novih inbred linija kukuruza i o fotosintetički-fluorescentnim pokazateljim: promeni inteziteta zakasnele fluorescencije hlorofila u njenom toku i dinamici, Arenijusovom kriterijumu za određivanje kritičnih temperature (temperature faznih prelaza) i o energiji aktivacije kao meri strukturnih promena u hloroplastima i tilakoidnoj membrani. U radu se analiziraju struktura zrna uključujući i njegove fizičke i hemijske pokazatelje novih inbred linija kukuruza. Isto tako, u radu se razmatraju relevantna selekcionarska, semenarska i tehnološka svojstva, karakteristike i parametri novih inbred linija i perspektivnih hibrida kukuruza. Ukupno izloženi rezultati pokazuju da su svojstva novih inbred linija i perspektivnih hibrida kukuruza, zasnovana na prirodi strukturnih i funkcionalnih promena koje se odigravaju u hloroplastima i tilakoidnim membranama, kao i na progresivnim efektima u oplemenjivanju, savremenoj proizvodnji hibridnog semena i merkatilnog kukuruza.

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