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INDUCTION CHLOROPHYLL FLUORESCENCE INDICATORS IN LENTIL DEPENDING ON SEED PRE-TREATMENT AND EXTRA-ROOT NUTRIENTS

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Abstract. The article contains the results of many years of research on the formation of basic and calculated indicators of chlorophyll fluorescence induction depending on the options for inoculation, treatment of seeds with microelements and use of various options for foliar fertilization for growing lentils. There was applied the generally accepted methodological protocol to determine and calculate the main and derivative parameters of chlorophyll flowering induction during dark adaptation of the assimilation apparatus of plants. The indicator reaction of changes in the basic parameters – initial flowering (F_0), flowering of the plateau zone (F_{pl}), maximum flowering (F_m) and stationary flowering (F_{st}) to the optimization of the lentil nutrition system both at the stage of pre-sowing seed preparation and at the stage of carrying out single and binary foliar fertilizing with microfertilizers. The significance of the reaction was noted due to the corresponding increase in F_0 , F_{pl} , F_m , F_{st} indicators by 52.6%, 69.1%, 91.4% and 36.2%, respectively, for the combination of seed inoculation, pre-sowing treatment with trace elements and the use of two-time foliar fertilization in comparison with the control variant. The use of derived calculation indices based on the basic indicators of the induction of chlorophyll flowering proved the possibility and expediency of using such an indicator method in the assessment of the abiotic-adaptive state of plants and the optimality of applied agrotechnological measures in the cultivation of lentils.

Key words: Lens culinaris; Chlorophyll fluorescence; Seed inoculation; Pre-sowing treatment; Microfertilizers.

INTRODUCTION

The photosynthetic activity of the plant is basic to ensure the realization of its productive potential. One of the innovative methods that allows to effectively assess the response of this system to additional agrotechnological measures is the induction of chlorophyll fluorescence (Kalaji, H.M. et al. 2017).

Fluorescence of chlorophyll is an indicator that allows to study in living objects the flow of photochemical reactions associated with the work of photosystem II (PSII), which is the most sensitive to environmental factors. The results of studies of chlorophyll fluorescence intensity (CFI) contribute to a deeper understanding of the regulatory mechanisms that ensure efficient energy conversion in the primary and subsequent stages of photosynthesis (Martinazzo, E.G. et al. 2012; Kargar, M. et al. 2019).

The graph of changes in fluorescence (Fig. 1) from the moment of illumination until reaching a stationary level carries information about the state of the photosynthetic apparatus of a plant leaf. Changes O–I–D–P are called the first wave, or rapid induction of fluorescence. It occurs in 1–3 seconds, depending on the intensity of light and other factors, and is observed both in living objects and in isolated chloroplasts. The slower P–S–M–T changes are known as the second wave, or slow induction of fluorescence. These changes take place over a period of several tens of seconds to several minutes, depending on the object and the conditions of the experiment (Tseng, Y.-C., Chu, S.-W. 2017).

The time dependence of the fluorescence intensity of chlorophyll has the characteristic shape of a curve with one or several maxima and was named the curve of induction of chlorophyll fluorescence (Kautsky curve). The shape of this curve is quite sensitive to the changes that occur in the photosynthetic apparatus of plants when adapting to different environmental conditions, which became the basis for the widespread use of the Kautsky effect in the study of photosynthesis (Chen, Kh. et al. 2019).

At the initial moment of time, all photosynthetic electron transfer channels are open and the maximum energy of excited electrons goes to the photosynthetic process. During this period, chlorophyll fluorescence is minimal, and its intensity on the Kautsky curve is denoted by F_0 . The transition from

 F_0 to Fpl is caused by the transfer of electrons from the reaction centers of FS II through feotin to the primary acceptors (chions). The transition from F0 to Fpl is observed, for example, during a short period of dark adaptation (Kalaji, H.M. et al. 2018).

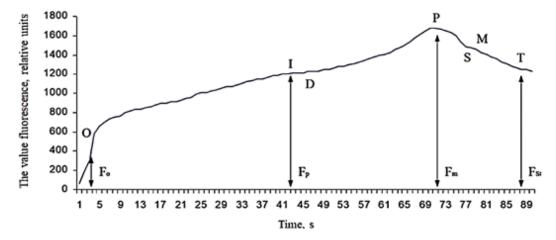


Figure 1. Typical chlorophyll fluorescence induction curve Kautsky: F_0 is the initial value of fluorescence induction; F_p (or F_{pl}) is the "plateau" fluorescence induction value; F_m is the maximum value of fluorescence induction; F_{st} is the stationary value of fluorescence induction after light adaptation of a plant leaf (Brestic, M., Zivcak, M. 2013).

The entire segment from F_0 to F_m is called the fast phase of fluorescence or variable fluorescence. The slow phase of chlorophyll fluorescence induction represents all induction transitions after reaching the Fm peak (Gorbunov, M.Y., Falkowski, P.G. 2021).

It is known that certain segments of the chlorophyll fluorescence induction curve are indicators of the corresponding physiological processes in the photosynthesis chain. Therefore, violations of individual links of photosynthesis, which are caused by exo- and endogenous factors, are manifested in characteristic changes of the corresponding segments of the curve of induction of chlorophyll fluorescence (Korres, N.E. et al. 2003).

Features of the induction of chlorophyll fluorescence depend on the state of the entire photosynthesis system and reflect the kinetics of all links of the biochemical chain of photosynthesis (Govindjee, G. 2004). Changes in any photosynthetic chain lead to a change in the shape of the chlorophyll fluorescence induction curve. Therefore, based on the appearance of this curve, it is possible to diagnose the current state of the plant's photosynthetic apparatus, evaluate changes in the efficiency of photosynthesis with changes in the light regime, temperature, humidity and, in particular, the use of fertilizers and growth-stimulating drugs.

At the same time, it is indicated that the influence on this process of options for the use of macroand microelements, biological growth-stimulating drugs is a debatable issue, as it is determined by the nature and chemistry of the specified substances, their physiological activity and terms of use (Ruban, A.V. 2016; Gorbunov, M.Y., Falkowski, P.G. 2021).

The results of our research, taking into account the aspect of the importance of the chlorophyll fluorescence indicator in evaluating the effectiveness of additional measures to regulate plant nutrition (inoculation, feeding, seed treatment with microfertilizers), will contribute to the deepening of knowledge about the physiological component of such methods.

MATERIALS AND METHODS

The Floratest portable fluorometer was used to study the processes of influence of the application of lentil nutrition optimization measures. The portable fluorometer 'Floratest' consists of a basic unit with a graphic liquid crystal display, control buttons, a remote optoelectronic sensor, a cable for connecting to a USB port of a personal computer and a network adapter (Romanov, V.A. et al. 2010).

The total measurement duration is 4 minutes, the functional measurement period is 3 minutes. Leaf plates for measurement were taken at the bean formation phase (BBCH 65–68) from identical tiers in

each series of experimental variants in the amount of 25 for each repetition. Measurements were made in the middle part of the leaf. The measurement was carried out after adaptation of the leaf to the dark for 10 minutes in 4-fold repetition for each option at 90 points with a time interval from 3 μ s to 300 s with subsequent calculation of relative units for 3 minutes with registration of changes in chlorophyll fluorescence.

In the course of the experiments, generally accepted indicators of the curve were analyzed (Brestic, M., Zivcak, M. 2013; Kalaji, H.M. et al. 2017) in relative units of the fluorescence standard: F0 – minimal fluorescence, Fpl – value of fluorescence induction 'plateau', Fm – maximal fluorescence, Fst – fluorescence in steady state.

The calculated indicators used in the studies based on the basic CFI curve (F0, Fpl, Fm, Fst) are presented in Table 1.

Table 1. Derived index indicators of the CFI curve used in research (according to the protocol for the analysis of the chlorophyll fluorescence curve (Brestic, M., Zivcak, M. 2013; Kalaji, H.M. et al. 2017)

The CFI curve sndex	The applied formula			
Fluorescence rise	$dF_{pl} = F_{pl} - F_0$			
Maximum variable fluorescence	$F_{v} = F_{m} - F_{0}$			
Index of the effect of exogenous and endogenous factors	$rac{dF_{pl}}{F_{_{_{_{_{_{_{_{_{}}}}}}}}}}$			
Photochemical efficiency or quantum efficiency (EP)	$EP = \frac{F_{v}}{F_{m}}$			
Photochemical quenching (Q _{ue})	$Q_{ue} = \frac{F_0}{F_v}$			
Leaf water potential (L_{wp})	$L_{wp} = \frac{F_{\scriptscriptstyle m}}{F_{\scriptscriptstyle 0}}$			
Plant viability index (RF _d)	$RF_{d} = \frac{F_{m} - F_{st}}{F_{st}}$ $K_{ef} = \frac{F_{st}}{F_{m}}$			
Indicator of endogenous (stress) factors (Kef)	$K_{ef} = \frac{F_{st}}{F_m}$			
Value of photochemical quenching of fluorescence (QP)	$QP = \frac{F_m - F_{st}}{F_m - F_0}$			
Index of the efficiency of the primary reactions of photosynthesis (K_{prp})	$K_{prp} = rac{F_{_{ar{ u}}}}{F_{0}}$			
Fluorescence decay coefficient (K _{fd})	$K_{_{\mathit{fd}}} = \frac{F_{\mathit{m}}}{F_{\mathit{st}}}$			
Relative change of fluorescence at time t (V _t)	$V_t = \frac{F_{st} - F_0}{F_m - F_0}$			

The main and derivative indicators of the induction of chlorophyll fluorescence were calculated in the system of the field multivariate experiment (Table 2). Sowing was carried out in the second decade of April by the usual row method. The rate of sowing was 2.1–2.2 million similar seeds per 1 ha at a depth of 4–5 cm. After sowing, rolling was used. The zoned variety of Linza lentils (*Lens culinaris* M. a subspecies of *edible green lentils*) was used in the research.

The hydrothermal parameters of lentil vegetation period varied, having formed certain typological features of the research years (Table 2).

Considering the biological features of lentils, the most extreme conditions for realizing the potential of lentils were in the conditions of 2021, especially in the period of June–July. The weather conditions of the 2019 lentil growing season were marked as the year with the maximum moisture supply against the background of average evaporation for the conditions of the research zone. As a result, the ratio of

excessive moisture in certain periods of vegetation with sharp fluctuations in average daily temperatures had a heterogeneous effect on the growth processes and qualitative transformations of lentil plants.

Year of research		Mo	Average for the period of		
	V	VI	VII	VIII	vegetation
2019	4,710	1,555	1,003	0,235	1,690
2020	5,489	1,474	0,649	0,474	1,859
2021	4,204	2,662	0,530	1,077	1,543

Table 2. Monthly average hydrothermal coefficient* over the growing season of lentil, 2019–2021

Thus, according to the presented data, the weather conditions during the years of research can be characterized as relatively favorable for the growth and germination of lentils both in terms of moisture content and in the nature of the sum of effective temperatures, and their sharp contrast in the years of research allows us to use the abiotic component in the evaluation system received data.

The experiment was repeated four times. Placement of options is systematic in two tiers. The area of the registered experimental plot is 25 m², the total area is 40 m². Factorial formula 2:2:4=16 options (the total number of sections for systems of 4 repetitions is 64) (Table 3).

Statistical evaluation of all analyzes was performed using the statistical software package Statistica 10.0 for Windows according to the analysis of variance (Hinnkelmann, K., Kempthorne, O. 2019).

RESULTS AND DISCUSSION

Taking into account methodical approaches to the collection, processing and visualization of data in the system of application of portable fluorometers (Posudin, Yu. et al. 2008), in the section of variants of the system of our research, an array of data indicators of the CFI curve was sorted (Table 4, Fig. 2 a–b).

According to the presented results, the photosystem of lentil plants reacts sensitively to the optimization of their nutrition, both when applying measures during pre-sowing seed preparation, and directly when carrying out a system of foliar fertilization. If in the first case it should be attributed to the factor of general optimization of growth and assimilation-forming processes, which was proved in the previous subsections, then in the second case it is directly related to the direct effect of microfertilizers on the photosystem of the leaf.

The detailed analysis of the formed block of indicators, taking into account the date of fixation and data collection, which is as close as possible to the reaching phase was done. The F0 criterion is considered the initial reading of the CFI curve and, in its essence, determines the overall activity of the photosystem of plant leaves. Its lower value indicates the slowness of reaction centers in terms of PAH excitation and the speed of implementation of energy transfer in these centers (Brestic, M., Zivcak, M. 2013; Kalaji, H.M. et al. 2017).

Based on this, the block of options where inoculation was not applied with the average value of F_0 at the level of 452.8 relative units of the fluorescence standard and the average of the options with the use of inoculation – 609.5 relative units of the fluorescence standard (comparative increase of 34.6%) allow us to emphasize the positive effect precisely inoculations in a prolonged perspective with an increasing effect of action from the sowing of treated seeds to the date of fixation of CFI indicators. The effect of seed treatment with microfertilizer was significantly lower and, compared to the average for options where this method is not used, the increase in the value of the indicator was 2.3% on the background without inoculation and 2.8% on the background with its application.

^{*-} $\frac{LTC}{0.1 \times \sum t_{.10}}$, where the amount of precipitation (ΣR) in mm over a period with temperatures above 10 °C, the sum of effective temperatures ($\Sigma t > 10$) over the same period, decreased by a factor of 10.

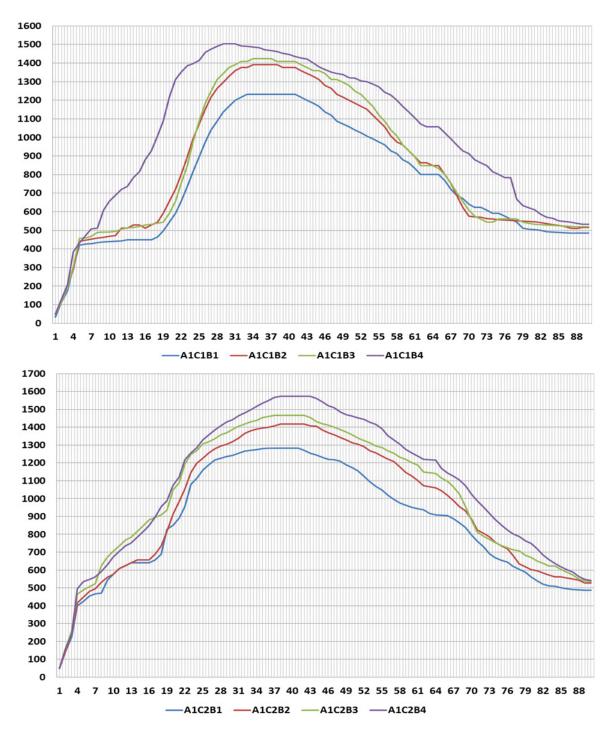
Table 3. Scheme of the experiment on the study of the combination of inoculation and the use of trace elements for the cultivation of lentils (pre-sowing fertilizer background for all variants $N_{30}P_{30}K_{30}$)

Seed inoculation with nitrogen- fixing microorganisms (factor A)	Pre-sowing treatment of seeds with trace elements (factor B)	Foliar fertilization with microfertilizers (factor C)					
		(C ₁) No feeding					
	Control (no treatment) (B ₁)	(C ₂) The active start of the PRO was activated (2.0 l/ha in the phase of the beginning of stalking (BBCH* 13–					
		(C ₃) Vanguard Complex Bobovi (2.0 l/ha in the phase of the beginning of budding (BBCH 53–55					
Without inoculation		(C ₄) Yarylo active start PRO (2.0 l/ha in the phase of the beginning of stemming (BBCH 13–15)) + Avangard Complex Beans (2.0 l/ha in the phase of the beginning of budding (BBCH 53–55))					
(A_1)		(C ₁) No feeding					
	Oracle seed (1 l/t) (B ₂)	(C ₂) The active start of the PRO was activated (2.0 l/ha in the phase of the beginning of stalking (BBCH* 13–15					
		(C ₃) Vanguard Complex Bobovi (2.0 l/ha in the phase of the beginning of budding (BBCH 53–55))					
		(C ₄) Yarylo active start PRO (2.0 l/ha in the phase of the beginning of stemming (BBCH 13–15)) + Avangard Complex Beans (2.0 l/ha in the phase of the beginning of budding (BBCH 53–55))					
		(C ₁) No feeding					
Inoculation Anderiz-r	Control (no treatment) (B ₁)	(C_2) The active start of the PRO was activated (2.0 l/ha in the phase of the beginning of stalking (BBCH* 13–1					
		(C ₃) Vanguard Complex Bobovi (2.0 l/ha in the phase of the beginning of budding (BBCH 53–55))					
		(C ₄) Yarylo active start PRO (2.0 l/ha in the phase of the beginning of stemming (BBCH 13–15)) + Avangard Complex Beans (2.0 l/ha in the phase of the beginning of budding (BBCH 53–55))					
(multicomponent inoculant (2 l/t))	Oracle seed (1 l/t) (B ₂)	(C ₁) No feeding					
(A ₂) "		(C ₂) The active start of the PRO was activated (2.0 l/ha in the phase of the beginning of stalking (BBCH* 13–15))					
		(C ₃) Vanguard Complex Bobovi (2.0 l/ha in the phase of the beginning of budding (BBCH 53–55))					
		(C ₄) Yarylo active start PRO (2.0 l/ha in the phase of the beginning of stemming (BBCH 13–15)) + Avangard Complex Beans (2.0 l/ha in the phase of the beginning of budding (BBCH 53–55))					

 $^{^{\}ast}-$ phase of lentil development according to the Zadoks scale.

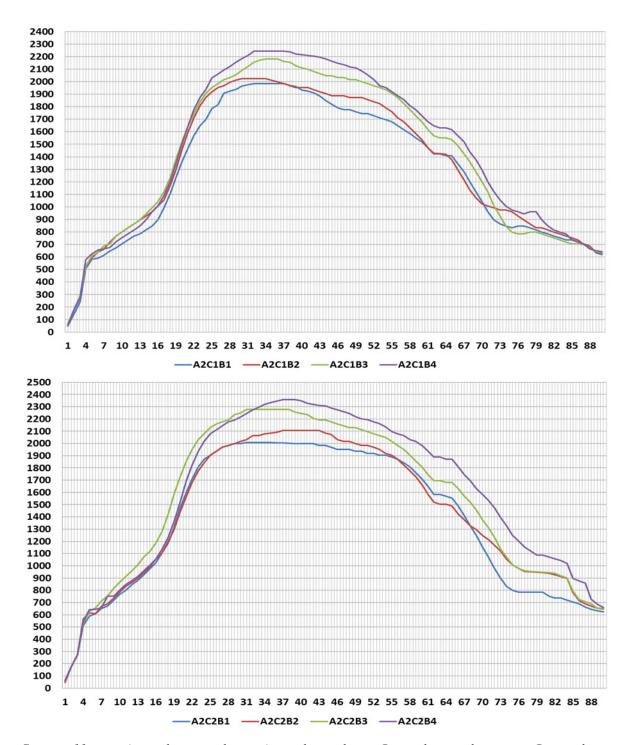
Table 4. Basic and calculated indicators of the chlorophyll fluorescence induction curve depending on the applied options for seed treatment and foliar fertilization of lentils of the Linza variety at the stage of bean formation (BBSN 65–68) (average for 2019–2021 in relative units of the fluorescence standard)

Seed inoculation (B)	Pre-sowing treatment of seeds (C)	Foliar fertilization (D)	$\mathbf{F_0}$	$\mathbf{F}_{ ext{pl}}$	$\mathbf{F}_{\mathtt{m}}$	F _{st}	dF _{pl}	F _v	dF_{pl}/F_{v}	EP
		1 (C ₁)	420	444	1232	484	24	812	0,030	0,659
	Control (no	2 (C ₂)	442	472	1392	516	30	950	0,032	0,682
Without inoculation	treatment)	3 (C ₃)	456	491	1424	519	35	968	0,036	0,680
	(B ₁)	4 (C ₄)	472	512	1504	533	40	1032	0,039	0,686
(A_1)		1 (C ₁)	424	471	1282	487	47	858	0,055	0,669
	Oracle	2 (C ₂)	448	496	1418	528	48	970	0,049	0,684
	seed (1 1/t) (B ₂)	3 (C ₃)	464	524	1467	535	60	1003	0,060	0,684
	(D ₂)	4 (C ₄)	496	562	1574	541	66	1078	0,061	0,685
		1 (C ₁)	581	648	1984	619	67	1403	0,048	0,707
	Control (no	2 (C ₂)	592	662	2024	627	70	1432	0,049	0,708
Inoculation	treatment) (B ₁)	3 (C ₃)	608	688	2180	634	80	1572	0,051	0,721
Anderiz-r	(D ₁)	4 (C ₄)	624	723	2244	642	99	1620	0,061	0,722
(multicomponent inoculant (2 l/t))		1 (C ₁)	588	651	2008	625	63	1420	0,044	0,707
(A ₂)	Oracle	2 (C ₂)	614	668	2105	648	54	1491	0,036	0,708
2.	seed (1 1/t) (B ₂)	3 (C ₃)	628	712	2279	655	84	1651	0,051	0,724
	(D_2)	4 (C ₄)	641	751	2358	659	110	1717	0,064	0,728
			L_{wp}	Q _{ue}	RF_d	\mathbf{K}_{ef}	QP	K _{prp}	\mathbf{K}_{fd}	$\mathbf{V}_{\mathbf{t}}$
	Control (no treatment) (B ₁)	1 (C ₁)	2,933	0,517	1,545	0,393	0,921	1,933	2,545	0,079
		2 (C ₂)	3,149	0,465	1,698	0,371	0,922	2,149	2,698	0,078
		3 (C ₃)	3,123	0,471	1,744	0,364	0,935	2,123	2,744	0,065
Without inoculation		4 (C ₄)	3,186	0,457	1,822	0,354	0,941	2,186	2,822	0,059
(A_1)		1 (C ₁)	3,024	0,494	1,632	0,380	0,927	2,024	2,632	0,073
	Oracle seed (1 l/t)	2 (C ₂)	3,165	0,462	1,686	0,372	0,918	2,165	2,686	0,082
	(B_2)	3 (C ₃)	3,162	0,463	1,742	0,365	0,929	2,162	2,742	0,071
	(-2)	4 (C ₄)	3,173	0,460	1,909	0,344	0,958	2,173	2,909	0,042
	Control (no treatment) (B ₁)	1 (C ₁)	3,415	0,414	2,205	0,312	0,973	2,415	3,205	0,027
		2 (C ₂)	3,419	0,413	2,228	0,310	0,976	2,419	3,228	0,024
Inoculation		3 (C ₃)	3,586	0,387	2,438	0,291	0,983	2,586	3,438	0,017
Anderiz-r (multicomponent		4 (C ₄)	3,596	0,385	2,495	0,286	0,989	2,596	3,495	0,011
inoculant (2 l/t))	Oracle seed (1 1/t) (B ₂)	1 (C ₁)	3,415	0,414	2,213	0,311	0,974	2,415	3,213	0,026
(A ₂)		2 (C ₂)	3,428	0,412	2,248	0,308	0,977	2,428	3,248	0,023
		3 (C ₃)	3,629	0,380	2,479	0,287	0,984	2,629	3,479	0,016
	· <u>2</u> ·	4 (C ₄)	3,679	0,373	2,578	0,279	0,990	2,679	3,578	0,010
LSD ₀₅	F_0					78; AC 7,7 D 15,56; B			35; BD 8,9 22,01	98;
	F _{pl} A 5,86; B 4,78; C 4,78; D 6,76; AB 8,28; AC 8,28; AD 11,72; BC 6,76; BD 9,57; CD 9,57; ABC 11,72; ABD 16,57; ACD 16,57; BCD 13,53; ABCD 23,43					57;				
	F_m	A 17,67; B 14,42; C 14,42; D 20,40; AB 24,98; AC 24,98; AD 35,33; BC 20,40; BD 28,85; CD 28,85; ABC 35,33; ABD 49,97; ACD 49,97; BCD 40,80; ABCD 70,66								
	F _{st} A 4,10; B 3,34; C 3,34; D 4,73; AB 5,79; AC 5,79; AD 8,19; BC 4,73; BD 6,69; CD 6,69; ABC 8,19; ABD 11,58; ACD 11,58; BCD 9,46; ABCD 16,38);		



 $\label{eq:content} \begin{subarray}{l} Content of factors: A_1-without inoculation; A_2-with inoculation; B_1-without seed treatment; B_2-seed treatment (Oracle 1 l/t); C-feeding: C_1-without feeding; C_2-Yarylo active start PRO; C_3-Avant-garde Complex Bobovi; C_4-Yarylo active start PRO + Vanguard Complex Bobovi \\ \end{subarray}$

Figure 2 a. Inducible changes in the fluorescence curves of chlorophyll of Linza lentil varieties in relative units of the fluorescence standard (vertical axis) depending on inoculation, seed treatment and the application of trace elements in top dressing for the phase of bean formation (BBCH 65–68) during on 90-second fixation period (horizontal axis), 2019–2021



 $\label{eq:content} \begin{subarray}{l} Content of factors: A_1-without inoculation; A_2-with inoculation; B_1-without seed treatment; B_2-seed treatment (Oracle 1 l/t); C-feeding: C_1-without feeding; C_2-Yarylo active start PRO; C_3-Avant-garde Complex Bobovi; C_4-Yarylo active start PRO + Vanguard Complex Bobovi. \end{subarray}$

Figure 2 b. Inducible changes in the fluorescence curves of chlorophyll of Linza lentil varieties in relative units of the fluorescence standard (vertical axis) depending on inoculation, seed treatment and the application of trace elements in top dressing for the phase of bean formation (BBCH 65–68) during on 90-second fixation period (horizontal axis), 2019–2021.

Increases in indicators when using foliar top dressings compared to the average for variants without its use amounted to 4.1% with a single top dressing at the beginning of stemming, 7.0% with a single top dressing at the beginning of budding and 10.9% with a combination of the specified top dressings during the lentil vegetation. The total increase in the comparison of the two limiting options in the experiment scheme showed a 52.6% higher value of F_0 in the option of combined and complex application of lentil nutrition optimization factors. The F_{pl} indicator (fluorescence of the 'plateau' zone) was the zone of achieving a temporary slowdown in the response signal of the photosystems of the leaf to excitation. The fluorescence indicator at the F_{pl} level was due to the rapid energy saturation of the reaction centers (RCs), which do not transfer energy to the electron transport chain (they do not reduce the primary acceptor QA and thus are reaction centers that do not restore the electron transport chain) (Brestic, M., Zivcak, M. 2013).

In its essence, the Fpl indicator expresses the stage of slowing down of the CFI curve and determines certain specific features of the levels of organization of the plant photosystem itself. At the same time, additional mineral nutrition due to the stabilizing optimization of the photosystem and the increase in the concentration of chlorophyll (which was noted in studies (Chen, J. et al. 2018)) contributes to the achievement of this point of the IPH curve at a higher value in conventional units of fluorescence. This was also confirmed in this studies. Thus, the average value of the indicator in the variants without the use of inoculation is 38.5% lower than with its application. Its growth was also noted in the variant of applying seed treatment with microfertilizer from 2.2% on the background without inoculation to 6.9% with it. The influence of extra-root nutrition was also positive -3.8% in the variant of one-time fertilizing at the beginning of stemming, 9.0% – for one-time fertilizing at the beginning of budding, and 15.1% for the combined application of microfertilizers (double application). It should also be noted the severity of this sections of the CFI curve for different variants of the experiment. Thus, in the variants without inoculation, the 'plateau' zone is clearly visible for 5–13 s of registration when mode with a slow transition to an intensive increase in the IPH curve in the area with an interval of 10–19 s of registration (Fig. 2 a) in the variants with inoculation, the specified zone is less noticeable and occupies the interval of time fixation of the device within 5–7 s with an intensive transition to the increasing section of the CFI curve (Fig. 2 b).

The dynamics of formation of the value of the maximum fluorescence indicator Fm, which characterizes the potential productivity of plant photosynthesis, had similar dynamics of changes as that of the F0 indicator. The value of F_m is positively correlated with the overall efficiency of the photosystem of plants in the transmission of an excitatory signal in the donor-acceptor physiological scheme of the photosystem of a plant leaf (Stirbet, A., Govindjee, 2011; Kalaji, H.M. et al. 2016). Its formation depended both on the physiological activity of the photosystem of the leaf and on the studied factors. Thus, the ratio between the inoculated and non-inoculated backgrounds of the variants was a ratio of 1.52. The increase from the application of seed treatment with microfertilizer was 3.4 and 3.8% on the background without inoculation and on the background with inoculation, respectively. The share of applied microfertilizers in top dressing was 6.7% in the Yarylo active start PRO application version, 13.0% in the Avangard Complex Bean application version, and 18.1% for its combination during the growing season.

The conclusions made above and the results of the assessment of another basic criterion of the CFI curve $-F_{st}$ – was confirmed. The degree of decrease in the level of chlorophyll fluorescence from maximum (F_{m}) to stationary (F_{st}) is often used as an integral indicator of the activity of the photosynthetic apparatus of plants (Stirbet, A., Govindjee, 2011).

It is noted (Flexas, J. et al. 2002) that the stationary level of Fst fluorescence shows the amount of chlorophylls that do not participate in the transfer of energy to the PSII reaction centers. The growth of this indicator indicates inhibition of the outflow of reduced photoproducts from the reaction centers due to adverse environmental factors (Strasser, R. et al. 2004). In the research of the authors (Guidi, L. et al. 2019; Larouk, C. et al. 2021) it is noted that the value of this indicator can be used to diagnose the intensity of the influence of a stress factor by estimating the time of its achievement on the CFI curve, the ratio of its value between the initial the level of induction of chlorophyll fluorescence (F_0) and its maximum value, as well as comparison of the difference between the value of F_0 and F_{st} . According to our estimates, its change had the least dependence on the studied factors of the experiment, considering the formation of this indicator to a greater extent as such, which is determined by the conditions of interaction

of physiological aspects of the photosystem caused by genetic nature in interaction with abiotic factors of the environment. Thus, the total increase in the value of the indicator of the inoculated background of the variants compared to the non-inoculated background amounted to 23.3% during the research period. The increase from the application of microfertilizers for seed treatment was 1.9% and 2.6%, respectively, on the variants without inoculation and on the variants with it. The use of foliar top dressings provided increases compared to the control without their use on average for all variants of the experiment system of 4.7%, 6.0% and 7.2%, respectively, for sequential application of top dressing options.

At the same time, it should be noted the different static stability of the lines of the CFI curves for different variants of the experiment. Thus, the highest fluctuations in the tropism of the points of this curve were noted in the variants without inoculation and seed treatment with microfertilizer (Fig. 2 a). On the contrary, the minimum amplitude of tropism of the test points was noted in the variants of the combination of inoculation, seed treatment and foliar fertilization (Fig. 2 b). It should also be noted that the application of foliar fertilization provides an increase in the specified fluctuations compared to the option without its application. In our opinion and in view of the research (Magney, T.S. et al. 2020), this character is due to the effect of direct interaction of the active substance of microfertilizer in a prolonged period with a direct system of getting through the leaves of the plant.

The calculated indicators of the CFI curve based on the basic indicators are presented in Table 4. According to the similar positive dynamics of their growth caused by their calculation on the basis of the analyzed indicators F_0 , F_{pl} . F_m and F_{st} , their brief analysis will allow to determine the optimality of the applied measures in the system of applied variants of the experiment. Thus, the F_v indicator, which determines the amplitude of the transition from the beginning of the CFI curve to its maximum value (F_m) and determines the speed of reaction of the reaction centers of the photosystem to photosynthetic excitation (Moustakas, M. et al. 2021), increases within 200 relative fluorescence units in the comparison of control variants and options with the use of two foliar feedings in each group of the interaction option of inoculation and seed treatment with microfertilizer.

The Lwp indicator, which characterizes the water potential of the leaf and determines the resistance of the assimilation apparatus of plants to atmospheric drought and low values of relative air humidity (Brestic, M., Zivcak, M. 2013) had a dynamic and stable growth in the section of the experiment variants with an increase of 13.1% in compared variants on the background without inoculation and with it and was gained of 2.9%, 6.0% and 6.6%, respectively, from the sequential staged application of foliar feeding.

The RF_d indicator, which determines the level of viability of the agrocenosis for this variant (Kalaji, H.M. et al. 2016), significantly increased with the sequential addition of lentil nutrition optimization measures and had a maximum value of 2.578 in the variant with the complex application of all factors of such optimization with an increase in the control of its complete absence in the amount of 66.8%. At the same time, due to a decrease in the rate of quantum quenching of chlorophyll fluorescence induction (Que indicator), an increase in the level of quantum energy output (QP indicator) according to (Murchie, E.H., Lawson, T. 2013; Ni, Z. Et al. 2019) the total duration of the change the time of flowering for options with the use of inoculation, treatment of seeds with microfertilizer and the use of two-time foliar feeding had significantly smaller values, which characterizes the state of the photosystem, was reactively more effective than in the option without the use of optimization factors.

According to the value of the such criteria L_{wp} , RF_{d} , K_{prp} and K_{fd} , which determine the presence of abiotic pressure on the assimilation apparatus of plants (Wang, H. et al. 2018; Sánchez-Moreiras, A.M. et al. 2020), the application of inoculation against the background of seed treatment with microfertilizer and improvement of physiological growth processes due to the application of two foliar feedings in the critical phases of growth and development of lentils provided a general reduction in the stress state of plants and an increase in its vitality.

CONCLUSIONS

Based on the main parameters of the CFI curve (F_0 , F_{pl} , F_m , F_{st}), it was established that the photosystem of lentil plants responds sensitively to the optimization of their nutrition, both when applying measures during pre-sowing seed preparation, and directly when carrying out a system of foliar fertilization. The resulting increase with the application of inoculation was noted by 34.6% for the index of

initial flowering (F_0), by 38.5% for the index of flowering of the 'plateau' zone ($F_{\rm pl}$), by 52.1% for the index of maximum flowering ($F_{\rm m}$) and by 23.3% for stationary florescence ($F_{\rm st}$). For the same indicators, the increase compared to the control from the application of seed treatment with trace elements on the background without inoculation was 2.3%, 6.9%, 3.4%, 1.9%, and on the background with inoculation was 2.7%, 7, 2%, 3.7% and 2.6%, respectively. A single foliar top dressing at the beginning of stemming compared to the average of the control options without top dressings provided an increase in the same basic indicators of the CFI curve by 4.1%, 3.8%, 6.7% and 4.7%. Similar increases in top dressing options at the beginning of budding were 7.1%, 9.1%, 13.0% and 5.8%, respectively. The maximum increases from foliar fertilization were noted in the variant of combined application of microfertilizers – 10.9%, 15.1%, 18.0% and 7.2%, respectively.

Due to the positive increase in the values of the basic indicators of the IPH curve (F_0 , F_{pl} , F_m , F_{st}), an increase in the efficiency of the photosystem of lentil plants in the comparison of the marginal studied technological options for optimizing lentil nutrition in terms of the growth of the agrocenosis viability index (RF_d) by 66.8% was noted. The water potential of leaves (L_{wp}) on the level of 13.1% was determined, increasing the stress resistance of plants to abiotic factors due to increasing the energy efficiency of the photosystem (according to indicators, the value of photochemical quenching of fluorescence (QP - by 2.1%) and reducing the indicator of photochemical quenching (Q_{ue} – in the interval from 5.2 to 16.2% depending on the option) against the background of the decline in the relative fluorescence change at time t (Vt).

REFERENCES

- 1. BRESTIC, M., ZIVCAK, M. (2013). PSII fluorescence techniques for measurement of drought and high temperature stress signal in plants: protocols and applications. In: Rout G.R., Das A.B., eds. Molecular stress physiology of plants, pp. 87-131.
- 2. CHEN, J., BURKE, J.J., XIN, Z. (2018). Chlorophyll fluorescence analysis revealed essential roles of FtsH11 protease in regulation of the adaptive responses of photosynthetic systems to high temperature. In: BMC Plant Biology, vol. 18 (1), p. 11.
- 3. CHEN, X., MO, X., HU, S., LIU, S. (2019). Relationship between fluorescence yield and photochemical yield under water stress and intermediate light conditions. In: Journal of Experimental Botany, vol. 70 (1), pp. 301-313.
- 4. FLEXAS, J., ESCALONA, J.M., EVAIN, S., GULÍAS, J., MOYA, I., OSMOND, C.B., MEDRANO, H. (2002). Steady-state chlorophyll fluorescence (Fs) measurements as a tool to follow variations of net CO2 assimilation and stomatal conductance during water-stress in C3 plants. In: Physiologia Plantarum, vol. 114 (2), pp. 231-240.
- 5. GORBUNOV, M.Y., FALKOWSKI, P.G. (2021). Using chlorophyll fluorescence kinetics to determine photosynthesis in aquatic ecosystems. In: Limnology and Oceanography, vol. 66 (1), pp. 1-13.
- 6. GOVINDJEE (2004). Chlorophyll a fluorescence: a bit of basics and history. In: Chlorophyll a Fluorescence: a Signature of Photosynthes, Advances in Photosynthesis and Respiration, vol. 19, pp. 1-41.
- GUIDI, L., LO PICCOLO, E., LANDI, M. (2019). Chlorophyll Fluorescence, Photoinhibition and Abiotic Stress: Does it Make Any Difference the Fact to Be a C3 or C4 Species? In: Frontiers in Plant Science, vol. 10, p. 174.
- 8. HINNKELMANN, K., KEMPTHORNE, O. (2019). Design and Analysis of Experiments, 10th Edition. Vol. 1. New York: Wiley and Sons, 688 p. ISBN 978-1-119-49244-3.
- KALAJI, H.M., SCHANSKER, G., BRESTIC, M., BUSSOTTI, F., CALATAYUD, A., FERRONI, L., SHE-LONZEK, H. (2016). Frequently asked questions about chlorophyll fluorescence, the sequel. In: Photosynthesis Research, vol. 132, Is. 1, pp. 13–66.
- KALAJI, H.M., RASTOGI, A., ŽIVČÁK, M. (2018). Prompt chlorophyll fluorescence as a tool for crop phenotyping: an example of barley landraces exposed to various abiotic stress factors. In: Photosynthetica, vol. 56, pp. 953-961.
- 11. KALAJI, H.M., SCHANSKER, G., BRESTIC, M., BUSSOTTI, F., CALATAYUD, A., FERRONI, L., GOLT-SEV, V., GUIDI, L., JAJOO, A., LI, P., LOSCIALE, P., MISHRA, V.K., MISRA, A.N., NEBAUER, S.G., PANCALDI, S., PENELLA, C., POLLASTRINI, M., SURESH, K., TAMBUSSI, E., YANNICCARI, M., ZIVCAK, M., CETNER, M.D., SAMBORSKA, I.A., STIRBET, A., OLSOVSKA, K., KUNDERLIKOVA, K., SHELONZEK, H., RUSINOWSKI, S., BĄBA, W. (2017). Frequently asked questions about chlorophyll fluorescence, the sequel. In: Photosynthesis Research, vol. 132(1), pp. 13-66.

- 12. KARGAR, M., GHORBANI, R., RASHED MOHASSEL, M.H., RASTGOO, M. (2019). Chlorophyl fluorescence a tool for quick identification of Accase and ALS inhibitor herbicides performance. In: Planta Daninha, vol. 37. Available: https://doi.org/10.1590/S0100-83582019370100132
- 13. KORRES, N.E., FROUD-WILLIAMS, R.J., MOSS, S.R. (2003). Chlorophyll fluorescence technique as a rapid diagnostic test of the effects of the photosynthetic inhibitor chlorotoluron on two winter wheat cultivars. In: Annals of Applied Biology, vol. 143 (1), pp. 53-56.
- 14. LAROUK, C., GABON, F., KEHEL, Z., DJEKOUN, A., NACHIT, M., AMRI, A. (2021). Chlorophyll Fluorescence and Drought Tolerance in a Mapping Population of Durum Wheat. In: Contemporary Agriculture, vol. 70, Issue 3-4, pp. 123-134.
- 15. MAGNEY, T.S., BARNES, M.L., YANG, X. (2020). On the covariation of chlorophyll fluorescence and photosynthesis across scales. Geophysical Research Letters, 47. e2020GL091098.
- MARTINAZZO, E.G., RAMM, A., BACARIN, M.A. (2012). The chlorophyll a fluorescence as an indicator of the temperature stress in the leaves of Prunus persica. Brazilian Journal of Plant Physiology. 24(4). P. 237–246. Available: https://doi.org/10.2478/contagri-2021-0018
- 17. MOUSTAKAS, M., CALATAYUD, A., GUIDI, L. (2021). Chlorophyll fluorescence imaging analysis in biotic and abiotic stress. In: Frontiers in Plant Science, vol. 12.
- 18. MURCHIE, E.H., LAWSON, T. (2013). Chlorophyll fuorescence analysis: A guide to good practice and understanding some new applications. In: Journal of Experimental Botany, vol. 64 (13), pp. 3983–3998.
- 19. NI, Z., LU, Q., HUO, H., ZHANG, H. (2019). Estimation of Chlorophyll Fluorescence at Different Scales: A Review. Sensors (Basel), vol. 19(13), p. 3000.
- POSUDIN, Y.U., MELNYCHUK, M., KOZHEM'YAKO, Y.A., ZALOILO, I., GODLEVSKA, O. (2010). Application of portable fluorometer for estimation of plant tolerance to abiotic factors. In: International Agrophysics, vol. 24, pp. 363-368.
- 21. ROMANOV, V.A., GALELYUKA, I.B., BABENKO-SARAKHAN, Y. (2010). Portable fluorometer Floratest and specifics of its application. In: Sensor Electronics and Microsystem Technology, vol. 7 (3), pp. 39–44.
- 22. RUBAN, A.V. (2016). Nonphotochemical chlorophyll fluorescence quenching: Mechanism and effectiveness in protecting plants from photodamage. In: Plant Physiology, vol. 170 (4), pp. 1903–1916.
- 23. SÁNCHEZ-MOREIRAS, A.M., GRAÑA, E., REIGOSA, M.J., ARANITI, F. (2020). Imaging of Chlorophyll a Fluorescence in Natural Compound-Induced Stress Detection. In: Frontiers in Plant Science, vol. 11, pp. 1-15.
- 24. STIRBET, A., GOVINDJEE (2011). On the relation between the Kautsky effect (chlorophyll a fluorescence induction) and photosystem II: Basis and applications of the OJIP fluorescence transient. In: Journal of Photochemistry and Photobiology B, vol. 104 (1-2), pp. 236-257.

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