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SHORT COMMUNICATION

Analysis of the impact of drought on selected morphological, biochemical and physiological traits of rye inbred lines

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Abstract The complex nature of plant resistance to drought makes the process of selecting the appropriate genes that increase the resistance to drought very difficult. With the future in mind, the aim of our study was to search for physiological and biochemical parameters which could provide a basis for the identification of genes controlling rye resistance to drought stress. The experiments were carried out on three inbred lines of rye: S120, S76 and M112, a recombinant inbred line of population RIL-M; lines in the tillering phase were subjected to drought stress for 4 weeks. Selected physiological indicators of PSII photochemical system [chlorophyll *a* fluorescence kinetics (FC) and photosynthetic pigment contents (PPC)], biochemical indicators (proline, soluble sugars, total phenolics) and selected agronomic traits were determined. Drought did not significantly affect the majority of the measured FC and PPC parameters in any of the three lines. Due to the different reactions of the lines to drought stress, depending on the analyzed characteristics, the authors concluded that the analyzed indicators can be used to study QTL locations in response to drought stress in the RIL-M mapping population of rye.

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Introduction

Drought stress is one of the most common and important abiotic stress factors. Breeding programs aimed at increasing yield have usually attempted to improve drought tolerance of plants. Rye (*Secale cereale* L.), belonging to the tribe *Triticeae*, stands out from other cereals in terms of exceptional drought tolerance. It withstands adverse conditions better than wheat, oat or barley. Its drought resistance and ability to endure sand blasts enable rye to produce a soil-binding cover on lands where other cereals do not grow, e.g. in Australia, with its extensive arid regions (Schlegel 2013). However, rye is grown mainly in Central and Eastern Europe. Water deficit was not a serious problem for rye breeders due to its high resistance, and thus rye was not the subject of research in this area. However, we can expect that rye, as well as wild relatives (e.g. wild emmer), with their adaptive complexes to abiotic stresses offer a rich allelic repertoire of agronomically valuable traits for related species. They are perceived as the most promising cereals for crop improvement (Peleg et al. 2005, 2009; Tuberosa and Salvi 2006; Xie and Nevo 2008). Rye has already been used as a source of agronomically important genes for wheat; the short arm of rye chromosome 1 (1RS) has been introgressed into several hundreds of wheat cultivars (Bartoš et al. 2008). Obviously, prior to any genetic manipulation, it is important to characterize the genetic basis of different adaptive mechanisms to stressful conditions. A better knowledge of the rye genome and genes responsible for drought tolerance could both facilitate rye improvement and increase the efficiency of utilizing rye genes in wheat breeding. Many morphological,

biochemical and physiological traits needs to be thoroughly analyzed, because of the complex nature of drought tolerance. Drought affects growth, yield, membrane integrity, pigment content, osmotic adjustment, water relations, and photosynthetic activity. Therefore, the tolerance of crop species can be characterized by different parameters related to growth response, stomatal conductance, ion accumulation, photosynthetic machinery reaction, phospholipid signaling mechanisms as well as the content of sugars, other osmolytes and antioxidant compounds (Mahajan and Tuteja 2005; Anjum et al. 2011).

Our goal was to initiate a study of rye focusing on certain physiological and biochemical parameters in correlation with yield components in standard and drought stress conditions using three inbred lines of winter rye. Two of them served as parental lines for a rye mapping population, which provides an opportunity to expand the research to a larger set of plant genotypes with the possibility of mapping regions of the rye genome associated with drought response. We measured parameters of photosystem activity (PSII), the content of pigments (chlorophyll $a+b$ and carotenoids), proline, soluble sugars, total phenolics and selected agronomic traits. As the selection of suitable drought tolerant genotypes in the field is time-consuming and difficult for plant breeders, a search for quick and relatively easy screening methods of suitable genotypes for drought-prone areas was conducted.

Materials and methods

Plant material

The experiments were carried out on three inbred lines of rye: S120, S76 (parental lines) and one (M120) recombinant inbred line of population RIL-M, derived from the cross between S120 and S76. The parental lines were developed within a commercial breeding program conducted in the Plant Breeding Danko Ltd. (Choryń, Poland). These lines are partly related, but they differ genetically (Myśków et al. 2001; Milczarski et al. 2011). RIL-M is a mapping population obtained in ZUT, used to obtain a high-density genetic map of rye (Milczarski et al. 2011) and analysis of QTL controlling some morphological traits (Myśków et al. 2014), earliness and pre-harvest sprouting (Myśków et al. 2012).

Plant growth conditions

Three inbred lines (S120, S76 and M12) were used for phenotyping of biochemical traits during the experiment conducted in Krakow, Poland. The seedlings after an 8-week vernalization were placed in pots (Ø15 cm, 20 cm high, one seedling per pot) filled with a mixture of soil and sand in equal proportions by volume. At the beginning of the experiment, the pots were filled with the same soil mass (1.700 kg) and the same water content. Several pots were selected to determine the soil FWC (field water capacity). The pots were watered with the same amount of water and weighted. We determined the moment when the water stopped flowing out from the bottom of every pot (about 2 days). The moment when the pots with soil reached a stable weight was determined as the permanent wilting point (about 15% FWC). In our study, 20–25% FWC was adopted as drought (D) and 65–70% FWC as well-watered (control). The plants were watered with an appropriate volume of water, approximately for every level of drought and control, on the basis of plant viability and soil appearance. Every few days, the weight of some pots was controlled to determine the water volume for watering. The plants were grown in a rain-out shelter, close to natural conditions, with natural daylight duration and air temperature of the spring–autumn period (May–September). Differentiation of water content in the soil began in the phase of tillering. Drought stress was continued for 4 weeks. The kinetics of chlorophyll a fluorescence (FC) was measured on the last day of the drought-stress treatment, followed by the collection of flag leaves of main shoots for biochemical measurements. The analysis was performed in nine replicates.

Measurements

Chlorophyll "a" fluorescence parameters (FC)

FC was measured on the flag leaf using a Handy PEA portable fluorometer (Hansatech Instruments, King's Lynn, UK) at ambient temperature, after 20 min adaptation of leaves to dark conditions on the last day of drought stress. Technical details of the method are described by Czyczyło-Mysza et al. (2013). On the basis of chlorophyll fluorescence measurements, the theory of energy flow in PS II, and using the OJIP test proposed by Strasser et al. (2000), the following parameters were calculated and analyzed: F_v/F_m (the maximum photochemical efficiency), F_v/F_0 (a value that is proportional to the activity of the water-splitting complex on the donor side of PSII), PI (overall performance index of PSII photochemistry), ABS/CS (light energy absorption), TRo/CS (amount of excitation energy trapped in PSII reaction centers), DIo/CS (energy amount dissipated from PSII, RC/CS (number of active reaction centers) and ETo/CS (amount of energy used for electron transport).

Biochemical measurements

Frozen detached flag leaves of the main shoots of control plants and of those subject to drought conditions were

lyophilized for 72 h, then powdered in a MM 400 mixing mill ball grinder (Retsch, Kroll, Germany). Samples with mass suitable for particular biochemical analyses were weighed on a micro-analytical balance.

Photosynthetic pigments content (PPC)

The concentrations of total chlorophyll $a+b$ (TChl) and carotenoids (Car) were calculated according to extinction coefficients given in the equations of Lichtenthaler and Buschmann (2001) as described by Czyczyło-Mysza et al. (2013).

Proline content (PC)

Proline content was measured spectrophotometrically according to Ting and Rouseff (1979) with modifications, as described by Marcińska et al. (2013).

Soluble sugars content (SSC)

Sugars content was analysed spectrophotometrically, according to the method of Dubois et al. (1956), modified by Marcińska et al. (2013).

Total phenolics content (TPC)

Total phenolics content was measured according to the modified method by Singleton and Rossi (1965). The samples (100–250 mg of fresh weight) were homogenized in 2 cm^3 of 96% ethanol and centrifuged (2100×g for 15 min). If necessary, the supernatants were diluted with distilled water. An aliquot of the extract (0.1 cm^3) was transferred to a test tube containing 0.5 cm^3 of 25% Na_2CO_3, and then 0.125 cm^3 Folin–Ciocalteu reagent (diluted 1/1 with distilled water before use) was added. The samples were vortexed, transferred to 96-well micro-plates after a 30-min incubation, and absorbance at 760 nm was read. Chlorogenic acid was used as a standard.

Agronomic traits

At final maturity, the plants were cut at the soil surface, weighed after drying to obtain the above-ground biomass and separated into the main shoot and remaining parts. For each plant, the number of grains per plant (NG), weight of grains per plant (YP) and dry weight per plant at harvest (BIOMASS) were measured.

Statistical analysis

Results presented in the figure and table are mean values based on nine replicates. Data were analyzed using STATISTICA 12.0 software package (Stat-Soft, Inc., USA); ANOVA [Fisher–Snedecor test (F) and Duncan's multiple range test at $P \leq 0.05$] and Principal component analysis (PCA).

Results

Principal component analysis (PCA) and ANOVA analysis

PCA analysis was carried out for physiological traits (FC), biochemical traits (PPC, PC, SSC, TPC) and yield component parameters (NG, YP, BIOMASS) of plants, whose vegetation was analyzed in two environmental conditions (C, D). The control demonstrated 46.9% of the variability observed for PC1 and 24.2% of the variability for PC2; the total variability for all traits amounted to 71.1% (Fig. 1). These values under drought were 39.7 and 25.2% for PC1 and PC2, respectively, and 64.9% of the total variability. Both under control and drought conditions, a positive (acute angle between the measured parameters), negative (obtuse angle), and no correlation (right angle) was observed. A strong positive correlation was recorded for the control between yield parameters: BIOMASS, NG and the FC parameter (PI), between soluble sugar content (SSC) and FC parameters (ABS/CS and TRo/CS). A weaker positive correlation occurred between DIo/SC and Car and TPC. A strong negative correlation was observed between the DIo/SC parameter and yield components and FC parameters (PI, F_v/F_m and F_v/F_0). A moderate negative correlation was observed between PC and SSC and parameters related to electron transport. Under drought, a positive correlation was also observed between the biomass and F_v/F_m and F_v/F_0 parameters. There was no positive correlation between SSC and FC parameters, in contrast to the control. However, there were strong positive correlations between photosynthetic pigments and ABS/CS, TRo/CS and Eto/CS FC parameters. Similarly, as in control, a moderate negative correlation also occurred between PC and SSC content, and between DIo and biomass, as well as a strong negative correlation between DIo and F_v/F_m, F_v/F_0, PI, ABS/CS and TRo/CS.

F-statistics (Fisher test) was calculated using ANOVA (STATISTICA 12.0) to establish significance of differences for line, treatment, and line × treatment interaction of the all measured parameters (Table 1). Analysis of variance showed highly significant differences between line, treatment, and line × treatment interaction for PC parameter. ANOVA analysis for following parameters: ABS/CS, ETo/CS, TRo/CS, TChl, Car, PC, SSC, BIOMASS and NG showed significant differences between tested lines. In addition, highly significant interactions

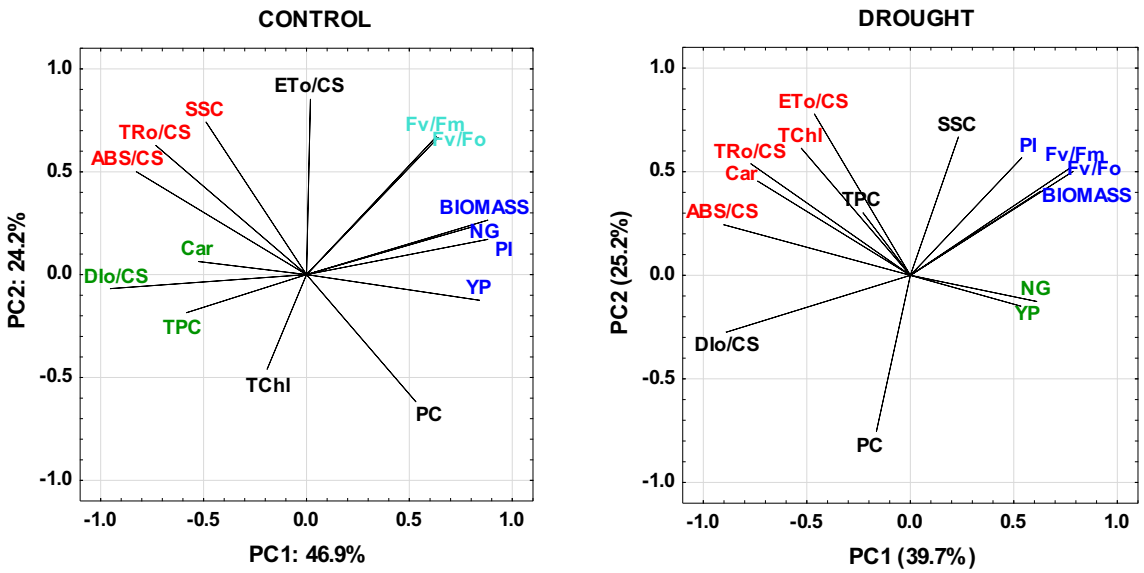

Fig. 1 Vector view of biplot showing interrelationship among studied traits under non stress (*control*) and water stress (*drought*) conditions. Traits marked by the *same color* are positively correlated

between lines and treatment were recorded for PC and TChl. No significant differences were observed between *F*-statistics measured for PI and NG.

Chlorophyll *a* fluorescence parameters

The PSII function index (PI) did not demonstrate significant variability among the discussed kinetic parameters of chlorophyll *a* fluorescence (FC) in all lines tested, under both optimal watering and drought stress (Table 1). All lines tested showed a decrease in the maximum quantum yield of PSII (F_v/F_m), maximum efficiency of the water dissociation reaction on the donor side of PSII (F_v/F_0) and an increase in excitation energy dissipation as heat (DIo/CS). However, it was significant only in the paternal line S76. Other parameters, indicative of electron transfer efficiency in the light phase, did not demonstrate decreasing trends, while for line M112, they were even significantly higher, compared with the control, for the following parameters: ABS/CS, Eto/CS, and TRo/CS (by 7, 10 and 6%, respectively). The photosynthetic apparatus in line M112 was more active and functioned better, compared with the parental lines, giving better results also under drought stress.

Photosynthetic pigments content

After 4 weeks of drought, no significant differences in TChl and Car contents between both parental lines under drought treatment were observed (Table 1). Pigment contents remained at a similar level, both under drought and control conditions for a given line. A higher content of pigments was found in the paternal line S76. The highest significant variability between treatments was recorded in the progeny line M112, where under drought stress, an increase in chlorophyll *a*+*b* and carotenoid contents was observed of 45 and 37%, respectively.

Proline content (PC)

PC in control plants was significantly different between line S76 in comparison to the remaining two genotypes (Table 1). An increase of proline in all rye lines tested was observed under water deficit. Only the paternal line (S76) was characterized by the significant increase in PC (136%) on drought conditions compared to the control. This line also showed an almost twofold higher proline increase under drought compared with other lines. Lines S120 and M112 showed no significant increase in proline.

Soluble sugars content (SSC)

The highest SSC content under optimal watering was observed in lines S120 and M112 (approx. 90 μg/mg). Rye lines tested showed a similar response to soil drought stress in terms of sugar contents in leaves. Drought stress caused a similar decrease in soluble sugar contents in leaves (24–34%), though this was significant only in line S120 (34%) compared with the control (Table 1).

Total phenolics content (TPC)

Of the three rye lines tested, line S120 showed the highest TPC content in the control (Table 1). Parental rye lines

Table 1 Mean values (±standard errors) for phenotypic traits related to chlorophyll "*a*" kinetics fluorescence parameters

Traits	Lines						*F*-statistics		
	S76		S120		M112				
	C	*D*	*C*	*D*	*C*	*D*	Lines	Treatment	Lines × treatment
F_v/F_0	5.08 ± 0.16^{b}	4.52 ± 0.249^{a}	4.96 ± 0.11^{ab}	4.72 ± 0.218^{ab}	5.20 ± 0.07^{b}	4.97 ± 0.110^{ab}	1.7	6.5*	0.6
PI	4.58 ± 0.23^{a}	3.75 ± 0.484^{a}	4.00 ± 0.40^{a}	3.88 ± 0.391^{a}	3.67 ± 0.16^{a}	3.93 ± 0.166^{a}	0.6	0.7	1.3
F_v/F_m	0.835 ± 0.004^{b}	0.816 ± 0.009^{a}	0.832 ± 0.003^{ab}	0.823 ± 0.008^{ab}	0.839 ± 0.002^{b}	0.832 ± 0.003^{ab}	1.7	5.9*	0.6
ABS/CS	335 ± 9.64^{a}	357 ± 11.68^{ab}	347 ± 3.74^{ab}	360 ± 8.00^{ab}	363 ± 5.85^{b}	387 ± 3.83^{c}	7.9**	9.4**	0.3
ETo/CS	179 ± 5.55^{a}	181 ± 2.28^{a}	176 ± 2.98^{a}	186 ± 6.10^{a}	181 ± 2.27^{a}	199 ± 3.35^{b}	3.9*	10.2**	2.1
TRo/CS	280 ± 7.05^{a}	290 ± 6.18^{abc}	289 ± 2.94^{ab}	296 ± 4.68^{bc}	305 ± 4.61^{c}	322 ± 2.8^{d}	18.3***	8.6**	0.6
DIo/CS	55 ± 2.89^{a}	66 ± 5.69^{b}	58 ± 1.40^{ab}	64 ± 4.02^{ab}	59 ± 1.39^{ab}	65 ± 1.64^{ab}	0.1	7.8**	0.3
TChl (μg/mg)	7.92 ± 0.43^{c}	7.73 ± 0.47^{bc}	5.72 ± 0.47^{a}	6.36 ± 0.58^{ab}	6.67 ± 0.25^{abc}	9.67 ± 0.567^{d}	12.2***	8.2**	5.7**
Car (μg/mg)	0.65 ± 0.12^{ab}	0.76 ± 0.03^{ab}	0.56 ± 0.07^{a}	0.55 ± 0.085^{a}	0.84 ± 0.07^{b}	1.15 ± 0.038^{c}	20.3***	4.8*	2.6
PC (μg/mg)	0.229 ± 0.023^{b}	0.540 ± 0.039^{c}	0.089 ± 0.008^{a}	0.186 ± 0.050^{ab}	0.111 ± 0.03^{a}	0.121 ± 0.039^{ab}	33.3***	21.7***	8.3***
SSC(μg/mg)	69.82 ± 5.83^{ab}	49.56 ± 7.78^{a}	94.39 ± 5.68^{b}	62.76 ± 4.94^{a}	91.74 ± 7.65^{b}	70.00 ± 12.11^{ab}	4.2*	13.9***	0.3
TPC (μg/mg)	7.44 ± 0.394^{a}	6.10 ± 0.594^{a}	9.026 ± 0.529^{b}	6.16 ± 0.400^{a}	7.180 ± 0.345^{a}	7.11 ± 0.374^{a}	1.7	15.1***	5.1*
BIOMASS (g)	4.95 ± 0.36^{bc}	3.42 ± 0.37^{a}	5.95 ± 0.41^{c}	5.13 ± 0.20^{c}	5.70 ± 0.91^{c}	3.88 ± 0.243^{ab}	6.9**	18.9***	0.9
NG (g)	43 ± 8.70^{ab}	18 ± 7.29^{ab}	58 ± 15.28^{ab}	71 ± 12.62^{b}	82 ± 46.51^{b}	0.13 ± 0.245^{a}	3.4*	3.1	2.3
YP (g)	0.78 ± 0.216^{abc}	0.33 ± 0.12^{ab}	0.74 ± 0.218^{abc}	1.06 ± 0.22^{ab}	1.57 ± 1.128^{c}	0.013 ± 0.001^{a}	1.0	5.2*	4.4*

On the right side *F*-statistics for line, treatment, and line × treatment interaction of the measured parameters

F_v/Fo the ratio of the variable to minimal fluorescence, *PI* overall performance index of PSII photochemistry, F_v/F_m the maximum photochemical efficiency, *ABS/CS* light energy absorption, *ETo/CS* amount of energy used for electron transport, *TRo/CS* amount of excitation energy trapped in PSII reaction centers, *DIo/CSm* energy amount dissipated from PSII, photosynthetic pigments (*TChl* total chlorophyll "*a*+*b*", *Car* carotenoids), proline (PC), soluble sugars (SSC) and phenolics (TPC) in leaves and productivity (*BIOMASS* dry weight per plant at harvest, *NG* the number of grains per plant, *YP* weight of grains per plant) of three rye inbred lines (S76, S120 and M112) under control (*C*) and drought (*D*) conditions

Significance levels: * $P \leq 0.05$; ** $P \leq 0.01$; *** $P \leq 0.001$

[a,b,c] marked with the same letters (for each parameter separately) differ not significantly, according to Duncan's test ($P \leq 0.05$), $n = 9$

were characterized by reduced contents of phenolic compounds in the leaves of plants grown in soil under drought conditions compared with control plants. However, this decrease was significant only for line S120. This line had 32% less TPC compared with the control, whereas the progeny line M112 showed the same TPC level both under drought and control conditions.

Agronomic traits

Yield components varied greatly in both treatments (*C*, *D*), with significant differences observed in all yield parameters for line M112 and high declining but non-significant differences for yield components in the paternal line S76 (Table 1). Line M112 did not form seeds in some plants under drought. As regards line S120, there were no significant differences found between the control plants and the plants treated with drought stress, and even an increasing number and weight of seeds under stress was observed, which might indicate its greater tolerance to soil drought.

Discussion

Adverse weather conditions (especially drought), in recent years often lasting for a long part of the crop growing season, have a negative effect on the yield of plants, not only of sensitive, but also resistant species, such as rye. Many research groups are studying the development, importance and inheritance of key traits associated with drought responses in plants and their genetic control, which was recently described by Tuberosa (2012). The literature on drought stress in rye lacks research concerning the identification of markers linked to drought tolerance genes associated with biochemical and physiological indices. To our knowledge, we present the first comprehensive experiments involving physiological and biochemical parameters in relation to yield components in rye during well-watered and drought conditions.

A water deficit in plant tissues under drought stress leads to a significant inhibition of photosynthesis. The plant reacts to water deficit with a rapid closure of stomata to avoid further loss of water through transpiration. Photosynthetic electron transport through PSII is inhibited, the oxygen evolving complex of PSII and the PSII reaction centers associated with the degradation of D1 protein are damaged (Zlatev 2009). Therefore, the ability to maintain the functionality of the photosynthetic machinery under water stress is of major importance in drought tolerance. This study shows that drought did not significantly affect the FC parameters in rye, which may be, *inter alia*, related to the resistance of rye plants to drought. The PI parameters (PSII performance index), which according to some authors (Rapacz 2007; Rapacz and Woźniczka 2009; Czyczyło-Mysza 2013) seem to be one of the most useful FC parameters, exhibited the lowest variation among the analyzed FC parameters in various kinds of stresses. In contrast, F_v/F_m that determines the quantum efficiency of PSII, but does not provide complete information about the photochemical properties of PSII, showed the highest variability. As the decrease of F_v/F_m was not accompanied by symptoms of electron transport disorders, according to Antonkiewicz and Rapacz (2006) this should not be regarded as a manifestation of damage, but rather a sign of adaptive or developmental changes. However, different reactions of lines to FC parameters indicate that these parameters can be used in QTL localization in response to drought stress or under conditions of optimal watering, which was confirmed in the studies of Yang et al. (2007), Bai et al. (2011), Van Heerden et al. (2007), Molik et al. (2014) and Czyczyło-Mysza et al. (2011).

Water stress also has the ability to reduce concentrations of chlorophylls and carotenoids, primarily by the production of ROS (reactive oxygen species) in the thylakoids. Both chlorophyll *a* and *b* are prone to soil drying. Photosynthetic pigments are used by plants mainly for light harvesting and production of reducing power. Low concentrations of photosynthetic pigments can directly limit photosynthetic potential, and thus primary production (Anjum et al. 2011). Carotenoids play additional roles and partially help the plants to withstand drought stress. Carotenes form a key part of the plant antioxidant defense system, but they are very susceptible to oxidative damage (Jaleel et al. 2009). In our research, no lines showed any decrease in photosynthetic pigments, which may indicate their protective role during drought in rye.

Phenolics, as with carotenoids, are known antioxidants helping to prevent cellular damages caused by oxidative stress. Phenolic compounds can act as metal chelators and can directly scavenge molecular species of active oxygen, but their exact role in plant stress responses in nature is still under debate (Bautista et al. 2016). A number of studies on winter triticale (Hura et al. 2007, 2009, 2010) confirmed that the TPC in leaves depends on the development phase of the plant, variety and different soil–water content. In our experiment, PSII reaction centers of line M112, with a higher TPC content under drought stress, compared with the parental lines, trapped excitation energy more efficiently, and then directed this energy to further photochemical reactions with simultaneous insignificant energy loss as heat (DI_O/CS). This is consistent with the study of Hura et al. (2009), who found that a variety with a more active photosynthetic apparatus was characterized by higher levels of phenolic compounds in triticale under drought stress applied in the vegetative phase of its growth. A decrease in TPC observed on the last day of drought in parental forms could be due to the fact that after/during the 4-week drought, these

compounds could have been used, *inter alia,* for strengthening the cell wall, probably during lignin synthesis. It is possible that soluble phenolics can be built into the cell wall structures with the involvement of hydrogen peroxide and peroxidase. Hura et al. (2012), based on their own and other studies, emphasized that saturation of the cell wall with phenolics leads to a limited utilization of carbohydrates, which could also be explained by the decrease of SSC on the last day of drought in the present research in the drought treatment compared with the control.

Various organic molecules, such as sugars, mono- and oligosaccharides, sugar alcohols (mannitol, sorbitol), polyols, amino acids and their derivatives, including proline and inorganic ions such as K^+ help the cells to maintain their hydrated state, thereby providing resistance against drought and cellular dehydration (Mahajan and Tuteja 2005). According to Bandurska (1991), free proline accumulation in leaves is one of the signs of metabolic response of plants to water stress, and proline accumulation under the same stress conditions differs amongst species and also different varieties of the same species. Proline accumulation is one of the common characteristics in many monocotyledons under water deficit. Proline can perform multiple functions under water stress; for example it regulates the accumulation of useable nitrogen, it can act as an osmotically active substance and improve the hydration of the cytoplasm, as well as serve as a factor stabilizing protein structure and enzyme activity (Bandurska 2000; Bandurska et al. 2008; Javadi et al. 2008). Hanson and Hitz (1982) argued that the accumulation of proline is not an adaptation trait, but more a symptom of stress. According to this reasoning, the results of Rampino et al. (2006) showed that drought-tolerant wheat plants had higher RWC associated with reduced accumulation of proline, which was also visible in our experiments.

Substantial yield losses have been observed in crops due to the reduced water supply, even for a short period of time. A reduction in plant yield or biomass due to water stress has been widely reported (for example: Blum 2005). Prevailing drought reduces plant growth and development, leading to restricted flower production and grain filling, and thus smaller and fewer grains, and this was also found in our study. The descendant line M112 exhibited an increase in most of the parameters tested, but had the lowest yield. This genotype was more sensitive in terms of measured agronomy traits. However, we consider this study as preliminary, because yield components are strongly modified by external conditions, the severity of stress, as well as the developmental phase of the plant. Therefore, it is necessary to conduct further experiments. Since ANOVA analysis for most of the analyzed parameters showed significant differences between tested lines continuation of the study on the whole mapping population of rye seems to be reasonable. Subsequent QTL analysis will allow us to discern the complex nature of the traits studied here, and these analyzes will be the first step in the identification of key regulatory sites for these traits in the rye genome under both optimum watering and drought stress.

Author contribution statement Study conception and design: C-M and M. Analysis and interpretation of data: CM. Drafting of manuscript: C-M and M.

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Compliance with ethical standards

Conflict of interest The authors of the manuscript have no conflict of interest to declare.

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