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ORIGINAL PAPER

The relations between drought susceptibility index based on grain yield (DSI_{GY}) and key physiological seedling traits in maize and triticale genotypes

Maciej T. Grzesiak · Piotr Waligórski · Franciszek Janowiak · Izabela Marcińska · Katarzyna Hura · Piotr Szczyrek · Tomasz Głąb

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Abstract The physiological reasons for the differences in sensitivity of C_3 and C_4 plant species to environmental stresses have not been thoroughly explained. In this study the effects of drought stress on the growth and selected physiological traits were examined in the seedlings of 13 single cross maize (C_4 plant) hybrids and 11 spring triticale (C_3 plant) breeding lines and varieties differing in drought sensitivity. For plants in the seedling stage the results demonstrated a genetic variation in dry matter accumulation of shoots and roots (DW_S, DW_R), number (*N*) and length (*L*) of particular components (seminal, seminal adventitious, nodal) of the root system, membrane injury by soil drought (LI_D), osmotic and high temperature stress (LI_{OS}, LI_{HT}), water potential (ψ), water loss (WL), grain germination in osmotic stress (FG, PI), and seedling survival (SS). Seedlings grown under moderate soil drought showed a decrease in dry matter of the top parts and roots and a decrease in the length of seminal, seminal adventitious and nodal roots in comparison to seedlings grown in control conditions. The observed harmful effects of drought stress were more distinct in drought sensitive genotypes. Used in this paper drought susceptibility indexes (DSI_{GY}) were calculated in other experiment by determining the changes in grain yield (GY) under two soil moisture levels (irrigated and drought). The variation of DSI_{GY} for maize ranges from 0.381 to 0.650 and for triticale from 0.354 to 0.578. The correlations between DSI_{GY} and laboratory tests (LI, FG, SS) confirmed that they are good indicators of drought tolerance in plants. The highest values of genetic variation were observed in LI, DW_S, SS and WL and the lowest in the measurements of ψ FG, PI, L_S, L_{SA} and L_N. The correlation coefficients between LI_{OS} and LI_{HT} tests were, in most of the considered cases, statistically significant, which indicates that in maize and triticale the mechanisms of membrane injury caused by simulated drought or high temperature are physiologically similar. It can be concluded that an approach to the breeding of maize and triticale for drought tolerance using these tests can be implemented on the basis of separate selection for each trait or for all of them simultaneously. In that case, it would be necessary to determine the importance of the trait in relation to growth phase, drought timing and level, as well as its associations with morphological traits contributing to drought tolerance. The obtained values of the correlation coefficient between laboratory tests suggest that the same physiological traits may be applied as selection criteria in drought tolerance of maize and triticale genotypes.

Keywords Triticale-(x *Triticosecale* Witt) · Maize-(*Zea mays* L.) · Soil drought · Osmotic stress · Germination · Membrane stability · Roots · Water potential · Water loss

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M. T. Grzesiak (✉) · P. Waligórski · F. Janowiak · I. Marcińska
The F. Górski Institute of Plant Physiology, Polish Academy of Sciences, Niezapominajek 21, 30-239 Kraków, Poland
e-mail: m.grzesiak@ifr-pan.krakow.pl

K. Hura
Department of Plant Physiology, Agricultural University, Podluzna 3, 30-239 Kraków, Poland

P. Szczyrek
Institute of Biology, Pedagogical University, Podbrzezie 2, 30-054 Kraków, Poland

T. Głąb
Institute of Machinery Exploitation, Agricultural University, Balicka 116 B, 31-149 Kraków, Poland

Abbreviations

DSI_{GY}	Drought susceptibility index
C	Control
D	Drought
SI	Stress index
FWC	Field water capacity
FG	Final germination
PI	Promptness index
SS	Seedling survival
LI	Membrane injury index
Ψ	Leaf water potential
WL	Water loss
DW_R	Root dry matter
DW_S	Shoot dry matter
FW	Leaf fresh weight
DW	Leaf dry matter
L_S, L_{SA}, L_N	Length of seminal, seminal adventitious and nodal roots, respectively
N_N	Number of nodal roots

Introduction

As drought is the most important environmental phenomenon affecting a plant's growth, development and crop yield, considerable progress has been made in understanding the changes in physiological processes caused by drought stress. It has been shown that the physiological responses of plants to drought stress are extremely complex and vary with plant species as well as with the degree and time of the exposure to drought (Levitt 1980; Bennett 1990; Evans et al. 1990, 1991; Jones 1993; Reynolds 2002; King 2011). Plants develop different morphological, physiological and biochemical mechanisms which inhibit or remove the harmful effects of drought stresses (Sullivan and Ross 1979; Boyer 1982; Larsson and Górny 1988; Chaves et al. 2002; Reynolds et al. 1998; Asharaf 2010). Drought tolerance of a plant species is usually determined by the plant's genes and also by morphological, phonological, physiological, and biochemical traits. The responses of plants to drought stress depend on the species, genotype, plant age, level and duration of drought, and physical parameters of the soil. Differences in tolerance to drought are known to exist within genotypes of plant species and were found in many studies, e.g. in maize (Martinielio and Lorenzoni 1985; Lorens et al. 1987; Grzesiak 1990; Grzesiak et al. 2012), wheat (Winter et al. 1988; Reynolds et al. 1998; Paknejad et al. 2007), rape seed (Richards and Thurling 1978), oat (Larsson and Górny 1988), coconut (Gomez et al. 2008) and triticale (Royo et al. 2000; Grzesiak et al. 2012).

Methods of evaluating the degree of drought tolerance allow for a direct or indirect estimation of the various physiological, biochemical or morphological traits of the examined genotypes. Measurements of different physiological processes of plant response to drought provide important information about the reactions of the plant intended to remove or to reduce the harmful effects of water deficit in the soil or plant tissues. Techniques of screening for drought tolerance were devised by selecting genotypes in a field or greenhouse study. Conducting field experiments is necessary for the verification of the drought tolerance estimated on the basis of physiological laboratory tests (Grzesiak 1990; Richards 1991; Kpoghomou et al. 1990). For proper field testing a number of methodological problems must be solved to enable water content in the soil to be controlled by irrigation or by limiting the inflow of water from rainfall. The relations between the plant yield obtained under conditions of drought and that obtained under conditions of optimal soil moistening were preferred among the field indices of drought tolerance. Such tests, however, are not accurate enough or too simplified to show important relations between the crop forming processes and soil–water-plant relationship. A more precise quantitative formulation of this relationship can be found in the studies by Fischer and Maurer (1978), Hanson and Nelson (1985), Winter et al. (1988), Stanley (1990) and in FAO reports by Doorenbos and Pruit (1977), Doorenbos and Kassam (1986).

Methods of screening for drought tolerance within a large number of genotypes should be easy, rapid and inexpensive (Hanson and Nelson 1985; Palta 1990; Zagdańska 1992). It is also necessary for the laboratory testing method to be characterized by a significant correlation with drought resistance observed under field conditions (Sullivan and Ross 1979; Blum et al. 1980; Bouslama and Schapauch 1984; Hanson and Nelson 1985; Kpoghomou et al. 1990; Chaves et al. 2002; Grzesiak et al. 2012). The most important laboratory methods suggested for screening for drought tolerance in crop plants were: germination in osmotic substances (mannitol, PEG), growth or survival of young seedlings subjected to soil or simulated water stress and high temperature stress (Sullivan and Ross 1979; Blum and Ebercon 1981; Martinielio and Lorenzoni 1985), leaf injury, leaf water content, leaf temperature and parameters of leaf gaseous exchange (Passioura et al. 1993; Farquhar et al. 1993; Dubey 1997). Other traits that may be promising as screening traits are different parameters of chlorophyll fluorescence (Reynolds 2002; Hura et al. 2007) and associations between dark respiration under drought conditions and heat tolerance of sorghum lines (Gerik and Eastin 1985) and of wheat (Reynolds et al. 1998).

The physiological reasons for the differences in sensitivity of C_3 and C_4 plant species to environmental stresses have not been thoroughly explained and understood (Edwards and Ku 1987; Medrano et al. 2002; Nayyar and

Gupta 2006; Lopes et al. 2011). Maize and triticale have different types of photosynthesis (C_3—in triticale, C_4—in maize), different metabolic pathways and structure of bundle sheath chloroplasts (Kranz syndrome). According to Iijima and Kono (1991), cereal species develop two types of root system, depending on the angle of the growth of branches (lateral roots) and their distribution in the soil profile. For triticale, the root system structure is "concentrated" whereas it is "scattered" for maize. Maize and spring triticale appear to be sensitive to drought stress during grain germination, seedling emergence, early vegetative growth and pollination. Moreover, both maize and triticale are important crops widely cultivated throughout the world (Fageria et al. 2006).

The objectives of this study were to evaluate the variation of selected physiological characteristics of maize and triticale seedlings grown in drought conditions and to compare them to variations in drought susceptibility index based on grain yield (DSI_{GY}) of plants grown in a stressed environment under field conditions and then determine which tests are most useful for screening drought resistance genotypes.

Materials and methods

Plant materials

Experiments were conducted on 13 single-cross maize hybrids and on 4 strains and 7 cultivars of spring triticale. Maize grains were obtained from Pioneer Overseas GmbH (Austria), Pioneer Saaten (Poland), Garst Seed Company (USA), Agriculture Canada and SAMPLO Holding (Slovakia). Triticale grains of breeding lines were obtained from DANKO in Choryn and cultivars from IHAR, Małyszyn (Poland). The choice of the maize hybrids and triticale lines and cultivars to be examined was done on the basis of the information on the effect of drought on plant yield received from breeders. Plant materials and drought susceptibility indexes (DSI_{GY}) according to Grzesiak et al. (2012) are listed in Fig. 1.

Experimental conditions

The experiment was carried out in a greenhouse under the conditions of 25/20 °C day/night temperature and 65 % relative humidity. Plants were grown in Mitscherlich pots and root-boxes filled with a mixture of garden soil, peat and sand (1:1:1). Air-dried soil substrate was sieved in a 0.25 cm mesh. Soil substrate pH was 7.1 and the percent of organic material was 0.7 %. For the determination of root length and number of root system components, seedlings were grown in root-boxes, which enabled non-destructive isolation of all compartments of the root system (Kono et al. 1987). A set for the "root-box and pin board method" consists of: a Plexiglas box (width—0.25 m, depth—0.40 m, thickness—0.02 m), a pin board for sampling the root system, and a polyethylene sheet (envelope) for handling and preserving the root system. In root-boxes soil compaction level was 1.30 g cm^{-3}. Mechanical impedance in soil substrate was measured with penetrometer DIK-5520 (Daiki Rika Kogyo Co. Ltd, Japan).

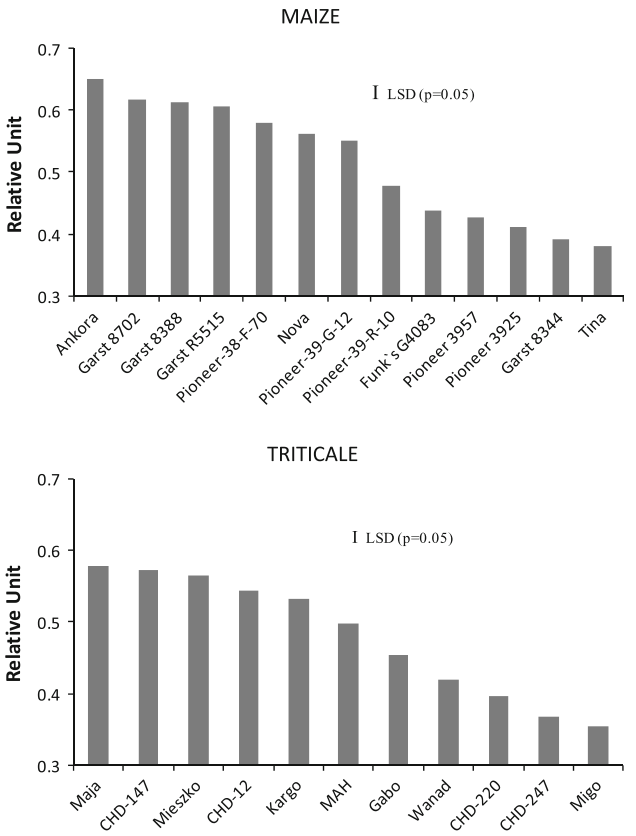

Fig. 1 Maize and triticale genotypes ordered according to their Drought Susceptibility Index (DSI_{GY})

Field soil water capacity (FWC) for soil mixture was determined according to Kopecky methods. Air-dried soil samples were placed inside metal cylinders, with a 1 mm hole at the bottom. The volume for the samples was 100 cm^3. Cylinders with the samples were placed inside a container with water for 30 min. After 8 h, maximal soil water content in the samples was 0.43 (g cm^{-3}) and after 48 h it decreased to 0.21 (g cm^{-3}). Following Hillel and van Bavel (1976), those last values were assumed to be 100 % of soil field water capacity (FWC). The pots and root-boxes were weighed every day, and the amount of water loss through transpiration was added to maintain the original weight in each treatment. For control (C) treatment soil water content was maintained from sowing for 42 days at the level of 65–70 % FWC. Soil water content in the root-box experiment for drought (*D*) treatment was kept at the level of 30–35 % FWC from

the 21st to 42nd day. Similarly, in the pot experiment from the 21st to 42nd day it was kept at the level of 30–35 % FWC for drought treatment D_{35} and at the level 15–20 % FWC for treatment D_{20}. In order to limit water evaporation from pots and root-boxes, soil surface was covered with 1 cm layer of ground Styrofoam. A single pregerminated grain was planted at the depth of 3–4 cm. After 42 days of plants' growth under C treatment Zadoks scale was about 17 or 18 for maize and about 16 or 17 for triticale.

On the 42th day seedlings grown in pots were used for measurements of leaf water potential, membrane injury and water loss in the excised leaf. For each species (2), genotypes (13 or 11) and treatments (3) 5 pots with 3 plants were used ($n = 5$). Similarly, seedlings grown in root-boxes were cut into top parts and roots for the determination of dry weigh (DW_S, DW_R) and number and length of particular components of the root system. The roots were sampled after the soil from the pot had been washed away by a gentle stream of water. After the measurements root samples were preserved in a FAA (formalin, acetic acid, and ethanol) solution. For each species (2), genotypes (13 or 11) and treatments (2) 4 root-boxes with 1 plants were used ($n = 4$).

Measurements

Germination and seedling survival (FG, PI, SS). Twenty grains of each genotype were germinated under 0.00, −0.47, −0.85 and −1.30 MPa of osmotic stress. Grains of the tested genotypes were surface sterilized in 70 % ethanol for 5 min and placed on petri dishes and incubated in an air-conditioned growth cabinet at the temperature of 25 °C. Osmotic stress was simulated using mannitol solutions ($C_6H_{14}0_6$, M.W. 182.17), Lobe Chemia. The concentration of solutions at the desired chemical water potential (ψ) was calculated according to Michel et al. (1983), the control was distilled water ($\psi = 0.0$ MPa). Germination was recorded when the radicle reached 5 mm in length. There were 4 replications for each treatments. Counts of germinated grains were made each day to compute the final germination percentage (FG) and promptness index (PI) and calculated as follows:

$$FG = (nd_{14} \cdot 100) \cdot 20^{-1}$$

$$PI = (nd_2 \cdot 1.00) + (nd_4 \cdot 0.75) + (nd_6 \cdot 0.50) + (nd_8 \cdot 0.25)$$

where nd_x = number of germinated grains by the xth day, Seedling survival (SS), 25 germinated grains were planted in 5 pots and after 14 days the number of growing seedling was recorded, and seedling survival index (SS) was calculated as follows:

$$SS = \text{number of living seedlings} \cdot 25^{-1}$$

Leaf water potential (ψ) was measured with thermocouple psychrometer HR 33T (Wescor Inc., Logan, USA) in "dew point" mode, equipped with sample chamber C-52 SF and digital multimeter *Metex M-3640 D*. Measurements were taken on leaf discs—diameter of 0.3 cm for triticale and 0.5 cm for maize—and immediately placed inside the psychrometer chamber and left to balance temperature and water vapor equilibrium for 30 min before measurements. For each treatments there were 5 replications. Psychrometric readout and ψ determination were made as described by Johnson and Brown (1977).

Excised-leaves water loss (WL) was measured only for control treatment (C) of each maize and triticale genotype. Leaves were cut from the upper half of the plant, weighed (FW_0) and left to desiccate at 25 °C in the dark. After 12 and 24 h samples were reweighed (FW_{12}, FW_{24}) and next oven dried at 70 °C and weighed again for the determination of dry weight of samples (DW). There were 5 replications. Water loss of excised leaves (WL) was calculated by the following formula:

$$WL = (FW_0 - FW_{12} \text{ or } FW_{24})DW^{-1}$$

Dry weight of shoots (DW_S) *and roots* (DW_R) was sampled in each root-box and was determined on the 42nd day after sowing through drying at 65 °C for 72 h.

Number (*N*) *and length* (*L*) *of seedling root components* (seminal-L_S, seminal adventitious-L_{SA}, nodal-$N_N{\cdot}L_N$) was measured with DELTA-T SCAN (England) analyzer.

Relative loss of intracellular electrolytes from leaf tissues (LI) was measured with the conductivity method using conductivity meter OK-102/1 (Radelkis, Hungary), according to the procedure and formula described by Sullivan and Ross (1979) and Blum et al. (1980).

$$LI = 1 - \left[1 - (T_I - T_2)^{-1}\right]\left[1 - (C_1.C_2)^{-1}\right]^{-1} 100$$

where C and T refer to the conductivity of control and treatment solutions, respectively, and subscript $_1$ and $_2$ refer to initial and final conductance, respectively.

Nine leaf discs (0.5 cm diameter for maize and 0.3 cm for triticale) were cut from leaves and immersed in test tubes containing 30 cm^3 redistilled water. After 24 h initial conductance measurements were taken. Final conductance measurements were taken after autoclaving all tubes at 110 °C for 15 min and cooling them to room temperature.

In the experiment additional conductivity tests of leaf injury by simulated drought (LI_{OS}) and by high temperature (LI_{HT}) were made. For these tests leaf discs were cut only from control (C) plants. For LI_{OS} leaf discs were immersed for 12 h in test tubes with 30 cm^3 of redistilled water (control 0.0 MPa) or 30 cm^3 of mannitol solution (treatment −0.47, −0.85 and −1.30 MPa). High

temperature stress (LI_{HT}) was imposed by immersing leaf discs for 1 h in test tube with 30 cm^3 of redistilled water by placing them in well-stirred water bath at three temperatures: 25 °C (control), 35 and 45 °C (treatments). There were 5 replications for each treatments.

All measurements except germination tests (FG, PI, SS) were made after 42 days of seedlings' growth. Samples for measurements of leaf water potential (ψ), relative loss of intracellular electrolytes (LI), and water loss of excised leaves (WL) were taken between 11:00 and 13:00 on most recent fully expanded leaf. Results of all measurements are presented as stress index (SI) which was calculated as indicated below:

$$SI = \text{treatment value} \cdot \text{control value}^{-1}.$$

Statistical analysis

All data were analyzed by one-way ANOVA and LSD tests ($p < 0.05$) using Statistica 10.0 (StatSoft, Inc., USA). Linear correlation analyses were used to determine the relationship between drought susceptibility index (DSI_{GY}) and stress indices (SI) obtained in laboratory tests. Also correlation coefficients were calculated for the determination of the relationship between and within laboratory tests.

Results and discussion

Drought susceptibility indexes (DSI_{GY}) of maize and triticale genotypes

According to our earlier research (Grzesiak et al. 2012), drought susceptibility indexes (DSI_{GY}) for maize and triticale genotypes were calculated by determining the changes in grain yield (GY) under two soil moisture levels (irrigated and drought). Variation of DSI_{GY} for maize ranges from 0.381 to 0.650 and for triticale from 0.354 to 0.578. The values of DSI_{GY} made it possible to rank the examined maize and triticale genotypes according to their drought tolerance. In the maize hybrids the drought resistant group ($0.381 < DSI_{GY} > 0.439$) comprises the hybrids Tina, Garst 8344, Pioneer 3925 and Pioneer 3957, while the drought sensitive group ($0.607 < DSI_{GY} > 0.650$) comprises the hybrids Ankora, Garst 8702, Garst 8388 and Garst R5515. For triticale the drought resistant group ($0.354 < DSI_{GY} > 0.419$) comprises the lines CHD 247 and CHD 220, and cultivars Migo and Wanad, while the drought sensitive group ($0.544 < DSI_{GY} > 0.578$) comprises the strains CHD 147 and CHD 12, and cultivars Mieszko and Maja (Fig. 1).

Germination under simulated drought conditions (FG, PI) and seedling survival (SS)

In the control treatment (0.0 MPa), maize and triticale genotypes did not differ in the final germination index (FG) and the correlation coefficients between FG and DSI_{GY} were insignificant. The imposed drought from −0.43 to 1.30 MPa caused a decrease in the values of stress indexes of FG and the correlation coefficients between FG and DSI_{GY} were statistically significant for maize in −0.43 MPa treatment and for triticale in −0.85 and −1.30 MPa treatments (Table 1). Differences between maize and triticale genotypes in promptness index (PI) values in the control were statistically significant only for maize, whilst the correlation coefficient with DSI_{GY} was statistically significant for both species. As with drought FG, osmotic stress caused a decrease in the values of PI and the correlation coefficients between PI and DSI_{GY} were statistically significant for maize in −0.43 and −0.85 MPa treatments, but for triticale only in −1.30 MPa treatment (Table 2). The results of seedling survival (SS) after grain germination in different water potential of mannitol solutions are presented in Table 3. Germination under osmotic stress conditions (−0.43, −0.85. −1.30 MPa) caused a decrease in the number of live seedlings in maize to 86, 79 and 76 %, respectively, and for triticale to 97, 86 and 84 %, respectively. Correlation coefficients between DSI_{GY} and the values of stress index of seedling survival were statistically significant in −0.43, −0.85 and −1.30 MPa treatments of maize genotypes, but for triticale only −0.85 and −1.30 MPa treatments were affected.

Germination is strongly influenced by plant species, grain age and storage conditions and is also highly sensitive to soil water quality and temperature (Ashraf and Mehmood 1990; Ahmad et al. 2009). Grain germination indexes were used with different results in selecting procedures for identifying drought resistant genotypes (Jajarmini 2009). Opinions on the efficiency of plant drought tolerance assessments on the basis of different parameters of germination are divergent and indicate their limited usefulness. It is believed that germination traits are affected by drought stress less than other physiological and biochemical traits (Winter et al.1988; Morgan 1992). Genotype tolerance to drought stress is process-specific with regard to grain water imbibition, endosperm utilization, activation of the dormant enzyme system and seedling emergence, growth and survival after stress. The tolerance to drought in any of those stages will affect the results and thus screening for drought tolerance reaction should also be process-specific, and the prediction of genotype performance from one process to another would not necessarily be possible (Blum et al. 1980; Ahmad et al. 2009). In this study the tolerance to simulated drought stress proved a relatively good indicator in the screening of maize

Table 1 Final germination index (FG) of maize and triticale genotypes after 14 days germination in mannitol solutions differing in water potential

Genotype	Control (C)	Stress index (SI) of osmotic stress treatments		
	0.0 MPa (%)	−0.43 MPa	−0.85 MPa	−1.30 MPa
Maize				
Ankora	98.5	0.818	0.698	0.627
Garst 8702	100.0	0.862	0.632	0.480
Garst 8388	98.1	0.826	0.654	0.502
Garst R5515	99.2	0.808	0.701	0.557
Pioneer-38-F-70	97.5	0.852	0.739	0.668
Nova	99.2	0.863	0.757	0.686
Pioneer-39-G-12	99.5	0.847	0.702	0.631
Pioneer-39-R-10	98.9	0.860	0.691	0.620
Funk's G4083	99.5	0.876	0.726	0.655
Pioneer 3957	98.9	0.841	0.740	0.669
Pioneer 3925	100.0	0.890	0.721	0.651
Garst 8344	98.0	0.879	0.668	0.597
Tina	100.0	0.872	0.777	0.652
Mean	99.0	0.854	0.708	0.615
Range	97.5–100.0	0.808–0.890	0.632–0.777	0.488–0.686
$LSD_{0,05}$	1.8	0.025	0.028	0.067
CV	0.8	2.9	5.8	10.5
r versus DSI_{GY}	-0.259^{NS}	−0.728**	-0.419^{NS}	-0.473^{NS}
Triticale				
Maja	100.0	0.989	0.809	0.679
CHD-147	100.0	0.956	0.713	0.583
Mieszko	98.5	0.984	0.801	0.620
CHD-12	99.9	0.943	0.723	0.593
Kargo	98.7	0.984	0.801	0.619
MAH	98.5	0.996	0.813	0.681
Gabo	99.2	0.990	0.808	0.677
Wanad	99.6	0.985	0.804	0.744
Migo	99.0	0.985	0.803	0.672
CHD-220	100.0	0.989	0.842	0.712
CHD-247	98.5	0.995	0.863	0.713
Mean	99.3	0.981	0.798	0.663
Range	98.5–100.0	0.943–0.996	0.713–0.863	0.583–0.744
$LSD_{0,05}$	1.31	0.016	0.047	0.077
CV	0.7	1.7	5.5	7.9
r versus DSI_{GY}	0.203^{NS}	-0.463^{NS}	−0.616*	−0.719*

Results for osmotic stress treatments are shown as a stress index (SI). Mean values, range of means, coefficient of variation (CV) and correlation coefficient (*r*) between measured traits and drought susceptibility index (DSI_{GY}). Genotypes are ordered according to the DSI_{GY} value ($n = 4$)

Number of degree of freedom (df) for critical values of (*r*) was for maize 11 and for triticale 9

NS, *r*-Correlation coefficient not significant

*, ** *r*-Correlation coefficient significant at the 0.05 or 0.01 level, respectively

and triticale genotypes. Our results showed that the efficiency of FG, PI and SS is related to how strongly a trait is expressed and the measurement must be performed at the right moment in order to maximize the expression of genetic variations of the particular trait.

Seedling dry matter (DW_S, DW_R), root length (L_S, L_{SA}, L_N) and the number of nodal roots (N_N)

Soil drought decreased the dry matter of the above ground parts (DW_S) in 13 genotypes of maize from 12.0 to 67.0 % and in 11 genotypes of triticale from 5.0 to 17.0 %. Similarly, the dry matter of roots (DW_R) decreased in maize genotypes from 3.0 to 30.0 % and in triticale genotypes from 1.0 to 25.0 %. Drought also strongly influenced the ratio of DW_S to DW_R within maize genotypes from 0.0 to 60.0 % and in triticale genotypes from 0.0 to 25.0 % (Table 4). The variation coefficients (CV) in control (C) seedlings of DW_S, DW_R and the ratio of DW_S to DW_R in maize were about 29, 18 and 37 %, respectively, and in triticale 23, 29 and 22 %, respectively. CV calculated for treatment D as a stress index (SI) for those traits in maize

Table 2 Promptness index (PI) of maize and triticale genotypes after 8 days germination in mannitol solutions differing in water potential

Genotype	Control (C)	Stress index (SI) of osmotic stress treatments		
	0.0 MPa (%)	−0.43 MPa	−0.85 MPa	−1.30 MPa
Maize				
Ankora	17.0	0.682	0.659	0.635
Garst 8702	16.4	0.744	0.659	0.634
Garst 8388	17.1	0.713	0.614	0.608
Garst R5515	17.6	0.722	0.642	0.619
Pioneer-38-F-70	18.0	0.783	0.611	0.572
Nova	16.9	0.769	0.686	0.604
Pioneer-39-G-12	18.1	0.823	0.707	0.575
Pioneer-39-R-10	17.6	0.778	0.699	0.619
Funk's G4083	17.9	0.726	0.704	0.626
Pioneer 3957	17.9	0.810	0.670	0.570
Pioneer 3925	18.1	0.818	0.652	0.575
Garst 8344	17.8	0.781	0.702	0.624
Tina	17.8	0.781	0.702	0.624
Mean	17.6	0.764	0.670	0.606
Range	16.0–17.9	0.682–0.823	0.611–0.707	0.570–0.635
$LSD_{0,05}$	1.11	0.130	0.078	0.020
CV	3.0	5.7	5.0	4.1
r versus DSI_{GY}	−0.632*	−0.592*	−0.574*	0.163^{NS}
Triticale				
Maja	14.8	0.946	0.905	0.757
CHD-147	17.8	0.837	0.618	0.506
Mieszko	15.0	0.933	0.847	0.627
CHD-12	18.1	0.773	0.718	0.497
Kargo	16.1	0.981	0.832	0.714
MAH	14.9	0.980	0.866	0.725
Gabo	15.9	0.987	0.818	0.711
Wanad	19.0	0.80	0.711	0.605
Migo	16.0	0.931	0.831	0.781
CHD-220	15.9	0.994	0.937	0.755
CHD-247	16.5	0.958	0.909	0.788
Mean	16.3	0.920	0.817	0.679
Range	14.8–19.0	0.773–0.987	0.618–0.937	0.497–0.788
$LSD_{0.05}$	1.80	0.122	0.131	0.107
CV	8.4	8.6	11.9	15.4
r versus DSI_{GY}	-0.121^{NS}	-0.229^{NS}	-0.333^{NS}	-0.557^{NS}

Results for osmotic stress treatments are shown as a stress index (SI). Mean values, range of means, coefficient of variation (CV) and correlation coefficient (*r*) among measured traits and drought susceptibility index (DSI_{GY}). Genotypes are ordered according to the DSI_{GY} value ($n = 4$)

Number of degree of freedom (df) for critical values of (*r*) was for maize 11 and for triticale 9

NS, *r*-Correlation coefficient not significant

* *r*-Correlation coefficient significant at the 0.05 level

were about 34, 13 and 31 %, respectively, and in triticale 15, 9 and 11 %, respectively (Table 4). In the control treatment (C), the statistically significant correlation between DSI_{GY} and DW_S, DW_R and the ratio of DW_S to DW_R were found only in triticale for the ratio of DW_S to DW_R. In the drought treatment, the statistically significant correlation between DSI_{GY} and DW_S, DW_R and the ratio of DW_S to DW_R were found in maize for DW_S and the ratio of DW_S to DW_R, and in triticale, only for DW_S (Table 4).

The results presented in Table 5 show that in the control (C) treatment, differences within drought resistant and drought sensitive genotypes of maize and triticale in the length of seminal (L_S), seminal adventitious (L_{SA}), nodal roots (L_N), and number of nodal roots (N_N) were in most cases statistically insignificant. Also under control conditions the correlation coefficient between DSI_{GY} and the measured traits was statistically insignificant. L_S in drought treatment decreased in the drought resistant genotypes of maize (Tina, Garst 8344) about 10 % and in drought sensitive genotypes (Ancora, Garst 8702) about 23 %. In drought resistant genotypes of triticale (CHD-220, CHD-247) the decrease was about 6 % and in drought sensitive

Table 3 Seedlings survival (SS) of maize and triticale genotypes germinated in mannitol solutions differing in water potential and after 14 days of grown in well watered soil

Genotype	Control (C)	Stress index (SI) of osmotic stress treatments		
	0.0 MPa (%)	−0.43 MPa	−0.85 MPa	−1.30 MPa
Maize				
Ankora	25.0	0.80	0.72	0.52
Garst 8702	25.0	0.72	0.64	0.72
Garst 8388	25.0	0.96	0.80	0.84
Garst R5515	25.0	0.72	0.68	0.52
Pioneer-38-F-70	25.0	0.80	0.76	0.76
Nova	25.0	0.92	0.84	0.92
Pioneer-39-G-12	25.0	0.72	0.64	0.72
Pioneer-39-R-10	25.0	0.84	0.80	0.72
Funk's G4083	25.0	0.88	0.80	0.84
Pioneer 3957	25.0	0.96	0.92	0.72
Pioneer 3925	25.0	0.92	0.88	0.72
Garst 8344	25.0	0.92	0.84	0.88
Tina	25.0	1.00	0.96	1.00
Mean	25.0	0.86	0.79	0.76
Range		0.72–1.00	0.64–0.96	0.52–1.00
$LSD_{0.05}$		0.088	0.107	0.099
CV		11.51	12.76	18.36
r versus DSI_{GY}		−0.638*	−0.763**	-0.544^{NS}
Triticale				
CHD-147	25.0	0.96	0.68	0.64
Maja	25.0	1.00	0.96	0.92
CHD-12	25.0	0.96	0.72	0.64
Mieszko	25.0	1.00	0.88	0.88
Kargo	25.0	0.96	0.8	0.76
MAH	25.0	0.92	0.84	0.80
Gabo	25.0	0.96	0.84	0.80
Wanad	25.0	1.00	0.96	0.96
Migo	25.0	0.96	0.96	0.96
CHD-220	25.0	1.00	0.96	0.96
CHD-247	25.0	1.00	0.92	0.92
Mean	25.0	0.97	0.86	0.84
Range		0.92–1.00	0.72–0.96	0.64–0.96
$LSD_{0.05}$		0.067	0.049	0.039
CV		2.77	11.56	14.44
r versus DSI_{GY}		-0.161^{NS}	−0.827**	−0.634*

Results for osmotic stress treatments are shown as a stress index (SI). Mean values, range of means, coefficient of variation (CV) and correlation coefficient (*r*) between measured traits and drought susceptibility index (DSI_{GY}). Genotypes are ordered according to the DSI_{GY} value ($n = 5$)

Number of degree of freedom (df) for critical values of (*r*) was for maize 11 and for triticale 9

NS, *r*-Correlation coefficient not significant

*, ** *r*-Correlation coefficient significant at the 0.05 and 0.01 level, respectively

genotypes (CHD-12, CHD-147) about 13 %. In maize the decrease in the total length of two seminal adventitious roots (L_{SA}) in comparison to the control was about 7 % in drought resistant hybrids and in drought sensitive about 25 %. For triticale the decrease in the total length of three seminal adventitious (L_{SA}) roots in comparison to the control was statistically insignificant in drought resistant and sensitive lines. In seedlings exposed to drought, significant differences between drought resistant and sensitive genotypes were observed in the measurements of the total length of nodal roots (L_N). In maize the decrease in L_N in drought sensitive genotypes was about 22 % and in the drought resistant ones about 10 %, and in triticale about 30 and 13 %, respectively. In C treatments no statistically significant correlation between DSI_{GY} and L_S, L_{SA}, N_N and L_N was found in either maize or triticale genotypes. In seedlings exposed to drought, a statistically significant correlation between DSI_{GY} and SI calculated for the measurements of root traits was found in maize, but in triticale only for L_N (Table 5).

As one might expect, a root system characteristic, such as the number and length of particular components of root

Table 4 Effect of moderate soil drought (35 % FWC) on dry matter of shoots (DW_S)

Genotype	Shoots (S)		Roots (R)		Ratio of S to R	
	C	D_{35}	C	D_{35}	C	D_{35}
Maize						
Ankora	7.08	0.412	3.29	0.761	2.15	0.542
Garst 8702	9.39	0.339	3.21	0.779	2.92	0.435
Garst 8388	10.41	0.410	2.94	0.850	3.54	0.482
Garst R5515	8.18	0.463	3.17	0.583	2.58	0.795
Pioneer-38-F-70	6.07	0.450	3.32	0.901	1.83	0.499
Nova	6.11	0.750	5.50	0.891	1.11	0.842
Pioneer-39-G-12	9.13	0.329	3.44	0.807	2.65	0.407
Pioneer-39-R-10	11.18	0.359	3.45	0.704	3.24	0.510
Funk's G4083	6.13	0.713	4.00	0.768	1.53	0.929
Pioneer 3957	8.11	0.755	3.65	0.840	2.22	0.898
Pioneer 3925	8.31	0.721	3.45	0.722	2.41	0.998
Garst 8344	4.50	0.644	4.00	0.950	1.13	0.678
Tina	4.00	0.875	3.50	0.971	1.14	0.901
Mean	7.58	0.555	3.61	0.810	2.19	0.686
Range	4.00–11.18	0.329–0.875	2.94–5.50	0.704–0.971	1.11–3.54	0.407–0.998
$LSD_{0.05}$	0.39	0.111	0.18	0.077	0.65	0.208
CV	28.7	34.4	17.81	13.3	36.91	31.3
r versus DSI_{GY}	0.413^{NS}	−0.732**	-0.228^{NS}	-0.337^{NS}	0.464^{NS}	−0.642*
Triticale						
Maja	3.99	0.779	3.59	0.864	1.11	0.903
CHD-147	3.06	0.654	1.68	0.832	1.82	0.786
Mieszko	4.02	0.746	2.41	0.974	1.67	0.766
CHD-12	3.45	0.725	1.90	0.791	1.82	0.917
Kargo	4.18	0.749	3.34	0.897	1.25	0.835
MAH	5.89	0.540	4.71	0.744	1.25	0.726
Gabo	4.11	0.852	3.50	0.857	1.17	0.994
Wanad	3.08	0.860	2.50	0.880	1.23	0.978
Migo	3.02	0.841	2.58	0.992	1.17	0.848
CHD-220	3.08	0.945	3.15	0.947	0.98	0.998
CHD-247	3.15	0.911	2.72	0.921	1.16	0.990
Mean	3.73	0.782	2.92	0.882	1.33	0.885
Range	3.02–5.89	0.725–0.945	1.68–4.71	0.744–0.992	0.98–1.82	0.726–0.998
$LSD_{0.05}$	0.25	0.139	0.11	0.223	0.38	0.141
CV	23.0	15.0	29.4	8.6	22.1	11.2
r versus DSI_{GY}	0.370^{NS}	−0.670*	-0.064^{NS}	-0.451^{NS}	0.627*	-0.554^{NS}

Roots (DW_R) and ratio of DW_S to DW_R of maize and triticale seedlings after 42 days of growth. Results for drought treatments (D_{35}) are shown as a stress index (SI). Mean values, range of means, coefficient of variation (CV) and correlation coefficient (*r*) between measured traits and drought susceptibility index (DSI_{GY}). Genotypes are ordered according to the DSI_{GY} value ($n = 4$)

Number of degree of freedom (df) for critical values of (*r*) was for maize 11 and for triticale 9

NS, *r*-correlation coefficient not significant

*, ** *r*-Correlation coefficient significant at the 0.05 and 0.01 level, respectively

system structure, their depth and abundance, are known to be associated with performance under drought conditions in many studies of cereal species (Richards 1996; Reynolds 2002). Nevertheless, decreased allocation in roots in the top layer of the soil has been shown to be an effective drought stress adaptive mechanism (Richards 1991). In the studies by Kono et al. (1987) the specific response of cereal species to drought stress was clearly noticeable in root

Table 5 Effects of moderate soil drought (35 % FWC) on length and number of particular components of root system in maize and triticale seedlings after 42 days of growth in root-boxes

Genotype	Length of seminal root—L_S (cm)		Length of seminal adventitious roots—L_{SA} (cm)		Number of nodal roots—N_N		Total length of nodal roots—L_N (cm)	
	C	D_{35}	C	D_{35}	C	D_{35}	C	D_{35}
Maize								
Ankora	37.7	0.775	68.1	0.730	11.5	0.974	345.9	0.740
Garst 8702	36.4	0.769	71.1	0.755	12.5	0.968	333.2	0.739
Garst 8388	35.2	0.793	67.1	0.757	11.8	1.017	361.0	0.740
Garst R5515	35.4	0.850	70.6	0.776	12.9	1.008	374.1	0.738
Pioneer-38-F-70	36.4	0.810	65.4	0.751	10.7	1.047	300.2	0.741
Nova	37.1	0.803	78.1	0.939	11.5	1.043	355.5	0.889
Pioneer-39-G-12	37.2	0.892	68.4	0.939	11.0	0.982	321.1	0.739
Pioneer-39-R-10	38.2	0.809	65.0	0.749	11.3	1.044	325.4	0.738
Funk's G4083	35.3	0.918	72.5	1.014	13.7	1.058	407.8	0.887
Pioneer 3957	31.0	0.926	69.0	0.930	11.2	1.232	324.5	0.900
Pioneer 3925	35.0	0.903	70.2	1.014	13.0	1.092	377.7	0.890
Garst 8344	39.4	0.916	64.2	0.922	11.7	0.983	340.2	0.890
Tina	34.2	0.907	75.0	0.949	13.5	1.015	411.7	0.887
Mean	36.0	0.852	69.6	0.863	12.0	1.036	352.2	0.809
Range	31.0–39.4	0.769–0.926	64.2–78.1	0.730–1.014	10.7–13.7	0.968–1.232	300.2–411.7	0.738–0.900
$LSD_{0.05}$	3.49	0.039	3.91	0.074	1.18	0.113	39.7	0.013
CV	5.8	7.1	5.7	12.8	8.2	6.7	9.5	9.7
r versus DSI_{GY}	-0.144^{NS}	−0.838**	-0.050^{NS}	−0.746**	-0.347^{NS}	-0.447^{NS}	-0.363^{NS}	−0.798**
Triticale								
Maja	34.1	0.818	90	0.903	15.4	0.773	199.3	0.688
CHD-147	31.5	0.867	89.5	0.911	16.4	0.930	213.5	0.797
Mieszko	28.7	0.951	95.4	0.820	17.9	0.765	232.4	0.612
CHD-12	32.3	0.870	100.2	0.831	18.2	0.813	200.3	0.741
Kargo	28.4	0.891	98.6	0.722	14.1	0.816	222.2	0.653
MAH	30.5	0.866	88.2	0.783	15.0	0.820	194.9	0.796
Gabo	29.5	0.953	107.3	0.702	14.2	0.894	175.6	0.74
Wanad	30	0.900	108.7	0.786	19.9	0.905	298.3	0.889
Migo	33.3	0.937	95.1	0.918	17.2	0.884	257.0	0.817
CHD-220	30.5	0.938	95.4	0.894	17.2	0.797	228.6	0.854
CHD-247	34.2	0.939	90.2	0.945	16.2	0.938	211.3	0.953
Mean	31.2	0.903	96.2	0.838	16.5	0.849	221.2	0.776
Range	28.4–34.2	0.818–0.953	88.2–108.7	0.702–0.945	14.1–19.9	0.765–0.938	175.6–298.3	0.612–0.953
$LSD_{0.05}$	2.18	0.027	5.39	0.083	2.11	0.101	13.0	0.088
CV	6.6	4.9	7.3	9.9	10.8	7.4	15.2	13.2
r versus DSI_{GY}	-0.234^{NS}	-0.547^{NS}	-0.210^{NS}	-0.222^{NS}	-0.198^{NS}	-0.489^{NS}	-0.392^{NS}	−0.760**

Results of drought treatment (D_{35}) are shown as stress index (SI). Mean values, range of means, coefficient of variation (CV) and correlation coefficient (*r*) between measured traits and drought susceptibility index (DSI_{GY}). Genotypes are ordered according to the DSI_{GY} value ($n = 4$)

Number of degree of freedom (df) for critical values of (*r*) was for maize 11 and for triticale 9

NS, *r*-Correlation coefficient not significant

*, ** *r*-Correlation coefficient significant at the 0.05 and 0.01 level, respectively

distribution, nodal root number, leaf number and grain yield. Those authors reported that the responses in root growth for maize and rice were different in the downward penetration of the main axis and in the higher order laterals. Species with a "concentrated" type of root system showed less restriction of root and shoot growth compared to species with a "scattered" type. A decrease in size of the root system and an increasing irregularity of root

distribution resulted in water and nutrients being transported greater distances to the nearest roots (Tardieu 1991; Lipiec et al. 1996). Drought also modifies root system components, such as the main root axis and lateral roots of different orders in rice and maize (Iijima and Kono 1991). Changes in the morphological structure of the root system in triticale and maize were also observed in our earlier studies in response to waterlogging and soil compaction (Grzesiak et al. 2012).

Leaf water potential (ψ) and water loss of excised leaves (WL)

Moderate (D_{35}) and severe (D_{20}) soil drought decreased ψ in maize and triticale (Table 6). Differences between resistant genotypes of maize (Garst 8344, Tina) and triticale (CHD-247, CHD-220) in terms of a decrease of ψ were statistically significant in comparison with sensitive genotypes of maize (Ankora, Garst 8702, Garst 8388) and triticale (Maja, CHD-247).

In seedlings exposed to drought, a statistically significant correlation between DSI_{GY} and SI calculated for the measurement of ψ in maize was found for both treatments but in triticale only for treatment D_{20}. The differences in WL between drought resistant and sensitive genotypes of maize and triticale genotypes were statistically significant. A statistically significant correlation between DSI_{GY} and WL in maize was found only in the measurements taken after 12 h of leaf desiccation but in triticale in the measurements taken after 12 and 24 h (Table 6).

Decreases in leaf water potential initially induced stomatal closure, resulting in a decrease in the supply of CO_2 to the mesophyll cells and subsequently in a decrease in the rate of leaf photosynthesis (Williams et al. 1999; Lawlor and Cornic 2002). According to Hura et al. (2007), a statistically significant correlation between water potential and photosynthetic rate and stomatal conductance was found in maize and triticale during various stages of plant development. Dehydration in C_3 and C_4 plants impairs various physiological processes, especially the changes in leaf water content, water potential and photosynthesis. There is a controversy as to whether drought limits photosynthesis due to leaf water status, stomatal closure, metabolic impairment or injuries to photosynthetic apparatus (Flexas et al. 2006). The first response to leaf water deficit is stomata closure, which limits CO_2 diffusion to chloroplasts (Berkowitz et al. 1983; Cornic and Masacci 1996; Muller and Whitsitt 1996). Non-stomatal mechanisms under prolonged or severe soil drought include changes in chlorophyll synthesis, functional and structural changes in chloroplasts and also disturbances in accumulation and distribution of assimilation products (Medrano et al. 2002). However, it is known that during drought stress, plants with C_4 photosynthesis increase water use efficiency and suppress photorespiration. Thus, C_4 plants are often more competitive than C_3 plants in drought-prone areas (Edwards and Ku 1987; King 2011).

Relative loss of intracellular electrolytes from leaf tissues (LI)

In drought sensitive genotypes of maize and triticale, the values of indexes of leaf injury by soil drought (D_{35}, D_{20}), osmotic stress (−0.47, −0.85, −1.30 MPa), and heat temperature (25, 35, 45 °C) were in general higher than in drought resistant genotypes and in most cases the differences between resistant and sensitive genotypes were statistically significant (Table 7). In treatment D_{35}, the values of coefficients of variation (CV) in maize and triticale were higher than in D_{20} treatment. Under osmotic stress, higher values of CV were found in −0.85 MPa treatment and under high temperature stress in 35 °C treatment.

The ability to maintain the structure and function of cytoplasmatic membranes under water deficit is one of the most important physiological traits. Conductometric measurements of LI are applied as a screening test for the estimation of tolerance to various stresses (Vietor et al. 1977; Richards 1978; Blum and Ebercon 1981; Poljakoff-Mayber 1981; Martinielio and Lorenzoni 1985; Palta 1990). Differences between sensitive and resistant genotypes might stem from the fact that drought resistant genotypes possess more efficient mechanisms protecting membrane functions and structure. Drought stress causes a loosening of lamellar membranes in chloroplasts, loss of a certain amount of grana, and increase in the level of coarse-grain matrix (Haupt-Harting and Fock 2002; Lawlor and Cornic 2002; Tang et al. 2002). Some authors suggest that drought resistant plant species show stronger binding of chlorophyll molecules to the lipid-protein complex of chloroplast membranes (Smirnoff and Colombe 1988; Bukhov et al. 1990). Our earlier results indicate that leaf age is very important because differences in LI between drought resistant and sensitive genotypes were smallest in the oldest and youngest leaves, though the greatest differences were observed in the leaves where cellular divisions had taken place and which had reached the maximal area (Grzesiak et al. 2006).

Correlations among stress parameters (DW_S, DW_R, DW_S/DW_R) were significant, except for the relationship between DW_S and DW_R in maize. In this experiment the correlations between DW_R and other traits were not significant except for seedling survival (SS). For both species, high and significant correlations were found between measurements of membrane injures due to drought (LI_D), osmotic stress (LI_{OS}), and high temperature (LI_{HT}). Also for both species, high and significant correlations were

Table 6 Leaf water potential (ψ) of maize and triticale genotypes grown in moderate (D_{35}) and severe (D_{20}) soil drought and index of excised-leaf water loss (WL) from leaf seedlings grown in control condition after 12 and 24 h

Genotype	Leaf water potential (ψ)		Water loss (WL—g H_2O/g DW)	
	D_{35}	D_{20}	12 h	24 h
Maize				
Ankora	2.69	2.40	5.07	7.11
Garst 8702	2.39	2.39	5.65	6.99
Garst 8388	2.31	2.41	5.69	6.07
Garst R5515	2.49	2.49	4.18	7.13
Pioneer-38-F-70	2.71	2.35	5.55	7.25
Nova	2.58	2.10	5.55	8.39
Pioneer-39-G-12	2.55	2.41	5.64	8.14
Pioneer-39-R-10	2.11	2.39	5.84	7.1
Funk's G4083	2.39	2.18	3.41	6.07
Pioneer 3957	2.41	2.27	3.18	5.23
Pioneer 3925	2.31	2.25	2.97	6.21
Garst 8344	2.19	2.18	3.39	6.21
Tina	2.07	2.21	3.13	6.55
Mean	2.40	2.31	4.56	6.80
Range	2.07–2.71	2.10–2.41	2.97–5.84	5.23–8.39
$LSD_{0.05}$	0.05	0.07	0.58	0.39
CV	8.5	5.1	26.0	12.8
r versus DSI_{GY}	0.682*	0.645*	0.773**	0.511^{NS}
Triticale				
Maja	2.39	2.39	2.42	3.50
CHD-147	2.31	2.49	3.58	4.07
Mieszko	2.25	2.18	2.54	5.07
CHD-12	2.45	2.35	3.57	4.04
Kargo	2.33	2.25	3.39	5.11
MAH	2.21	2.00	2.41	4.41
Gabo	2.37	2.25	3.18	4.39
Wanad	2.41	2.11	2.07	3.52
Migo	2.18	2.18	1.27	3.07
CHD-220	2.13	2.07	2.07	3.50
CHD-247	2.11	2.11	2.13	3.00
Mean	2.28	2.22	2.60	4.00
Range	2.11–2.45	2.00–2.49	1.27–3.58	3.00–5.11
$LSD_{0.05}$	0.04	0.08	0.18	0.25
CV	5.1	6.7	27.9	18.7
r versus DSI_{GY}	0.579^{NS}	0.637*	0.714*	0.655*

The results of ψ are shown as a stress index (SI). Mean values, range of means, coefficient of variation (CV) and correlation coefficient (*r*) between measured traits and drought susceptibility index (DSI_{GY}). Genotypes are ordered according to the DSI_{GY} value ($n = 5$)

Number of degree of freedom (df) for critical values of (*r*) was for maize 11 and for triticale 9

NS, *r*-Correlation coefficient not significant

*, ** *r*-Correlation coefficient significant at the 0.05 and 0.01 level, respectively

found between leaf water potential (ψ) and water loss (WL) and between FG and PI, except for maize (Table 8).

Most of physiological processes are affected by the stresses of soil drought, osmotic and high temperatures (Levitt 1980). The usefulness of methods for studying plant drought tolerance has been discussed in many papers and reviews (Blum et al. 1980; Kpoghomou et al. 1990; Zagdańska 1992; Reynolds 2002). According to some authors,

Table 7 Leaf injury index (LI) of maize and triticale seedlings grown in moderate (D_{35}) and severe (D_{20}) soil drought conditions and data of conductivity tests of leaf injury by osmotic drought stress (−0.47, −0.85, −1.30 MPa) and by high temperature stress (25, 35, 45 °C)

Genotype	Soil drought		Osmotic stress (MPa)			High temperature stress (°C)		
	D_{35}	D_{20}	−0.47	−0.85	−1.30	25	35	45
Maize								
Ankora	41.1	50.9	22.2	42.0	48.7	20.9	32.2	35.0
Garst 8702	39.5	45.3	25.6	45.0	27.0	24.3	30.0	58.0
Garst 8388	39.5	45.0	25.5	34.5	45.1	24.2	40.7	47.5
Garst R5515	39.5	42.3	24.3	31.8	42.4	23.0	34.3	44.8
Pioneer-38-F-70	31.2	36.8	19.5	26.3	26.5	18.2	29.5	40.4
Nova	31.2	40.8	20	30.3	40.9	18.7	30.0	43.3
Pioneer-39-G-12	31.2	36.8	18.6	18.0	22.2	17.3	28.6	39.0
Pioneer-39-R-10	30.6	35.3	18.2	21.8	22.5	16.9	28.2	37.8
Funk's G4083	13.1	36.6	20	17.5	35.0	18.7	25.0	40
Pioneer 3957	18.4	36.3	22.2	25.8	26.0	20.9	24.2	38.8
Pioneer 3925	13.2	35.8	20.1	25.3	26.0	17.8	20.0	40.3
Garst 8344	18.1	33.9	22.2	23.4	34.0	20.9	32.2	36.4
Tina	11.1	30	20.9	17.5	23.6	19.6	21.2	35.5
Mean	27.5	38.9	21.5	27.6	32.3	20.1	28.9	41.3
Range	11.1–41.1	30.0–50.9	18.2–25.6	17.5–45.0	22.2–48.7	16.9–24.3	20.0–40.7	35.0–58.0
$LSD_{0.05}$	1.51	1.88	1.25	1.35	1.13	1.18	1.08	1.13
CV	40.9	14.6	11.4	32.0	28.7	12.5	19.2	15.0
r versus DSI_{GY}	0.953**	0.868**	0.412^{NS}	0.751**	0.540^{NS}	0.438^{NS}	0.707**	0.511^{NS}
Triticale								
Maja	24.5	30.6	16.5	37.5	39.4	12.7	36.4	52.4
CHD-147	26.4	35.9	18.7	33.3	38.1	14.4	34.2	48.5
Mieszko	25.3	31.5	20	34.5	37.9	18.2	34.8	55.9
CHD-12	25.4	39.7	18.5	28.8	45.7	17.8	42.8	49.5
Kargo	25.4	32.8	16.2	39.1	37.4	12.0	35.4	50.2
MAH	23.4	30.6	14.5	27.6	28.8	12.7	25.8	41.6
Gabo	21.4	27.5	22.2	25.4	26.4	15.7	22.5	44.4
Wanad	19.8	28.6	17.5	18.8	21.5	10.2	18.6	44.5
Migo	19.9	25.5	15.1	18.8	25.8	13.3	22.7	48.2
CHD-220	13.5	23.6	15.5	19.5	22.7	13.7	20.8	45.2
CHD-247	11.5	23.3	14.5	20.3	21.1	11.8	18.2	44.6
Mean	21.5	30	17.2	27.6	31.3	13.9	28.4	47.7
Range	11.5–25.4	23.3–35.9	14.5–22.2	18.8–39.1	21.1–45.7	10.2–17.8	18.2–42.8	41.6–55.9
$LSD_{0.05}$	1.18	1.39	1.41	1.07	1.08	0.59	1.11	0.99
CV	23.3	16.8	14.3	27.8	27.3	18.0	30.0	8.7
r versus DSI_{GY}	0.857**	0.820**	0.404^{NS}	0.911**	0.886**	0.436^{NS}	0.876**	0.603*

Mean values, range of means, coefficient of variation (CV) and correlation coefficient (*r*) between measured traits and drought susceptibility index (DSI_{GY}). Genotypes are ordered according to the DSI_{GY} value ($n = 5$)

Number of degree of freedom (df) for critical values of (*r*) were for maize 11 and for triticale 9

NS, *r*-Correlation coefficient not significant

*, ** *r*″-Correlation coefficient significant at the 0.05 or 0.01 level, respectively

besides the evaluation criteria used in the present research, positive results were also obtained by utilizing the measurements of canopy infra-red temperature, changes of leaf color and responses of root architecture (Clarke and McCaig 1982; Stanley 1990). Partly positive results were obtained in tests of the plant's ability to reduce leaf area, the development of wax bloom on leaves, leaf rolling and the ability of the leaf to hold water (Passioura et al. 1993).

Table 8 Correlation coefficients (r) among physiological stress traits in maize and triticale genotypes

Traits	Species	Traits										
		DW_R	DW_S/DW_R	L_S, L_{SA}, L_N	LI_D	LI_{OS}	LI_{HT}	ψ	WL	FG	PI	SS
DW_S	Maize	0.427^{NS}	0.900***	0.845***	−0.581**	-0.381^{NS}	−0.537*	−0.634**	−0.672**	0.712***	0.362^{NS}	0.793***
	Triticale	0.689**	0.854***	0.433^{NS}	−0.670**	-0.560^{NS}	-0.408^{NS}	-0.275^{NS}	−0.586*	0.555^{NS}	0.343^{NS}	0.657**
DW_R	Maize		-0.002^{NS}	0.315^{NS}	-0.216^{NS}	-0.233^{NS}	-0.189^{NS}	-0.390^{NS}	-0.092^{NS}	0.349^{NS}	0.267^{NS}	0.706***
	Triticale		0.216^{NS}	0.288^{NS}	-0.423^{NS}	-0.303^{NS}	-0.084^{NS}	-0.349^{NS}	-0.518^{NS}	0.340^{NS}	0.380^{NS}	0.641**
DW_S/DW_R	Maize			0.793***	−0.533*	-0.301^{NS}	−0.489*	−0.489*	−0.714***	0.616**	0.232^{NS}	0.533*
	Triticale			0.342^{NS}	−0.555*	-0.490^{NS}	-0.437^{NS}	-0.065^{NS}	-0.376^{NS}	0.461^{NS}	0.149^{NS}	0.406^{NS}
L_S, L_{SA}, L_N	Maize				−0.760***	-0.406^{NS}	−0.618**	−0.619**	−0.670**	0.664**	0.597**	0.633**
	Triticale				−0.678**	−0.714**	-0.573^{NS}	-0.386^{NS}	−0.722**	0.324^{NS}	0.042^{NS}	0.325^{NS}
LI_D	Maize					0.589**	0.857***	0.539*	0.423^{NS}	−0.778***	−0.695***	−0.495*
	Triticale					0.973***	0.875***	0.770***	0.902***	−0.831***	-0.530^{NS}	−0.691**
LI_{OS}	Maize						0.837***	0.216^{NS}	0.299^{NS}	−0.660**	-0.350^{NS}	-0.317^{NS}
	Triticale						0.922***	0.733**	0.880***	−0.752***	-0.364^{NS}	−0.594*
LI_{HT}	Maize							0.353^{NS}	0.356^{NS}	−0.868***	−0.576**	-0.381^{NS}
	Triticale							0.635**	0.715**	−0.770***	-0.373^{NS}	-0.466^{NS}
ψ	Maize								0.532*	-0.286^{NS}	−0.652**	−0.799***
	Triticale								0.673**	−0.743***	−0.672**	-0.532^{NS}
WL	Maize									-0.253^{NS}	-0.155^{NS}	-0.472^{NS}
	Triticale									−0.733**	-0.459^{NS}	−0.806***
FG	Maize										0.434^{NS}	0.523*
	Triticale										0.748***	0.819***
PI	Maize											0.357^{NS}
	Triticale											0.567^{NS}

Data for calculation of correlation coefficient (r) for traits DW_S, DW_R and DW_S/DW_R were SI for treatment D_{35}, for L_S, L_{SA} and L_N were SI for total length for seminal, seminal adventitious and nodal roots, for LI_D were mean values of treatment D_{35} and D_{20}, for LI $_{OS}$ and LI_{HT} mean values of treatments −0.47, −0.85 and −1.30 MPa and 25, 35 and 45 °C, respectively, for ψ were mean values of SI in treatment D_{35} and D_{20}, for WL were mean values of treatments 12 and 24 h and for FG, PI and SS were mean values of treatments −0.47, −0.85 and −1.30 MPa. Number of degree of freedom (df) for critical values of (r) was for maize 11 and for triticale 9

^{NS} r-Correlation coefficient not significant

*, **, *** r-Correlation coefficient significant at the 0.10, 0.05 and 0.01 level, respectively

On the other hand, the effectiveness of the evaluation of tolerance based on the measurement of different parameters of leaf gaseous exchange and the content of various metabolites, including proline, has not been definitely confirmed. (Hanson and Nelson 1985; Farquhar et al. 1993; Dubey 1997; Bandurska and Stroiński 2003).

The maize and triticale genotypes used in these experiments show a relatively wide range of drought tolerance. This study contributes to the understanding of responses of different genotypes to drought, though in this research, the method of estimating drought susceptibility index was relatively simple and did not take into account other important factors of soil–water-plant relationship. The correlations between DSI_{GY} and laboratory tests (LI, FG, SS) showed that they are good indicators of plant drought tolerance. The correlation coefficients between LI_{OS} and LI_{HT} tests were, in most of the considered cases, statistically significant, thus indicating that in maize and triticale the mechanisms of membrane injury caused by simulated drought or high temperature were physiologically similar. It can be concluded that an approach to the breeding of maize and triticale for drought tolerance using these tests can be implemented on the basis of separate selection for each trait or for all of them simultaneously. In that case, it would be necessary to determine the importance of the trait in relation to the growth phase, drought timing and level as well as associations with morphological traits contributing to drought tolerance (Kono et al. 1987; Kpoghomou et al. 1990).

The results presented in this paper, our earlier research (Grzesiak 1990; Grzesiak et al. 2012) and the results of other authors (Lorens et al. 1987; Martinielio and Lorenzoni 1985; Kono et al. 1987) confirm the existence of a wide range of genotypic variability of response to drought in cereal plants. In maize and triticale, similar to other crop plants, the physiological reasons for this variability have not as yet been fully recognized and explained. In the literature one can find many contradictory conclusions, as the reduction of crop yield of the tested genotypes depends not only on the drought level and its duration, but also on the phase of plant growth and development and interaction with other environmental factors (Hanson and Nelson 1985; Blum 1988; Naylor and Su 1998).

The grounds for genotypic variation of drought tolerance in cereals has not yet been entirely elucidated and future research is necessary (Zagdańska 1992; Royo et al. 2000; Reynolds 2002). The frequent occurrence of drought stress in many regions of the world and the deteriorating water conditions for plant growth and productivity have raised interest in research into the responses of crops to periodic water deficiency. Progress in the breeding of drought resistant cereal plants requires future study of the physiological mechanisms underlying the responses of plants to water stress. It is a widely held opinion that the breeding of drought resistant crop plants will not be an easy task (Hanson and Nelson 1985; Zagdańska 1992; Reynolds 2002). The complexity of the property of drought tolerance will make it necessary to take into consideration various tolerance evaluation criteria in the breeding programs (Levitt 1980; Turner 1986; Blum 1988; Richards 1991; Jones 1993; King 2011).

Author contribution MTG, FJ, PW, KH designed the research; MTG, PS, IM and TG conducted the research; MTG, FJ, PS and KH analyzed the data; MTG, FJ, IM and KH wrote the paper; MTG had primary responsibility for the final content. All authors have read and approved the final manuscript.

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