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EFFECTS OF ARTIFICIALLY DROUGHT INDUCED BY CHEMICAL DESICCATION IN SEVERAL WINTER WHEAT (*Triticum aestivum* L.) GENOTYPES

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ABSTRACT

Climatic changes affect crop production and wheat as one of the main crop worldwide suffers production losses year by year caused by drought stress. The aim of this study was to identify valuables winter wheat genotypes concerning to drought tolerance. A set of 113 Romanian and foreign genotypes were analysed for drought tolerance over two experimental years in field conditions at Agricultural Research and Development Station Turda. Simulating drought effect was realized by applying the desiccant solution ($NaClO_3$ - 2%) on wheat plants at 14 days after anthesis. Morpho-productive traits, such as number of grains per spike; weight of grains per spike; thousand kernel weight and grain yield were analysed as a response of drought stress conditions. Of the main yield components: weight of grains/spike and thousand kernel weight were more affected traits. Simulation of drought effect allowed identification of valuable cultivars, capable to limiting the drought impact on yield components and finally on grain yield. Among studied cultivars, six perspective lines from Turda breeding program were noted for a small reduction rate of thousand kernel weight and weight of grain per spike as main yield components. They will be used as genetic resources for improvement of drought tolerance.

Keywords: climate changes, desiccant, drought tolerance, winter wheat.

INTRODUCTION

Wheat (*Triticum aestivum* L.) represents more than 30% of the world' cereals area, with over 220 million ha cultivated worldwide, often under abiotic stress. Wheat crop is sensitive to heat and drought stresses mainly at the flowering and grain development stages, which have a negative impact on yield and grain quality (Kulkarni et al., 2017). Grain yield under post-anthesis drought stress is one of the most complex traits, which is inherited quantitatively (Zaynali et al., 2011). A recent study analysing the data of studies published from 1980 to 2015 reported up to 21 and 40% yield reductions in wheat (*Triticum aestivum* L.) and maize (*Zea mays* L.) caused by drought on a global scale (Daryanto et al., 2016). Zampieri et al. (2017) also affirmed that annual production variability estimated at ~40% is mainly caused by heat waves and drought situations in major wheat producing areas throughout the world. Among all the abiotic stress factors that limit crop productivity, drought is the most devastating one and the most difficult to approach by breeders. Past breeding efforts to improve drought tolerance have been hindered by its quantitative genetic basis and by the poor understanding of the physiological basis of yield under water-limited conditions (Tuberosa and Salvi, 2006). Tolerance is a complicated trait which is controlled by polygenes and their expressions are influenced by various environmental factors. Current breeding programs continue to make progress through commonly used breeding approaches.

The outcome of combining heat-adaptive physiological traits by hybridisation is not always predictable in terms of the net effect

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on crop productivity, especially over a wide range of environments (Cossani and Reynolds, 2012). Because field screening for post-anthesis drought tolerance is difficult, effective and validated methods to simulate drought in order to identify sources of tolerance can facilitate screening of breeding materials (Kamal et al., 2018). For the selection of cereal cultivars tolerant to drought, chemical desiccants can be applied to simulate the effect of stress under field conditions, by inhibiting carbon assimilation (Hossain et al., 1990; Nicolas et al., 1993; Budakli et al., 2007)

Cultivars that translocate more carbohydrate reserves to the grains were better able to maintain a stable kernel weight under desiccation conditions (Blum et al., 1983). In addition, the resultant reduction in grain weight is correlated significantly with the reduction in grain weight due to natural drought (Nicholas and Turner, 1993).

Drought during reproductive and grain-filling phases causes substantial yield reductions in wheat, which are primarily due to accelerated leaf senescence (Yang et al., 2002), oxidative damage to photo-assimilatory machinery (Farooq et al., 2014), reduced rates of carbon fixation and assimilate translocation (Asada, 2006), pollen sterility (Dorion et al., 1996; Cattivelli et al., 2008), reduced grain set and development (Ahmadi and Baker, 2001; Nawaz et al., 2013), and reduced sink capacity (Farooq et al., 2014).

As a counteraction of environment limiting conditions to plant performance expression, wheat breeders must continuously improve the wheat genome by using different genes sources. Exposure to extremely high temperatures (i.e. heat stress) leads to plant damages by inducing perturbations in cellular structures and metabolic processes (Nakamoto and Hiyama, 1999). On the other hand the presence of a wheat cultivar capable to perform in drought conditions can capitalize the land unused because of deficient water availability (Bănică et al., 2008). The climate changes characterized by serious droughts have led to an increasing frequency of drought years.

The present study had as purpose to identify winter wheat genotypes characterized by a high yield capacity, endowed with a good tolerance to the main limited yield factor - drought.

MATERIAL AND METHODS

One hundred thirteen Romanian and foreign genotypes of winter wheat (Table 1) were tested for drought tolerance. Drought tolerance test was done using the method proposed by Blum (1988). The working method consists in using the desiccant $NaClO_3$, which induces the most severe drought effect (Ongom et al., 2016) in a concentration of 2%, applied 14 days after flowering, in order to cause the drying of the plant. Previously, Haley and Quick (1993) reported the use of $NaClO_3$, applied at 2% concentration, to achieve early generation selection of wheat (*Triticum aestivum* L.) for post-anthesis drought tolerance. Significant kernel weight reduction was reported among wheat varieties under $NaClO_3$ desiccation stress, and this response was correlated with reactions to late season drought (Cseuz et al., 2002).

Table 1. Names of tested local and foreign genotypes

From ARDS Turda:							
1.	Andrada	31.	T.23-12	61.	T.65-12	89.	Glosa
2.	Apullum	32.	T.24-04	62.	T.66-12	90.	Gruia
3.	Arieşan	33.	T.24-11	63.	T.67-10	91.	Izvor
4.	Dumbrava	34.	T.263-03	64.	T.71-11	92.	Litera
5.	T.100-01	35.	T.27-12	65.	T.7-12	93.	Miranda
6.	T.10-12	36.	T.279-03	66.	T.72-11	94.	Otilia
7.	T.104-11	37.	T.28-12	67.	T.74-12	95.	Partener
8.	T.109-11	38.	T.29-12	68.	T.76-12	96.	Pitar
9.	T.1-12	39.	T.30-12	69.	T.77-12	97.	Retezat
10.	T.112-11	40.	T.34-11	70.	T.78-12	98.	Roditor
11.	T.113-11	41.	T.38-12	71.	T.83-05	99.	Rovine
12.	T.118-11	42.	T.39-12	72.	T.83-10	100.	Spornic
13.	T.124-11	43.	T.40-12	73.	T.9-05	101.	Gasparom
14.	T.125-05	44.	T.41-12	74.	T.9-12	*Foreign:*	
15.	T.126-11	45.	T.42-04	75.	T.94-11	102.	Apache
16.	T.13-12	46.	T.42-05	76.	T.94-12	103.	Capo
17.	T.135-08	47	T.45-12	77.	T.99-11	104.	Cubus
18.	T.135-11	48.	T.47-12	78.	T.158-11	105.	Element
19.	T.136-03	49.	T.49-12	*From NARDI Fundulea R&D Institute and other research centers:*		106.	Exotic
20.	T.143-11	50.	T.5.12			107.	Gallio
21.	T.145-11	51.	T.52-12	79.	Aniversar	108.	Josef
22.	T.150-11	52.	T.53-12	80.	Ardeal	109.	Kolo
23.	T.15-12	53.	T.54-01	81.	Boema	110.	Renan
24.	T.16-12	54.	T.55-01	82.	Crişana	111.	Serina
25.	T.17-12	55.	T.57-12	83.	Delabrad	112.	Bezostaya
26.	T.178-11	56.	T.60-12	84.	Drobeta	113.	Cristina
27.	T.19-11	57.	T.61-12	85.	Dropia		
28.	T.205-11	58.	T.6-12	86.	Esenţial		
29.	T.208-11	59.	T.62-01	87.	Faur		
30.	T.216-03	60.	T.65-11	88.	Flamura 85		

The research was conducted at the Agricultural Research and Development Station from Turda, as multifactorial trials in two repetitions during two field experimental years. The experimental factors and the graduations of our research were:

- factor A – genotype, with 113 graduations;
- factor B – the year with 2 graduations;
- factor C – the desiccation with 2 graduations.

The experimental design consisted in four rows with 100 cm length. Two rows were considered as control and the others as treatment (sprayed with desiccant solution on the whole green plant). All spikes from rows in each plot were harvested individually at physiological maturity. Morphological traits, such as number of grains/spike (NGS), weight of grains/spike (WGS), thousand kernel weight (TKW) was measured. Also, based on these measurements grain yield (GY) was estimated.

To compare the effects of treatments and to estimate drought tolerance of genotypes we calculated the indices mentioned in Table 2.

Table 2. Drought tolerance indices

Index name	Equation	Reference
Stress tolerance index	$STI = (Y_p \times Y_s) / \overline{Y_p}^2$	(Fernandez, 1992; Kristin et al., 1997)
Yield Index	$YI = Y_s / \overline{Y_s}$	(Gavuzzi et al., 1997)
Drought resistance index	$DI = (Y_s \times (Y_s / Y_p)) / \overline{Y_s}$	(Lan, 1998)
Stress susceptibility index	$SSI = 1 - (Y_s / Y_p) / SI;$ $SI = 1 - (\overline{Y_s} / \overline{Y_p})$	(Fischer and Maurer, 1978)
Stress susceptibility percentage	$SSPI = (Y_p - Y_s / 2(\overline{Y_p})) \times 100$	(Moosavi et al., 2008)
Geometric mean productivity	$GMP = \sqrt{Y_s \times Y_p}$	(Kristin et al., 1997)
Mean production	$MP = (Y_s + Y_p)/2$	(Rosielle and Hamblin, 1981)
Relative drought index	$RDI = [Y_s / Y_p] / [\overline{Y_s} / \overline{Y_p}]$	(Fischer and Wood, 1979)
Harmonic Mean	$HARM = \frac{2(Y_p \, x \, Y_s)}{Y_p \, x \, Y_s}$	(Jafari et al., 2009)

* Y_s are stress and and Y_p optimal (normal) yield of a given genotype, respectively;
$\overline{Y_s}$ and $\overline{Y_p}$ are average yield of all genotypes under stress and optimal conditions, respectively.

The climate conditions of the two experimental years (Table 3) had some particularities regarding the rainfall conditions, alternating the optimal rainfall conditions with periods of drought accompanied by heat stress during some important phenological stages.

Table 3. The climate conditions during the experimental years

Month	Rainfall (mm/m^2)		Temperature (°C)		Multiannual mean	
	Year I	Year II	Year I	Year II	mm/m^2	°C
October	66.8	67.4	11.2	10.8	35.6	9.5
November	5.9	34.2	7.1	5.7	28.5	3.9
December	3.3	86.6	-1.7	1.3	27.1	-1.4
January	51.6	12.3	0.5	-0.7	21.8	-3.4
February	15.5	20.9	3.8	0.6	18.8	-0.9
March	23.1	12.8	8.8	5.5	23.6	4.7
April	72.0	32.2	11.4	9.6	45.9	9.9
May	66.2	66.0	15.1	15.8	68.7	15.0
June	48.4	115.7	18.5	19.4	84.8	17.9
July	144.4	52.2	20.4	22.3	77.1	19.7
Total Average	497.2	500.3	9.51	9.03	431.9	7.49

RESULTS AND DISCUSSION

Significant differences were obtained for the traits measured both for genotype and treatments (Table 4). The reaction of genotypes regarding yield and yield components to simulated drought stress suggest that among these genotypes, sources of drought tolerance for new cultivar development could be identified. Desiccant application had a severe impact on plants agronomic traits, the impact intensity being directly related to the development stage of studied traits; for instance, the intensity of treatment application on number of grains/spike was very significant, this yield component being already present at the time of treatment. However, the influence of treatment on this character can be explained through that wheat plants can abandon the development of some seed in stress conditions.

Table 4. Analysis of variance for main yield components and grain yield under two different treatments

Source of variation	df	MS	F	P-value
Number of grains/spike				
Genotypes	112	40.718	6.67	< 0.0001
Treatment	1	515.801	84.43	< 0.0001
Error	112	6.109		
Weight of grains/spike				
Genotypes	112	0.10023	4.00	< 0.0001
Treatment	1	33.00003	1316.84	< 0.0001
Error	112	0.02506		
TKW				
Genotypes	112	21.746	3.28	< 0.0001
Treatment	1	10937.96	1651.68	< 0.0001
Error	112	6.6223		
Grain yield				
Genotypes	112	0.9849	4.72	< 0.0001
Treatment	1	277.5565	1331.11	< 0.0001
Error	112	0.2085		
Total	225			

Application of desiccant to simulate the drought effect had a significant impact on the main yield components and also on grain yield (Table 5). Even if the effect of drought simulation is normal to influence the traits as weight of grains/spike, thousand kernel weight and grain yield due to the long-term formation of those; number of grains/spike were also influenced by desiccant application caused by plant adaptation to stress condition which has a tendency to reduce its performance. Generally, treatment application caused decreased values in comparison to untreated plants, determining in the same time an increased variability of studied traits. High range value of weight of grains/spike (1.46) at stress conditions, suggests that among the studied winter wheat there was a large variability for this character and also possibility to identify contrasting genotypes.

The large amplitude for yield components and grain yield for treated material highlights contrasting reaction of different genotypes to artificially simulated drought stress conditions.

Table 5. The parameters of variability for and yield components in the studied winter wheat genotypes

Statistics		Number of grains/spike		Weight of grains/spike		TKW		Grain yield	
		Treated	Untreated	Treated	Untreated	Treated	Untreated	Treated	Untreated
		113 genotypes							
Average		44.60	47.62	1.54	2.30	34.30	48.22	6.90	9.12
Range		28.95	26.10	1.46	1.03	22.04	15.88	3.90	4.19
Median		44.90	47.30	1.55	2.30	34.19	47.96	6.86	9.06
Standard deviation		4.96	4.72	0.25	0.25	4.15	3.34	0.78	0.76
Amplitude	Min.	32.38	33.2	0.94	1.84	24.38	40.65	4.80	7.73
	Max.	61.33	59.30	2.39	2.87	46.43	56.53	8.70	11.92
CV (s%)		11.12	9.91	16.23	10.87	12.10	6.93	11.30	8.33

The relationship between yield components at drought stress conditions and normal conditions is presented in Table 6. In case of normal conditions, the coefficient of correlation between yield components and grain yield are strong and positive, being negative in case of relation between number of grains per spike and thousand kernel weight. Comparing the yield components, untreated and treated plants, the relation between those varied depending on ability of individual genotype to adapt to stress conditions.

Application of desiccant to simulate the drought effect increased the intensity of relationship between some yield components, suggesting a different reaction of biologic material especially between number of grains/spike and thousand kernel weight, grain yield and all yield components.

Table 6. Correlation between yield components under normal and stress conditions

Traits		NGS	WGS	TKW	GY
Untreated (control)					
Untreated	NGS	1.00	0.80**	-0.21*	0.35**
	WGS		1.00	0.40**	0.38**
	TKW			1.00	0.11
	GY				1.00
Untreated (control)					
Treated	NGS	0.74**	0.58**	-0.18	0.26**
	WGS	0.48**	0.60**	0.26**	0.28**
	TKW	-0.08	0.23*	0.55**	0.13
	GY	0.35**	0.46**	0.25**	0.65**
Treated					
Treated	NGS	1.00	0.69**	-0.07	0.46**
	WGS		1.00	0.65**	0.71**
	TKW			1.00	0.53**
	GY				1.00

* Significant at 5% level of significance;
** Significant at 1% level of significance.

The estimated indicators of drought tolerance (Table 7) indicated that the identification of drought tolerant cultivars can be appreciated based on yield and yield components.

The increasing values of correlation coefficient between drought tolerance indicators and yield components in case of desiccant application confirm that this indicators can be used for drought stress conditions. Naghavi et al. (2013) and Gholinezhad et al. (2014) suggested that YI, SSPI, STI, MP, GMP and HARM could be used as the most suitable indicators for screening drought tolerant cultivars. Using more estimated drought tolerance indices can improve the chance to identify a desirable genotype.

The increased values of correlation coefficients between yield/yield components and drought tolerance indicators suggest that weight of grains per spike, alongside grain yield are the most suitable traits for drought tolerance.

Table 7. Correlation coefficients between yield traits and drought tolerance indices

	STI	YI	DI	SSI	SSPI	GMP	MP	RDI	HARM
Untreated (control)									
NGS	0.38**	0.35**	0.27**	-0.12	-0.01	0.39**	0.39**	0.12	0.35**
WGS	0.46**	0.46**	0.40**	-0.25**	-0.11	0.47**	0.46**	0.25**	0.46**
TKW	0.20*	0.25**	0.26**	-0.23*	-0.18	0.21*	0.20*	0.23*	0.25**
GY	0.88**	0.65**	0.35**	0.10	0.40**	0.88**	0.91**	-0.10	0.65**
Treated									
NGS	0.41**	0.46**	0.44**	-0.36**	-0.24*	0.41**	0.40**	0.36**	0.46**
WGS	0.57**	0.71**	0.75**	-0.67**	-0.53**	0.58**	0.55**	0.67**	0.71**
TKW	0.39**	0.53**	0.60**	-0.58**	-0.49**	0.40**	0.37**	0.58**	0.53**
GY	0.93**	1.00**	0.94**	-0.69**	-0.44**	0.94**	0.91**	0.69**	1.00**

* Significant at 5% level of significance;
** Significant at 1% level of significance.

Considering weight of grain/ spike and thousand kernel weight as the main yield components, which are more influenced by simulated post-anthesis drought stress condition, we calculated the rate of reduction of those as an indicator of drought tolerance for each genotype (Figure 1). Based on this coefficient we could make a distribution of genotypes regarding the rate of reduction of the two characters. Thus, the genotypes with a rate of reduction between 0-12.5% were considered to have a high tolerance to drought stress conditions, between 12.51-25.0% as medium-good tolerance, 25.01-37.5% with a medium tolerance, 37.51-50.0% as medium sensitive and over 50.01% sensitive genotypes to drought, respectively.

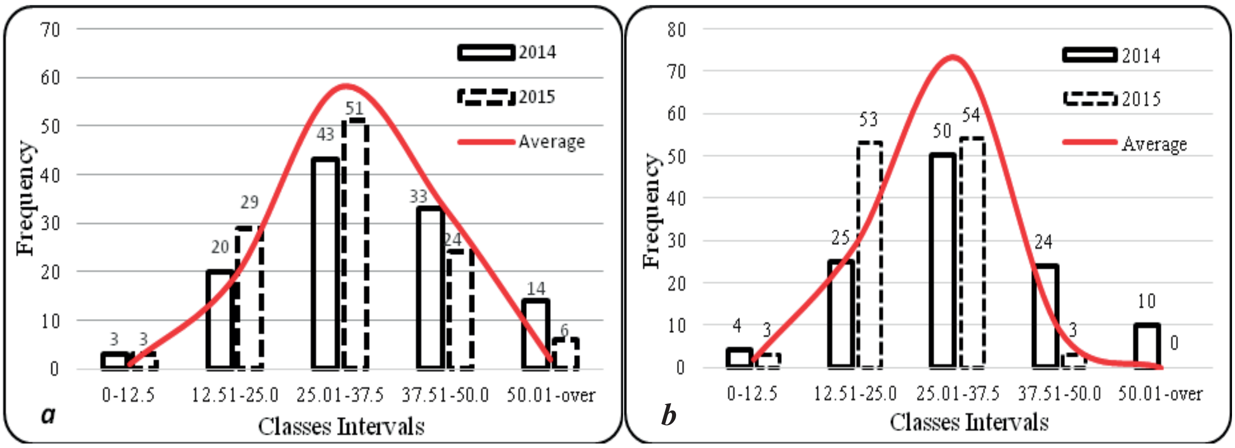

Figure 1. The distribution of the studied wheat genotypes regarding the rate of reduction of the grain weight/spike (a) and TKW (b)

From the group of studied genotypes we identified some winter wheat perspective lines with a medium-good or a high tolerance of drought stress conditions, both for thousand kernel weight and weight of grains/spike (Figure 2). Also, it can be observed that in stress conditions caused by simulated drought relationship between thousand kernel weight and weight of grains/spike became stronger (r = 0.65***).

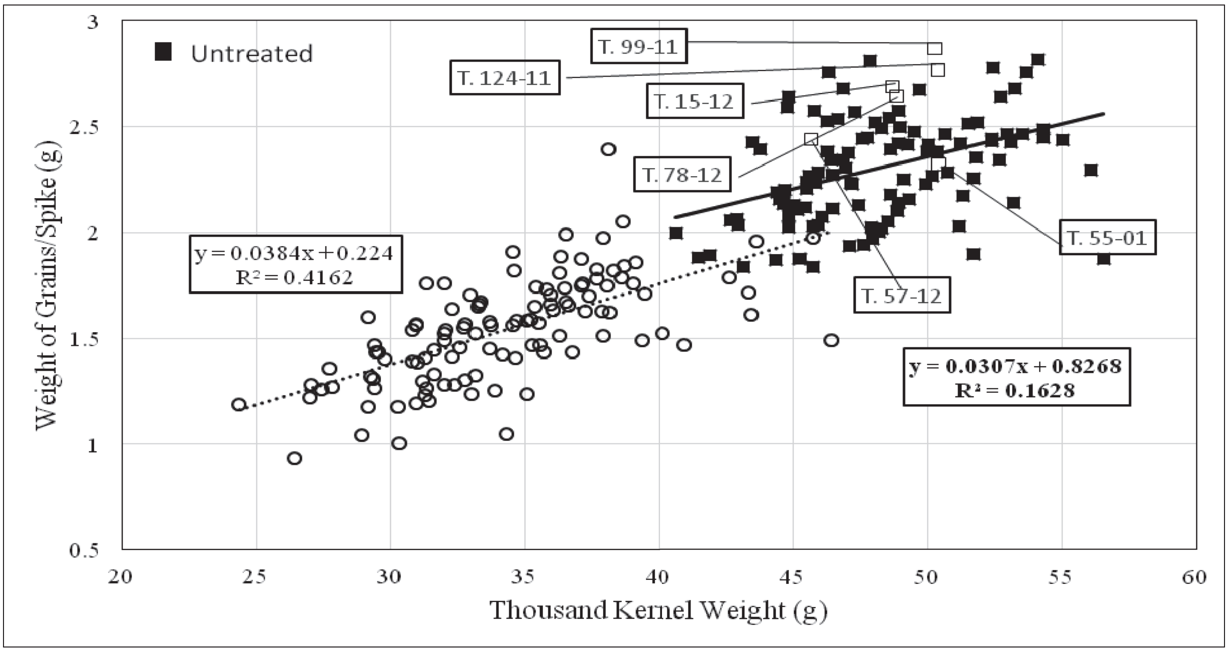

Figure 2. The behaviour of wheat genotypes under normal and drought conditions regarding weight of grains/spike and thousand kernels weight

The principal component analysis of the winter wheat group genotypes (Figure 3) highlights almost the same genotypes which were identified before based on the relationship between weight of grains/spike and thousand kernel weight.

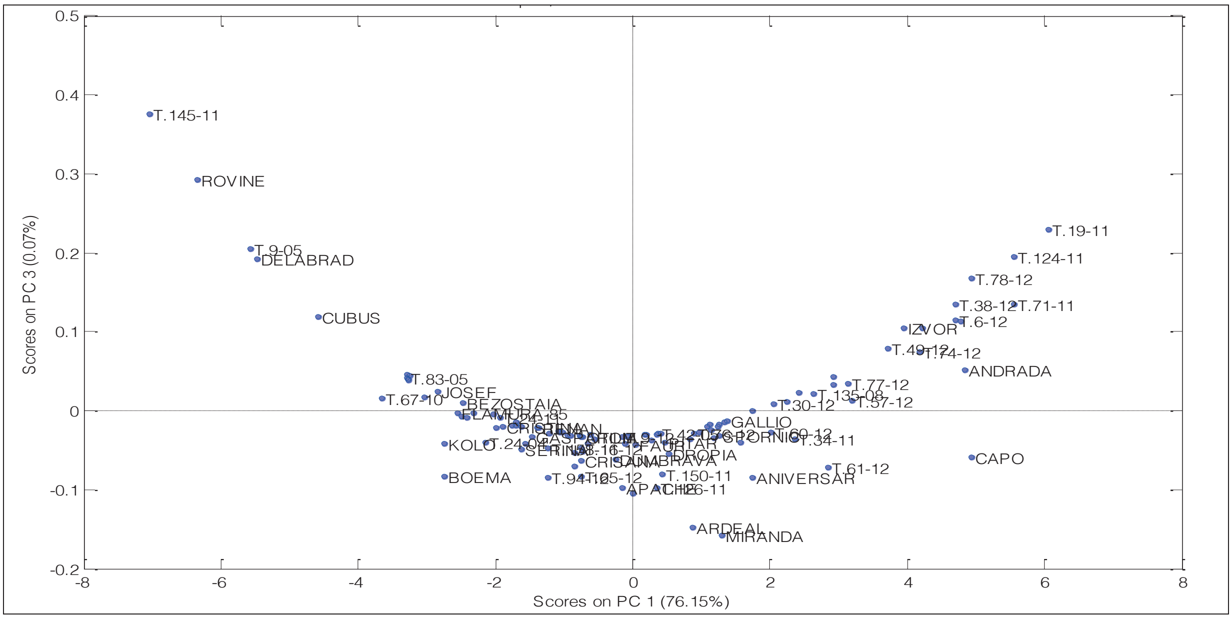

Figure 3. Principal component analysis

The similar behaviour of identified genotypes as being tolerant to drought stress because of ability of translocation assimilate from straw wheat is strengthened by the reaction of Izvor variety which is known as drought tolerant cultivar due to its high osmotic adjustment. Also, Andrada variety has highlighted a good tolerance to drought stress, but in its case this physiological trait can be the result of a complex of minor genes. Even if some identified genotypes did not excel in normal conditions, these could be used as a valuable germoplasm sources for future breeding to drought stress conditions.

CONCLUSIONS

Analysing the reaction of winter wheat genotypes to simulated artificial drought stress conditions obtained by desiccant applications on the main elements of productivity (grain weight/spike, grain number/spike, TKW and grain yield) we noticed that grain number/spike was also very significantly influenced by treatments (despite the fact that at 14 days after anthesis this character was already established), while the TKW and the grain weight/spike suffered major changes with important effects on grain yield.

Using this method of drought tolerance simulation allowed identification of valuable biological material: T. 55-01, T. 99-11; T. 124-11; T. 15-12; T. 57-12 and T. 78-12 capable of better remobilization of stored assimilates.

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