Volume 15(2), 85- 91, 2011
JOURNAL of Horticulture, Forestry and Biotechnology

Effects of osmotic stress in the germination stage of some barley (*Hordeum vulgare*) genotypes

Velicevici Giancarla*[1], Sumalan R.[1], Madosa E. [1], Ciulca S[1].,Ciulca Adriana[1], Bitea Nicoleta[1]

[1]Banat University of Agricultural Sciences and Veterinary Medicine, Faculty of Horticulture and Forestry, Calea Aradului, 119, 300645, Timisoara, Romania

*Corresponding author: giancarlavely2000@yahoo.com

Abstract Osmotic stress caused by drought (water stress) or salinity is a major problem facing agriculturae and the wildlife allower the world and especially in the developing countries. These environmental factor affect plants at various stages of growth and the development from germination, seedling stage, vegetative growth and productivity. In this study were investigated 19 romanian and foreign genotypes of barley (Hordeum vulgare) regarding our germinative capacity in stress conditions. The osmotic stress were induced by PEG 6000 solution with the osmotic potential (-2.72 Bars, -4.48 Bars -7.35 Bars) using method suggested by (10) and replicated three time, at a temperature of 20 ^{0}C.
Therefore, drought induced by using physiological germination solutions with a different osmotic pressure levels directly influence the germination of the seeding material. From this parameters stand point the superior genotypes were: Andrei, Dina, DH 261-22.

Key words

barley, germination., drought

Barley (*Hordeum vulgare* L.) is the major cereal in many dry areas of the Middle East, North Africa and West Asia (1.2)and is vital for the livelihood of many poor farmers, with suggestions that it could replace wheat as the dominant crop, due to its tolerance to drought (4). It is a major food source in many N. African countries, although in Pakistan it is mainly grown for grain and straw for small ruminants during winter (7).Drought is the main environmental constraint, which occurs in many parts of the world every year, often having devastating effects on crop productivity. Hence, improved tolerance to drought has been a goal in crop improvement programs since, the dawn of agriculture (9). Drought tolerance is not a simple response, but is mostly conditioned by many component responses, which interact and may different for crops, in relation to types, intensity and duration of water deficit. Moreover, most agronomical characters are expressed differently in normal and stress conditions and are known to be affected by environmental factors. Therefore, selection based on the phenotype would be difficult for such traits (8).
Seed germination is usually a critical stage determining the state of the crop, witch in turn determines the yield. The coursse of the seedling emergence depends on the interaction between agroecological condition and seed vigor. Wather deficiency in soil causes delayed and reduced germination on seed, (5) unegual seedling emergence, varied numbers of plants per unit area and decreased seed yield.
Germination is a one trait that has been found to greatly vary among populations (reviewed by Baskin & Baskin, 1998). Selection is expected to favour appropriate responses to local environmental cues that synchronize germination with periods that are optimal for seedling survival. Such a selection may result in site-specific adaptation of germination traits.

Material and Methods

Experiment were conducted in the laboratories of the Faculty of Horticulture and Forestry Timisoara, Departament of the Genetic Enginering in agriculture. The biological material used in this study was represented by a collection of 19 genotypes of romanian and foreign whinter barley. The seed was obtained from Agriculture Research Station Fundulea.
Seed were initially treated with 1,5% sodium hipoclorite for 15 min. residual chlorine was eliminated by throught washing of the seeds with distilled water. 100 seeds were then germinated on filter parer in Petri-dishes, in an incubator at 22+2^{o}C. The experiment was conducted under normal (0 Bars) and drought stress (-2.72 Bars,-4.48 Bars, and – 7.35Bars) conditions created with the help of Polyethylene- ghlychol (PEG6000) by the method suggested by Michael and Kaufman (1973) and replicated three time. Five ml of distilled wather or PEG6000 solutions was added to each Petri-dish under normal and drought stress conditions, respectively , after 2 days, to compensate the losses due to evaporation. The emergence of 2 mm of radical and plumule was taken as the criterion for germination. The data for germination percentage was recorded on 3, 5 and 10 days after sowing.

Results and Discussions

The peculiarities of barley caryopsis germination is of particular importance for the success of crops, because the germination period may overlap or be followed by environmental conditions characterized by lack of moisture.

It is known that the dehydration of the cell, which manifests itself through the extraction of the water from cells, called plasmolysis condition, occurs when the external solution is more concentrated than the internal solution. Plasmolysis occurs in plants during drought conditions or when soil solution becomes hypertonic (chemical pollutants, excess salt, etc.). (11)

In the laboratory, using a hypertonic solution to induce water stress (with polyethylene ghlycol, sucrose), it is possible to test the drought tolerance of plants from seed germination stage.

The capacity of the caryopsis to germinate clearly illustrates the adverse conditions of hydric deficit. The drought simulated by different graduations of the osmotic value in the germination mediums affected the germination capacity of the caryopsis for all barley genotypes studied. The higher was the osmotic value of the solution used as a medium for germination, the more reduced was the germination capacity.

In table 1 is recorded the germination rate of each genotype normally hydrated, these results being considered the starting point of highlighting the influence of the germination medium on the germination of seed and germination rates of the three osmotic pressures. These results were recorded after three days.

Germination ranged from 9% to 32.00%. Compared with the control variety Dana, genotypes with higher germination were: Andrew (32%), Compact (27%), Dina and the Royal (25%), the results recorded being statistically insured. With a lower germination compared to the control line Dana were Tas genotypes and double haploid genotype DH254-10 (9%), the results recorded for these genotypes being significantly negative in statistical terms.

Germination recorded for the genotypes under hydric stress with PEG 6000 at a pressure of (-2.72 Bars) ranged between 4% and 20%. Genotypes that recorded higher results than the control genotype were : Andrew (20%), Dina (20%), Lyric (16%), double haploid line DH 261-22 (16%), Compact (13%), Sunrise (12%) , Djerbel (12%), the results recorded by them being statistically insured. Genotypes with a lower germination than the control genotypes` germination were Tas and DH 254-10 (4%).

The percentages of germination of barley genotypes obtained in a PEG6000 solution (corresponding to an osmotic pressure of -4.48 Bars.) are presented in Table 1.1. It may be noted that at this concentration of the solution, the germination decreased significantly ranging between 3 and 16%. Once again Andrew genotypes, Dina and DH 261-22 double haploid line scored very significantly superior to control. Genotypes with a lower germination capacity compared to the control were Palaisant. DH 254-10 (2%), but the results don`t have statistical significance.

The increase of osmotic pressure (-7.35 Bars) determined the germination percentage to decrease even further between 1% and 7%. And in this case genotypes Andrei and Dina (7%) recorded a higher germination percentage than the control Dana the results being very significant positive.

In Table 2 are recorded germination percentages after 7 days. The variant hydrated with distilled water had a germination percentage between (83% and 100%). With a 100% germination rate was observed Adi variety, but the result is not statistically significant. Plaisant and Regal were inferior to the control Dana (Plaisant (83%) and Regal(89%)) the results scored by these two being significantly negative and significantly distinct from the controls.

The variant that was the subject of water stress with PEG 6000 (-2.72 Bars), germination percentages were between 36% and 94%. Genotypes with a higher germination percentage compared to the control Dana were: DH 261-22 (94%), Orizont (86%), Secura (84%), Adi (81%), Andrei (76%) the results recorded being distinct significant positive. With a lower germination percentage was observed genotype Tas (36%), this result being significantly negative compared to the controls.

As the osmotic pressure increases one may notice a decrease in the percentage of germination, so at a pressure of (-4.48 Bars), it was between 14% and 88%. Once again the genotypes that were superior to the control genotype were DH 261-22 (83%), Adi (60%), Compact (65%), Regal (52%), Andrei (50%), the results being very significantly positive. Inferior to the control was Tas (14%) very significantly negative. The more intense the fluid deficit was (-7.35 Bars), a more drastic decrease in the percentage of germination may be noticed, being ranged between 10% and 46%. The genotypes superior to the control were Djerbel (46%), Adi (44%), Compact (42%), Orizont (40%), double haploid line DH 260-18 (42%), Andrei (34%), the results having statistical insurance. Also in this case genotype Tas recorded the lowest percentage of germination.

In Table 3 are presented the data on the percentage of germination recorded at 10 days. The variant hydrated with distilled water had a germination percentage between 83% and 100%. Genotypes which registered the lowest percentage of germination were Plaisant (83%) and DH line 254-10 (95%) results registered by this were statistical significance.The variant with (-2.72%) PEG 6000 the percentage of germination varied between 74% and 96%. Very significant negative results from the control genotypes

were recorded at the Plaisant, Madalin, Dina and Sunrise.

In case of an increased osmotic pressure the germination percentage was between 40% and 85%. The genotypes that have registered very significantly negative differences from control were Mădălin, Dina Sunrise, Djerbel, Adi. In this case DH260-12, DH254-10 surpassed the control but the differences don`t have statistical insurance.

Germination percentage recorded in the osmotic pressure (-7.35 Bars) was between 20% and 64%. In this case there were recorded very significant positive differences compared with the control at the following genotypes: Andrei, Regal, Compact, DH 260-18, Orizont. Distinctly significant negative differences were recorded for the variety Tas, DH 254-10 line.

Therefore, drought induced by using physiological germination solutions with a different osmotic pressure levels directly influence the germination of the seeding material. From this parameters stand point the superior genotypes were: Andrei, Dina, DH 261-22.

Analising the data from table 4 we can observe the falowins : concentration PEG influence on germination seed of barley, a high drought tolerance correlated with high values of germination percentage was presented by cultivars:Regal,DH260-18,Dana, DH 261-22, Orizont and low values of germination confirms a high drought sensibility, in this case is genotypes Plaisant, Tas, Secura.

Table1

Results regarding germination percentage registered from 3 days

		V0				V1				V2				V3			
No. Crt.	Genptypes	meen germinative %	%	diference	signif.	meen germinative%	%	diference	signif.	meen germinative %	%	diference	signif.	meen germinative %	%	diference	signif.
1	DANA	17,000	100,000	0,000		6	100	0		4	100	0		2	100	0	
2	**ORIZONT**	14,333	84,314	-2,667		10	166,667	4		7	175	3		5	250	3	*
3	**PRECOCE**	14,333	84,314	-2,667		7	116,667	1		5	125	1		3	150	1	
4	**ADI**	12,000	70,588	-5,000		5	833,333	-1		4	100	0		2	100	0	
5	**MADALIN**	13,000	76,471	-4,000		5	833,333	-1		3	75	-1		2	100	0	
6	**ANDREI**	32,000	188,235	15,000	***	20	333,333	14	***	15	375	11	***	7	350	5	***
7	**REGAL**	25,000	147,059	8,000	*	10	166,667	4		9	225	5		6	300	4	**
8	**COMPACT**	27,000	158,824	10,000	*	13	216,667	7	**	7	175	3		4	200	2	
9	**DJERBEL**	17,000	100,000	0,000		12	200	6	*	9	225	5		5	250	3	*
10	**LYRIC**	20,000	117,647	3,000		16	266,667	10	***	8	200	4		5	250	3	*
11	**PLAISANT**	13,000	76,471	-4,000		5	833,333	-1		2	50	-2		1	50	-1	
12	**TAS**	9,000	52,941	-8,000	0	4	666,667	-2		3	75	-1		1	50	-1	
13	**SECURA**	10,000	58,824	-7,000		6	100	0		2	50	-2		1	50	-1	
14	**DINA**	25,000	147,059	8,000	0	20	333,333	14	***	16	400	12	***	7	350	5	***
15	**SUNRISE**	24,000	141,176	7,000		12	200	6	*	7	175	3		3	150	1	
16	**DH 254-10**	9	5,294,118	-8	0	4	666,667	-2		2	50	-2		1	50	-1	
17	**DH260-18**	12	7,058,824	-5		9	150	3		5	125	1		2	100	0	
18	**DH 260-12**	15	8,823,529	-2		8	133,333	2		6	150	2		2	100	0	
19	**DH 261 22**	20	1,176,471	3		16	266,667	10	***	12	300	8	***	4	200	2	
			DL 5%	7.492			DL 5%	4.791			DL 5%	5.458			DL 5%	2.474	
			DL1%	10.038			DL1%	6.42			DL1%	7.314			DL1%	3.315	
			DL 0.1%	13.249			DL 0.1%	8.474			DL 0.1%	9.653			DL 0.1%	4.375	

Table2

Percent of germination registered at 7 days

		V0				V1				V2				V3			
No. Crt.	Genptypes	meen germinative %	%	diference	signif.	meen germinative %	%	diference	signif.	meen germinative %	%	diference	signif.	meen germinative %	%	diference	signif.
1	DANA	96	100	0		53	100	0		36	100,000	0		26	100	0	
2	**ORIZONT**	99	103,125	3		86	162,2642	33	***	45	125,000	9	*	40	153,846	14	***
3	**PRECOCE**	98	102,0833	2		68	128,3019	15	*	38	105,556	2		15	57,6923	-11	000
4	**ADI**	100	104,1667	4		81	152,8302	28	***	60	166,667	24	***	44	169,230	18	***
5	**MADALIN**	95	98,95833	-1		69	130,1887	16	*	44	122,222	8		35	134,615	9	**
6	**ANDREI**	97	101,0417	1		76	143,3962	23	**	50	138,889	14	***	34	130,769	8	**
7	**REGAL**	89	92,70833	-7	00	63	118,8679	10		52	144,444	16	***	30	115,384	4	
8	**COMPACT**	92	95,83333	-4		71	133,9623	18	*	65	180,556	29	***	42	161,538	16	***
9	**DJERBEL**	98	102,0833	2		66	124,5283	13		56	155,556	20	***	46	176,923	20	***
10	**LYRIC**	92	95,83333	-4		68	128,3019	15	*	35	97,222	-1		24	92,3076	-2	
11	**PLAISANT**	83	86,45833	-13	000	44	83,01887	-9		28	77,778	-8	00	23	88,4615	-3	
12	**TAS**	96	100	0		36	67,92453	-17	0	14	38,889	-22	000	10	38,4615	-16	000
13	**SECURA**	96	100	0		84	158,4906	31	***	33	91,667	-3		23	88,4615	-3	
14	**DINA**	96	100	0		74	139,6226	21	***	30	83,333	-6	0	18	69,2307	-8	00
15	**SUNRISE**	97	101,0417	1		52	98,11321	-1		47	130,556	11	***	27	103,846	1	
16	**DH 254-10**	96	100	0		89	167,9245	36	***	80	222,222	44	***	20	76,9230	-6	0
17	**DH260-18**	94	97,91667	-2		82	154,717	29	***	72	200,000	36	***	42	161,538	16	***
18	**DH 260-12**	98	102,0833	2		92	173,5849	39	***	88	244,444	52	***	35	134,615	9	**
19	**DH 261 22**	98	102,0833	2		94	177,3585	41	***	83	230,556	47	***	25	96,1538	-1	
			DL 5%	4.889			DL 5%	14.025			DL 5%	5.646			DL 5%	5.557	
			DL1%	6.551			DL1%	18.793			DL1%	7.565			DL1%	7.446	
			DL 0.1%	8.646			DL 0.1%	24.803			DL 0.1%	9.984			DL 0.1%	9.827	

Table 3

Percent of germination registered at 10 days

		V0				**V1**				**V2**				**V3**			
No. Crt.	Genptypes	meen germinative %	%	diference	signif.	meen germinative %	%	diference	signif.	meen germinative %	%	diference	signif.	meen germinative %	%	diference	signif.
1	DANA	**100**	100	0		**96**	100,000	0		**85**	100,000	0		**39**	100,000	0	
2	**ORIZONT**	**99**	99	-1		**86**	89,583	-10	0	**80**	94,118	-5		**65**	166,667	26	***
3	**PRECOCE**	**100**	100	0		**96**	100,000	0		**79**	92,941	-6		**42**	107,692	3	
4	**ADI**	**99**	99	-1		**88**	91,667	-8		**66**	77,647	-19	000	**60**	153,846	21	***
5	**MADALIN**	**99**	99	-1		**79**	82,292	-17	000	**69**	81,176	-16	000	**46**	117,949	7	*
6	**ANDREI**	**98**	98	-2		**90**	93,403	-6		**82**	96,471	-3		**52**	133,333	13	***
7	**REGAL**	**99**	99	-1		**97**	100,694	1		**83**	97,647	-2		**53**	135,897	14	***
8	**COMPACT**	**96**	96	-4		**91**	94,792	-5		**80**	94,118	-5		**55**	141,026	16	***
9	**DJERBEL**	**100**	100	0		**81**	84,375	-15	00	**58**	68,235	-27	000	**50**	128,205	11	**
10	**LYRIC**	**99**	99	-1		**97**	101,042	1		**65**	76,471	-20	000	**37**	94,872	-2	
11	**PLAISANT**	**83**	83	-17	000	**47**	48,958	-49	000	**40**	47,059	-45	000	**29**	74,359	-10	00
12	**TAS**	**99**	99	-1		**95**	98,958	-1		**49**	57,647	-36	000	**20**	51,282	-19	000
13	**SECURA**	**99**	99	-1		**82**	85,417	-14	00	**78**	91,765	-7		**38**	97,436	-1	
14	**DINA**	**98**	98	-2		**74**	77,083	-22	000	**54**	63,529	-31	000	**42**	107,692	3	
15	**SUNRISE**	**97**	97	-3		**77**	80,208	-19	000	**65**	76,471	-20	000	**33**	84,615	-6	
16	**DH 254-10**	**95**	95	-5	0	**93**	96,875	-3		**88**	103,52	3		**24**	61,538	-15	000
17	**DH260-18**	**100**	100	0		**96**	100,000	0		**84**	98,8235	-1		**64**	163,248	24	***
18	**DH 260-12**	**99**	99	-1		**94**	97,917	-2		**88**	103,529	3		**32**	82,051	-7	0
19	**DH 261 22**	**99**	99	-1		**94**	97,917	-2		**85**	100	0		**50**	128,205	11	**
			DL 5%	4.099			DL 5%	9.477			DL 5%	8.528			DL 5%	6.608	
			DL1%	5.492			DL1%	12.698			DL1%	11.426			DL1%	8.854	
			DL 0.1%	7.249			DL 0.1%	16.760			DL 0.1%	15.081			DL 0.1%	11.686	

Table 4

Concentration PEG influence on germination seed of barley

Nr. Crt.	Genotypes	V0		V1		V2		V3		Suma ranguri
		Meen	Rang	Meen	Rang	Meen	Rang	Meen	Rang	
1	**Dana**	100	1,5	96	3,5	85	3,5	39	12	20,5
2	**Orizont**	99	3,5	86	12	80	8,5	65	1	25
3	**Precoce**	100	1,5	96	3,5	79	10	42	10,5	25,5
4	**Adi**	99	3,5	88	11	66	13	60	3	30,5
5	**Mădălin**	99	3,5	79	15	69	12	46	9	39,5
6	**Andrei**	98	5,5	90	10	82	7	52	6	28,5
7	**Regal**	99	3,5	97	1,5	83	6	53	5	16
8	**Compact**	96	8	91	9	80	8,5	55	4	29,5
9	**Djerbel**	100	1,5	81	14	58	16	50	7,5	39
10	**Lyric**	99	3,5	97	1,5	65	14,5	37	14	33,5
11	**Plaisant**	83	10	47	18	40	19	29	17	64
12	**Tas**	99	3,5	95	5	49	18	20	19	45,5
13	**Secura**	99	3,5	82	13	78	11	38	13	40,5
14	**Dina**	98	5,5	74	17	54	17	42	10,5	50
15	**Sunrise**	97	7	77	16	65	14,5	33	15	52,5
16	**DH 254/10**	95	9	93	8	88	1,5	24	18	36,5
17	**DH 260/18**	100	1,5	96	3,5	84	5	64	2	12
18	**DH 260/12**	99	3,5	94	6,5	88	1,5	32	16	27,5
19	**DH 261/22**	99	3,5	94	6,5	85	3,5	50	7,5	21

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Acknowledgement

This work was published during the project "POSTDOCTORAL SCHOOL OF AGRICULTURE AND VETERINARY MEDICINE", POSDRU/89/1.5/S/62371, co-financed by the European Social Fund through the Sectorial Operational Programme for the Human Resources Development 2007-2013.