Differential Genotypic Responses of String Wheat Early Seedling Growth to Limited Moisture Conditions

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Keywords: *Triticum* sp. — Durum wheat — Water stress — Selection.

Summary

The objective of this study was to measure the genotypic response of spring wheat seedling growth in a range of osmotic media and to determine which genotype could be identified as drought tolerant. Six durum wheat cultivars were subjected to moisture stress using polyethylene glycol PEG-9000. Aqueous solutions of 0, -3, -6 and -9 bars were prepared. For each cultivar 20 seeds were germinated in these solutions in a growth chamber. After 2 weeks, number of roots, leaf number, coleoptile length, seedling height, root length, first and second leaf length, and dry matter weight were measured. All traits measured were significantly influenced by water stress. The water stress treatments of -6 and -9 bars gave lower rates of seedling growth than the 0 and -3 bars treatments. The results suggest that good seedling vigor under water stress condition is a useful selection criterion. An ideotype for a drought tolerant wheat genotype should have good seedling vigor.

Résumé

L'objectif de cette étude était de mesurer la réponse génotypique des plantules de blé de printemps dans une gamme de milieu osmotique et de déterminer les génotypes qui pourraient être identifiés comme tolérants à la sécheresse. Six cultivars de blé dur ont été soumis au stress hydrique en utilisant le polyéthylène glycol PEG-9000. Des solutions aqueuses de 0, -3, -6 et -9 bars ont été preparées. Pour chaque cultivar, 20 semences ont germé dans ces solutions dans une chambre de culture. Après 2 semaines, le nombre de racines, le nombre de feuilles, la longueur du coléoptile, la hauteur des plantules, la longueur des racines, la longueur de la première et de la deuxième feuille et le poids de la matière sèche ont été mesurés. Tous les caractères mesurés ont été significativement affectés par le stress hydrique. Les traitements de -6 et -9 bars ont été associés avec des taux de croissance faibles par rapport à ceux observés chez les traitements de 0 et -3 bars. Les résultats suggèrent qu'une bonne vigueur des plantules sous des conditions stressées est un critère de sélection utile. Un idéotype de blé tolérant la sécheresse devrait avoir une bonne vigueur au stade jeune plantule.

Introduction

Drought limits crop production in many semi-arid areas of the world. In these areas plants are exposed to many environmental stresses including abnormal temperatures, poor nutrient and physical soil conditions, weeds, diseases, and insect pests. However, water stress which is both extensive and unpredictable reduces plant growth and yield more than the other stresses combined (4, 8). Yield instability in the drought prone areas requires the genetic improvement of cultivars adapted to these harsh environmental stresses.

The success of a spring wheat crop is initially dependent on good seedling establishment and vigor. Soil moisture at planting time is unreliable and often inadequate in arid and semi-arid areas. An important contribution to the successful spring wheat production would be the selection of cultivars germinating and producing vigorous seedlings in soil, with limited available moisture.

Seed germination and subsequent seedling growth have been used to screen for drought tolerance in many crop species. The procedures consist of testing seed germination and plant growth under induced water deficit (1, 2, 3, 7, 10, 12, 13). These techniques provide rapid tools for identifying physiological characteristics in crop plants that are reliable indicators for selecting genotypes that are drought tolerant. However, previous studies did not indicate which parameter to use as a selection criterion for drought tolerance. The objective of this study was to determine which parameters are effective criteria to select for drought tolerance in spring wheat and in practical terms to develop cultivars with good germination and seedling establishment under limited available moisture.

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Material and methods

Six durum wheat *Triticum durum* DESF. cultivars originating from various areas were used in this experiment. These cultivars are Ashu (Africa) 5, Indo (India) 1, Peru 1, Peru 4, Latino and Rouma. They originate respectively from Japan, India, Peru, Peru and Italy. They were provided by the «National Institute of Agrobiological Resources Genebank» in Japan.

Polyethylene glycol PEG-9000 was the moisture stress inducing medium. Aqueous solutions of 0, -3, -6 and -9 bars were obtained by dissolving 0, 115, 170 and 225 g of extra pure PEG-9000 in 1000 ml of distilled water, respectively. The quantity of PEG-9000 for the required osmotic potentials was determined from published curves (9) and these values were compared with those obtained by the freezing point method (Osmometer model OM 801, Vogel). Adjustments in the dilution of PEG-9000 were made until three replicated readings from the osmometer corresponded exactly to the desired water potentials.

The seeds were subjected to uniform seed sterilisation and then soaked in a 70% ethanol solution for 30 minutes and resoaked for the same time in a 1% sodium hypochlorite solution. Seeds were then put in a permeable white cloth and allowed to dry in a ventilated refrigerator at 10°C until the start of the experiment.

For each cultivar 20 seeds were germinated in the 4 aqueous solutions. Seeds were placed on two layers of Whatman no. 2 filter paper in 20 mm x 140 mm glass petri dishes and 10 ml of solution was added. The four replicates of each treatment (a total of 96 petri dishes) from a split-plot design were placed at random in a growth chamber for 2 weeks at a day/night light regime of 10/14h, a constant temperature of 20°C and 65% relative humidity.

After 2 weeks days the petri dishes were removed from the growth chamber and the following measurements were made on five seedlings from each petri dish: (1) number of roots, (2) number of leaves, (3) coleoptile length, seedling height, (5) root length, (6) first and second leaf length (cm). These five seedlings were then oven dried at 80°C and after 72th total dry matter weight was determined.

The experimental design was a split-plot with four replications. The main factor was water potential treatment and the sub-factor were the wheat cultivars randomly arranged within each main factor. The data obtained for each measured trait were subjected to analysis of variance. The comparison of the treatment means was done by the least significant value (LSD). Only significances at the 1% and 5% levels were considered.

Results and Discussion

The effects of water stress on the parameters measured are presented in Tables 1, 2 and 3. The analysis of variance of each separate data revealed that all traits were significantly ($P<0.01$) influenced by water stress. Significantly lower rates of seedling growth were observed with increasing levels of water stress. These results indicate that in wheat water potential is a parameter vital to early seedling growth and development. The data in Tables 1, 2, 3 and 4 illustrate that when wheat seeds are germinated into a dry soil a reduced seedling vigor is expected to occur. It was shown that plant water potential influences virtually all morphological, physiological, and metabolic processes and the rate of injury depends on the water stress level (6). Similar findings were reported by (2) and (3). It is worth mentioning that small differences in seedling vigor were observed between the 0 and -3 bars water potential treatments indicating that a slight reduction in the moisture stress level is not detrimental to optimal seedling growth whereas the -6 and -9 bars gave significantly lower rates of seedling growth than 0 and -3 bars. Since plants are highly integrated organisms, when stress is not severe, it disturbs some primary processes in the system. When stress increases gradually, the processes most sensitive to stress are normally altered first, and such alterations, in turn, may lead to many secondary and tertiary changes.

Genotypic differences were significant for all parameters except for root length and number of leaves per seedling. Differences among cultivars in their response to water stress may be attributed to differences in structural or physiological characteristics that maintain a relatively high plant water potential such as the capability to osmoregulate and the maintenance of membrane integrity.

Under high stress conditions (-6 and -9 bars) the differences in seedling vigor among cultivars were large while under low stress conditions (0 and -3 bars) these differences were small. These results suggest that seedling vigor under high stress conditions can be successfully used in the identification of drought tolerant wheat genotypes. The data in Tables 1, 2, 3 and 4 provide evidence of adaptation reactions that may be of value to the plant breeder in selecting a drought tolerant wheat cultivar based on seedling growth performance under various water stress levels.

There was significant genotype x moisture stress interaction effects on all traits studied, except for number of leaves per seedling. In fact, the reduction of the characters measured due to decreasing water potential was severe for some genotypes, moderate for others and low for some cultivars (Tables 1, 2 and 3). These results indicate that cultivars differed in their sensitivities to water stress. Significant differences in seedling growth among genotypes and their interaction with moisture treatments have one implication: under the conditions of this experiment seedling growth is a viable selection criterion for drought tolerance in durum wheat. Genotypes that maintain high seedling vigor are probably characterized by an adequate osmotic adjustment and hence obtain a low water pressure favorable to higher seed yield and dry matter production (2, 3). Results in this experiment show that it is possible to identify such genotypes based on the measurement of traits associated with seedling vigor.

The ranking of genotypes based on their seedling growth performance across the traits measured under the various water potential treatments was not consistent. This is

Table 1
Mean coleoptible and root length (cm) and number of roots per seedling of six durum wheat genotypes germinated at four water potential values

Genotype	Water potential (bars)			
	0	-3	-6	-9
Ashu (Africa) 5	2.1[1]	2.0	2.1	2.0
	11.7[2]	12.1	10.5	9.1
	5.5[3]	5.6	5.4	5.7
Indo (India) 1	1.9	2.0	1.8	1.7
	12.2	12.2	9.3	7.9
	7.3	7.2	7.1	6.7
Peru 1	1.9	2.0	1.2	2.0
	15.5	12.8	9.5	5.9
	7.4	7.0	6.5	5.5
Peru 4	2.3	2.2	2.1	1.9
	10.7	12.4	11.4	8.5
	7.7	7.1	7.4	6.8
Latino	2.1	2.4	2.2	2.2
	11.2	13.3	10.5	9.9
	6.7	6.5	6.8	6.4
Rouma	1.5	1.6	1.5	1.4
	14.1	13.7	9.5	7.6
	6.2	5.4	5.6	5.7

[1] Coleoptile length. - [2] Root length. - [3] Number of roots per seedling.
LSD (0.05) for comparing any pair of means = 0.1, 0.8 and 0.2 respectively for coleoptile length, root length and number of roots per seedling.

Table 2
Mean number of leaves, first and second leaf length (cm) of six durum wheat genotypes germinated at four water potential values

Genotype	Water potential (bars)			
	0	-3	-6	-9
Ashu (Africa) 5	2.0[1]	1.6	1.7	1.0
	7.2[2]	5.9	5.5	4.3
	3.0[3]	2.1	1.1	-
Indo (India) 1	2.0	1.7	1.3	1.0
	6.9	7.3	5.2	3.7
	3.8	1.4	1.5	-
Peru 1	2.0	1.7	1.3	1.1
	8.2	7.0	4.7	2.2
	5.8	3.0	1.1	-
Peru 4	2.0	2.0	1.5	1.0
	6.6	6.8	4.7	3.9
	3.2	1.9	0.8	-
Latino	2.1	1.9	1.5	1.2
	7.2	7.4	6.2	4.9
	4.3	2.4	1.7	-
Rouma	2.0	1.9	1.3	1.1
	6.2	6.2	4.4	3.9
	5.2	3.1	1.9	-

[1] Number of leaves. - [2] First leaf length. - [3] Second leaf length.
-: Second leaf was absent.
LSD (0.05) for comparing any pairs of means = 0.15 for number of leaves and 0.4 for first leaf length. LSD value for second leaf length was impossible to compute because of missing values at the -9 bars treatment.

because genotypes displayed genotype x environment interaction that resulted in differential responses. The different relationships among the traits measured within each genotype contributed to the failure of genotypes to perform similarly in their response to water stress levels across all traits studied. These results indicate that it is difficult to identify drought tolerant wheat genotypes based on the evaluation of many traits related to seedling vigor. To remedy this, the response of the six genotypes to water

Table 3
Mean seedling height (cm) and dry matter weight (g) of six durum wheat genotypes germinated at four water potential values

Genotype	Water potential (bars)			
	0	-3	-6	-9
Ashu (Africa) 5	9.9[1]	8.1	7.6	6.2
	0.109[2]	0.109	0.095	0.079
Indo (India) 1	9.4	9.6	7.0	5.4
	0.117	0.119	0.106	0.088
Peru 1	10.6	9.4	7.1	3.8
	0.136	0.117	0.065	0.044
Peru 4	9.1	9.4	6.9	5.9
	0.124	0.121	0.098	0.083
Latino	10.5	9.8	8.4	7.0
	0.130	0.141	0.129	0.103
Rouma	8.3	8.1	6.0	5.3
	0.128	0.122	0.092	0.083

[1] Seedling height. - [2] Seedling dry weight.
LSD (0.05) for comparing any pair of means = 0.5 and 0.06 respectively for seedling height and seedling dry matter weight.

Table 4
Stress and drought indices of six durum wheat genotypes derived from the evaluation of many characters related to seedling vigor under optimum water supply (0 bar) and high water stress condition (-9 bars)

Character	Genotype[1]					
	Ashu	Indo	Peru 1	Peru 4	Latino	Rouma
Coleoptile length	0.95	0.89	1.05	0.83	1.05	0.93
Root length	0.78	0.65	0.38	0.79	0.88	0.54
First leaf length	0.60	0.54	0.27	0.59	0.68	0.63
Second leaf length	-	-	-	-	-	-
Seedling height	0.63	0.57	0.36	0.65	0.67	0.64
Number of roots	1.04	0.92	0.74	0.88	0.96	0.92
Dry matter weight	0.73	0.75	0.32	0.67	0.79	0.65
Drought index	0.79	0.72	0.52	0.74	0.84	0.72

[1] Ashu = Ashu (Africa 5), Indo = Indo (India) 1.
-: Indices were not possible to compute because the second leaf was absent at the -9 bars water potential treatment.

stress was assessed by a stress tolerance index (5) defined as the ratio of seedling growth in the -9 bars treatment to that in the 0 bars treatment. In the construction of a drought tolerance index the characters mesured, (except for number of leaves per seedling whose genotypic effects were not significant) were weighted equally important in their contribution to final seedling growth expression. Thus, the drought tolerance index of each genotype is the mean of the stress indices averaged over all characters (Table 4). These indices are very useful because they allow a direct comparison of genotypic response to water stress (5). Low indices indicate susceptibility to drought; conversely, high indices indicate tolerance. From the results in Table 4 it may be concluded that the cultivar Latino is the most drought tolerant genotype. The cultivars Ashu(Africa) 5, Indo (India) 1, Peru 4 and Rouma are moderately tolerant to drought. The cultivar Peru 1 with a drought index of 0.52 is the most drought susceptible cultivar in this population. The results of the drought index show a difference in the ability to cope with water stress of more than 60% between the best cultivar (Latino) and the poorest cultivar (Peru 1). This demonstrates wide variability for drought tolerance in this group of durum wheat lines. The variation observed among cultivars tested here supports the idea that wide variation exists among wheat lines for the character of drought tolerance at the seedling stage.

Conclusion

These results suggest that good seedling vigor under water stress condition is a useful selection criterion. An ideotype for a drought tolerant wheat genotype should have good seedling vigor. The ability of a seed to produce vigorous seedling under drought stress indicates that it has genetic potential for drought tolerance, at least at this stage in the life cycle. This does not necessarily indicate that a seedling started under water stress could continue under drought stress and that the plant could complete its life cycle. Tolerance to drought at germination and emergence is, however, a highly desirable trait. For this reason, use of early seedling growth as first indicator of drought tolerance seems valid. This experiment dealt only with drought tolerance at the critical first stages of the life cycle which may or may not be associated with the ability to cope a later stages. A more ambitious program seems appropriate, which would include screening all available material, not only at seedling, but also at all the other stages of the life cycle. This would determine the evolution of performance among stages, if any exists, and would be more useful in a breeding program for the development of higher levels of drought tolerance in wheat at all stages of development. It would be preferable to use seedlings in drought stress selection techniques. Several workers have indicated that drought response of seedlings was reasonably well correlated with drought response of mature plants (11, 12).

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