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BRIEF COMMUNICATION

Growth, abscisic acid content, and carbon isotope composition in wheat cultivars grown under different soil moisture

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Abstract

Changes in dry matter accumulation and allocation, abscisic acid content and carbon isotope composition of three wheat cultivars from dry, middle and wet climate regions were recorded at full maturity after exposure to different watering regimes (100, 50 and 25 % field capacity). Compared with the wet climate cultivar, the dry climate cultivar showed lower stem height, total leaf area, total dry biomass and total grain dry mass, and higher root/shoot ratio, abscisic acid content and carbon isotope composition under all watering regimes. Both water-limited treatments significantly reduced leaf growth and increased dry matter allocation into the roots leading to a significant raise of root/shoot ratio in all cultivars tested. In addition, drought affected morphological and physiological properties more in the dry climate cultivar than in the wet climate cultivar.

Additional key words: dry matter accumulation and allocation, *Triticum aestivum*, water use efficiency.

Survival strategies in response to water deficits in many plants have been extensively studied (Socias *et al*. 1997, Heschel and Hausmann 2001, Nayyar and Walia 2003, Pospíšilová and Baťková 2004, Akhter *et al*. 2005). Some of the adaptive responses to drought are due to physiological and morphological changes, such as changes in plant structure, dry matter accumulation, tissue osmotic potential and stomatal conductance (reviewed by Blum 1997). Moreover, abscisic acid (ABA), a well-known stress-inducible plant hormone and growth inhibitor, has long been studied as a potential mediator for induction of drought tolerance in many plants (Sharp 2002, Pandey *et al.* 2003, Purty *et al*. 2005). It has been shown to be involved in drought tolerance, both from the use of applied exogenous ABA to intact plants (Xu *et al*. 2002, Wang *et al*. 2003, Li *et al*. 2004, Yin *et al*. 2004), and from measurement of the endogenous ABA content under water-limited conditions (Li and Wang 2003, Zhang *et al*. 2004). The relationship between drought tolerance and ABA content was also reported in wheat (Ali *et al*. 1999, Nayyar and Kaushal 2002, Nayyar and Walia 2003, Trapeznikov *et al*. 2003). However, for wheat cultivars from contrasting climate regions, their differences in physiological and morphological responses to water deficits were studied relatively little.

For long time there has been a considerable effort to elucidate the degree and nature of the genetic control over water use efficiency (WUE) in plants. WUE is defined either as the ratio of dry matter accumulation to water consumption over a season or as the ratio of net photosynthesis to transpiration over a period of seconds or minutes. Much attention has been devoted to the study of carbon isotope composition ($\delta^{13}C$) of plant tissues and $\delta^{13}C$ because a strong correlation is found between $\delta^{13}C$ and WUE (Farquhar *et al*. 1989). The carbon isotope ratio of plant tissue provides an integrated measurement of internal and external affections of photosynthetic gas exchange over the time when the carbon was fixed (Anderson *et al*. 1996, Brodribb and Hill 1998, Li 2000).

In this study, wheat, which is widely distributed from the dry to wet climate regions and exhibits large genetic variation, was employed as a model species to assess differences in dry matter accumulation and their allocation, ABA content and $\delta^{13}C$ of three cultivars selected after exposure to different watering regimes (100, 50 and 25 % field capacity). These differences between

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Abbreviations: ABA - abscisic acid content; GM - total grain dry mass; HT - stem height; LA - total leaf area; RM - root dry mass; RS - root/shoot ratio; SLA - specific leaf area; TB - total dry biomass; WUE - water use efficiency; $\delta^{13}C$ - carbon isotope composition.

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the cultivars in drought responses may be used as criteria for genotype selection in different management regimes.

Three cultivars of wheat (*Trititcum aestivum* L.) from contrasting habitats (No. 01 from dry climate region Lasa, 30º10'N, 91º30'E, annual rainfall 320 mm, No. 02 from middle climate region Kaifeng, 35º38'N, 114º52'E, annual rainfall 626 mm and No. 03 from wet climate region Nanjing, 31º20'N, 118º18'E, annual rainfall 1106 mm) were chosen for the study. Seeds were sown on wet tissue paper in Petri dishes. After seed germination, the seedlings were transferred into small plastic pots (180 cm^3) and grown for about one month. Ninety healthy seedlings of uniform height were chosen from each cultivar, and each seedling was transferred to a 2-dm^3 pot. A commercial peat-sand mixture (containing 80 % peat and 20 % sand) was used as growth medium in the pots. The plants were grown to full maturity in a naturally light greenhouse under the semi-controlled environment with a day temperature range of 12 - 31 °C and a night temperature ranged of 9 - 15 °C, and the relative humidity range of 35 - 85 %. A total of 5 g slow release fertilizer (7 % N, 5 % P and 26 % K) was added to each pot at the beginning of experiment.

A completely randomised design with two factors (cultivar and watering regime) was used. In well-watered treatment, individuals of each cultivar were watered to 100 % of field capacity by supplying an amount of water equal to transpiration losses every other day; in water-limited treatments, individuals of each cultivar were maintained at 50 % and 25 % of field capacity by watering every other day, respectively. 30 pots of each cultivar were used in each watering regime. Evaporation from the soil surface was prevented by enclosing the pots in plastic bags that were tied to the stems of the plants. Transpiration was measured gravimetrically by weighing all pots every other day. After six months, all individuals from each cultivar and treatment were at full maturity.

The flag leaves of ten replicates during mature period from each cultivar and treatment were used for carbon isotope analysis. The samples were oven-dried for 24 h at 80 ºC and homogenised by grinding in a ball mill. Stable carbon isotope abundance in combusted samples was measured with a mass spectometer (*Finnegan MAT Delta-E*, Waltham, USA). The overall precision in δ-values was better than 0.1 ‰ as determined from repeated samples. The abundance of ^{13}C in the sample was calculated as described by Li *et al.* (2000). The flag leaves of ten replicates during mature period from each cultivar and treatment were used for measurement of ABA content as described by Li *et al*. (2005).

All individual plants were harvested at full maturity after six months, and divided into leaves, stem, roots and grain. These biomass samples were dried to constant mass and weighed. Root/shoot ratio (RS, root dry mass divided by leaves, stem and grain dry mass) and specific leaf area (SLA, leaf area divided by leaf dry mass) were then calculated. Leaf area for each individual was determined by portable *Laser Area Meter* (*CI-203, CID Inc.*, Vancouver, BC, Canada).

Analyses of variance (*ANOVA*) of experimental data were used for testing the cultivar and treatment differences. Statistical analyses were done with the *SYSTAT* statistical software package.

Water-limited treatment (25 % of field capacity) decreased 34.44, 30.12 and 29.23 % in stem height (HT), 53.96, 49.17 and 48.04 % in total dry biomass (TB), 76.04, 69.59 and 67.13 % in total leaf area (LA), 27.45, 25.00 and 22.22 % in total grain dry mass (GM) in the dry, middle and wet climate cultivars, respectively (Table 1). Three cultivars of wheat differed significantly in HT, TB, LA and GM under all watering regimes. There was not significant cultivar difference in root dry

Table 1. Stem height (HT), total dry biomass (TB), root dry mass (RM), total leaf area (LA) and total grain dry mass (GM) in three wheat cultivars as affected by three watering regimes. Data are means ± SE of 10 replicates (*P*w - watering effect; *P*c - cultivar effect; $P_{W×C}$ - watering × cultivar interaction).

Soil moisture	Cultivar	HT [cm]	TB [g]	RM [g]	LA [dm^2]	GM [g]
100 %	No.01	78.4 ± 1.3	8.21 ± 0.25	2.69 ± 0.27	4.09 ± 0.15	0.51 ± 0.02
	No.02	81.0 ± 1.1	9.03 ± 0.31	2.63 ± 0.24	4.44 ± 0.17	0.60 ± 0.03
	No.03	84.5 ± 1.5	9.97 ± 0.39	2.53 ± 0.26	5.08 ± 0.21	0.72 ± 0.05
	P_C	< 0.001	< 0.001	ns	< 0.001	< 0.001
50 %	No.01	65.9 ± 0.7	5.26 ± 0.16	2.20 ± 0.29	2.26 ± 0.10	0.42 ± 0.02
	No.02	69.7 ± 0.9	6.05 ± 0.21	2.34 ± 0.25	2.71 ± 0.11	0.55 ± 0.03
	No.03	72.3 ± 1.0	7.08 ± 0.23	2.51 ± 0.22	3.35 ± 0.15	0.64 ± 0.03
	P_C	< 0.001	< 0.001	ns	< 0.001	< 0.001
25 %	No.01	51.4 ± 0.5	3.78 ± 0.11	1.90 ± 0.28	0.98 ± 0.06	0.37 ± 0.02
	No.02	56.6 ± 0.8	4.59 ± 0.16	2.21 ± 0.33	1.35 ± 0.07	0.45 ± 0.02
	No.03	59.8 ± 0.9	5.18 ± 0.19	2.40 ± 0.30	1.67 ± 0.09	0.56 ± 0.03
	P_C	< 0.001	< 0.001	ns	< 0.001	< 0.001
	P_W	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001
	P_C	< 0.001	< 0.001	ns	< 0.001	< 0.001
	$P_{W×C}$	< 0.001	< 0.001	ns	< 0.001	< 0.001

Table 2. Root/shoot ratio (RS), specific leaf area (SLA), abscisic acid content (ABA) and carbon isotope composition ($\delta^{13}C$) in three wheat cultivars as affected by three watering regimes. Data are means ± SE of 10 replicates (P_W - watering effect; P_C - cultivar effect; $P_{W \times C}$ - watering × cultivar interaction).

Soil moisture	Cultivar	RS	SLA [$dm^2\ g^{-1}$]	ABA [$\mu g\ g^{-1}$(FM)]	$\delta^{13}C$ [‰]
100 %	No.01	0.49 ± 0.05	1.92 ± 0.17	0.67 ± 0.05	-29.93 ± 0.37
	No.02	0.41 ± 0.03	2.21 ± 0.19	0.68 ± 0.05	-30.14 ± 0.34
	No.03	0.34 ± 0.02	2.47 ± 0.22	0.64 ± 0.07	-30.37 ± 0.31
	P_C	< 0.001	< 0.001	ns	ns
50 %	No.01	0.72 ± 0.07	1.55 ± 0.13	2.84 ± 0.17	-26.67 ± 0.28
	No.02	0.63 ± 0.05	1.73 ± 0.16	2.21 ± 0.20	-27.91 ± 0.29
	No.03	0.55 ± 0.04	1.89 ± 0.20	1.76 ± 0.13	-28.88 ± 0.32
	P_C	< 0.001	< 0.001	< 0.011	< 0.019
25 %	No.01	1.01 ± 0.09	1.23 ± 0.11	3.43 ± 0.28	-25.48 ± 0.23
	No.02	0.93 ± 0.06	1.45 ± 0.15	2.69 ± 0.25	-26.52 ± 0.26
	No.03	0.86 ± 0.04	1.56 ± 0.16	2.07 ± 0.23	-27.90 ± 0.29
	P_C	< 0.001	< 0.001	< 0.001	< 0.001
	P_W	< 0.001	< 0.001	< 0.001	< 0.001
	P_C	< 0.001	< 0.001	< 0.001	< 0.031
	$P_{W \times C}$	< 0.001	< 0.001	< 0.001	< 0.055

mass (RM), but significant differences in root/shoot ratio (RS) and specific leaf area (SLA) between the cultivars were detected under all watering regimes. Individuals from the wet climate cultivar showed greater HT, TB, LA, GM and SLA, and lower RS than individuals from the dry climate cultivar (Tables 1 and 2). In addition, the watering regimes significantly affected all these growth properties, and the watering × cultivar interaction effect was also significant in these properties except for RM.

Water deficit also significantly affected abscisic acid content (ABA) and carbon isotope composition ($\delta^{13}C$). Water-limited treatment (25 % of field capacity) increased 4.12, 2.96 and 2.23 times ABA, 14.87, 12.01 and 8.13 $\delta^{13}C$ in the dry, middle and wet climate cultivars, respectively (Table 2). Significant differences between the cultivars in ABA content and $\delta^{13}C$ were detected under water-limited treatments, but these differences were not found under well-watered treatment. Compared with the wet climate cultivar, the dry climate cultivar showed higher ABA content and $\delta^{13}C$ under water-limited treatments. In addition, the watering effect was highly significant for both properties, and the watering × cultivars interactions were also significant for ABA content, but not for $\delta^{13}C$ (Table 2), which is consistent with most previous studies (*e.g.* Johnson and Bassett 1991, Read *et al*. 1992, Zhang *et al*. 1996, Li 2000).

Differences in water use strategies between three wheat cultivars from contrasting climate regions were demonstrated and attributed to differences in morphological and physiological responses to water availability. In the present study, genetic influence was very obvious, because environmental differences under the same watering regime in the greenhouse were negligible, thus, the observed variations in growth strategy could be linked to genetic differences.

Significant differences between the cultivars were found in HT, TB, LA, GM, RS and SLA ($P < 0.001$) under different watering regimes. The effect of watering and interaction between watering and cultivar were also highly significant ($P < 0.001$). Water stress affected dry matter accumulation and their allocation more in the dry climate cultivar than in the wet climate cultivar. It implied that the dry climate cultivar was more responsive to water deficits and well-developed root systems than the wet climate cultivar. Root systems are complex and dynamic structures, water uptake may be limited by the amount of roots in a particular soil layer and enhancing root growth can increase drought-tolerance (Klepper and Rickman 1990, Yin *et al*. 2005). Plant productivity under drought stress is strongly related to the processes of dry matter partitioning and the spatial and temporal root distribution (Kage *et al*. 2004). Biomass allocation to root, and the quantity and length of functional roots increased under water-limited treatments (reviewed by Kozlowski and Pallardy 2002). Accordingly, water deficits mostly reduced leaf growth and increased at least relatively dry matter allocation into the root fraction, leading to a significant raise of root/shoot ratio.

Differences between the cultivars in these responses could be related with the local climatic condition of the cultivars, and reflect a particular strategy for adaptation. The dry climate cultivar, which has been cultivated in regions with a low rainfall during growing season, could be assumed to possess a high drought tolerance. In contrast, the wet climate cultivar, which occurs in regions with a high rainfall during growing season, could be assumed to utilize abundant soil water supplies and thus grow rapidly. Similar results were also reported for other plants (Gibson *et al*. 1995, Li *et al*. 2000, Zhang *et al*. 2005). Accordingly, evidence of an inter-relation between growth rate and drought tolerance in wheat cultivars suggests that selecting the wet climate cultivar may increase biomass productivity when water is available, and selecting the dry climate cultivar may possess a high drought tolerance that is achieved at the expense of biomass productivity when water is limited.

References

Akhter, J., Mahmood, K., Tasneem, M.A., Malik, K.A., Naqvi, M.H., Hussain, F., Serraj, R.: Water-use efficiency and carbon isotope discrimination of *Acacia ampliceps* and *Eucalyptus camaldulensis* at different soil moisture regimes under semi-arid conditions. - Biol. Plant. **49**: 269-272, 2005.

Ali, M., Jensen, C.R., Mogensen, V.O., Bahrun, A.: Drought adaptation of field grown wheat in relation to soil physical conditions. - Plant Soil **208**: 149-159, 1999.

Anderson, J.E., William, J., Kriedemann, P., Austin, M.P., Farquhar, G.D.: Correlations between carbon isotope discrimination and climate of native habitats for diverse eucalypt taxa growing in a common garden. - Aust. J. Plant Physiol. **23**: 311-320, 1996.

Blum, A.: Crop responses to drought and the interpretation of adaptation. - In: Belhassen, I. (ed.): Drought Tolerance in Higher Plants: Genetical, Physiological, and Molecular Biological Analysis. Pp. 57-70. Kluwer Academic Publishers, Dordrecht 1997.

Brodribb, T., Hill, R.S.: The photosynthetic drought physiology of a diverse group of southern hemisphere conifer species is correlated with minimum seasonal rainfall. - Funct. Ecol. **12**: 465-471, 1998.

Farquhar, G.D., Ehleringer, J.R., Hubick, K.T.: Carbon isotope discrimination and photosynthesis. - Annu. Rev. Plant Physiol. Plant mol. Biol. **40**: 503-537, 1989.

Gibson, A., Bachelard, E.P., Hubick, K.T.: Relationship between climate and provenance variation in *Eucalyptus camaldulensis* Dehnh. - Aust. J. Plant Physiol. **22**: 453-460, 1995.

Heschel, M.S., Hausmann, N.J.: Population differentiation for abscisic acid responsiveness in *Impatiens capensis* (*Balsaminaceae*). - Inter. J. Plant Sci. **162**: 1253-1260, 2001.

Johnson, R.C., Bassett, L.M.: Carbon isotope discrimination and water-use efficiency in four cool-season grasses. - Crop Sci. **31**: 157-162, 1991.

Kage, H., Kochler, M., Stützel, H.: Root growth and dry matter partitioning of cauliflower under drought stress conditions: measurement and simulation. - Eur. J. Agron. **20**: 379-394, 2004.

Klepper, B., Rickman, R.W.: Modeling crop root growth and function. - Adv. Agron. **44**: 113-132, 1990.

Kozlowski, T.T., Pallardy, S.G.: Acclimation and adaptive responses of woody plants to environmental stresses. - Bot. Rev. **68**: 270-334, 2002.

Li, C.: Population differences in water-use efficiency of *Eucalyptus microtheca* seedlings under different watering regimes. - Physiol. Plant. **108**: 134-139, 2000.

Li, C., Berninger, F., Koskela, J., Sonninen, E.: Drought responses of *Eucalyptus microtheca* provenances depend on seasonality of rainfall in their place of origin. - Aust. J. Plant Physiol. **27**: 231-238, 2000.

Li, C., Wang, K.: Differences in drought responses of three contrasting *Eucalyptus microtheca* F. Muell. populations. - Forest. Ecol. Manage. **179**: 377-385, 2003.

Li, C., Wu, N., Liu, S.: Development of freezing tolerance in different altitudinal ecotypes of *Salix paraplesia*. - Biol. Plant. **49**: 65-71, 2005.

Li, C., Yin, C., Liu, S.: Different responses of two contrasting *Populus davidiana* populations to exogenous abscisic acid application. - Environ. exp. Bot. **51**: 237-246, 2004.

Nayyar, H., Kaushal, S.K.: Alleviation of negative effects of water stress in two contrasting wheat genotypes by calcium and abscisic acid. - Biol. Plant. **45**: 65-70, 2002.

Nayyar, H., Walia, D.P.: Water stress induced proline accumulation in contrasting wheat genotypes as affected by calcium and abscisic acid. - Biol. Plant. **46**: 275-279, 2003.

Pandey, D.M., Goswami, C.L., Kumar, B.: Physiological effects of plant hormones in cotton under drought. - Biol. Plant. **47**: 535-540, 2003.

Pospíšilová, J., Baťková, P.: Effects of pre-treatments with abscisic acid and/or benzyladenine on gas exchange of French bean, sugar beet, and maize leaves during water stress and after rehydration. - Biol. Plant. **48**: 395-399, 2004.

Purty, R.S., Agrawal, V., Gupta, S.C.: Induction of a novel boiling stable protein in response to desiccation and ABA treatments in *Sebania sesban* var. *bicolor* leaves. - Biol. Plant. **49**: 137-140, 2005.

Read, J.J., Johnson, D.A., Asay, K.H., Tieszen, L.L.: Carbon isotope discrimination relationship to yield, gas exchange and water-use efficiency in field-grown crested wheat grass. - Crop Sci. **32**: 168-175, 1992.

Sharp, R.E.: Interaction with ethylene: changing views on the role of abscisic acid in root and shoot growth responses to water stress. - Plant Cell Environ. **25**: 211-222, 2002.

Socias, F.X., Correia, M.J., Chaves, M., Medrano, H.: The role of abscisic acid and water relation in drought responses of subterranean clover. - J. exp. Bot. **48**: 1281-1288, 1997.

Trapeznikov, V.K., Ivanov, I.I., Kudoyarova, G.R.: Effect of heterogeneous distribution of nutrients on root growth, ABA content and drought resistance of wheat plants. - Plant Soil **252**: 207-214, 2003.

Wang, Z.L., Huang, B.R., Xu, Q.Z.: Effects of abscisic acid on drought responses of Kentucky bluegrass. - J. amer. Soc. hort. Sci. **128**: 36-41, 2003.

Xu, X., Zheng, G.Q., Deng, X.P., Medrano, H.: Effects of exogenous abscisic acid and water stress on the growth response of subterranean clover of different genotypes. - Acta bot. sin. **44**: 1425-1431, 2002.

Yin, C., Duan, B., Wang, X., Li, C.: Morphological and physiological responses of two contrasting poplar species to drought stress and exogenous abscisic acid application. - Plant Sci. **167**: 1091-1097, 2004.

Yin, C., Wang, X., Duan, B., Luo, J., Li, C.: Early growth, dry matter allocation and water use efficiency of two sympatric *Populus* species as affected by water stress. - Environ. exp. Bot. **53**: 315-322, 2005.

Zhang, J.W., Marshall, J.D., Fins, L.: Correlated population differences in dry matter accumulation, allocation, and water-use efficiency in three sympatric conifer species. - Forest. Sci. **42**: 242-249, 1996.

Zhang, X., Wu, N., Li, C.: Physiological and growth responses of *Populus davidiana* ecotypes to different soil water contents. - J. arid Environ. **60**: 567-579, 2005.

Zhang, X., Zang, R., Li, C.: Population differences in physiological and morphological adaptations of *Populus davidiana* seedlings in responses to progressive drought stress. - Plant Sci. **166**: 791-797, 2004.