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Ecophysiological responses of two ecotypes of *Dactylis glomerata* L. under water-deficit conditions

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Abstract. Development of improved water-saving varieties is a high priority in forage grass breeding programs especially if the climate change predictions for the Mediterranean region become true. In this experiment the adaptation responses of two ecotypes of *Dactylis glomerata* L., one from a fir forest and the other from an adjacent open grassland, to water-deficit conditions were studied. Plants of these ecotypes were planted into pots and subjected during the growing season to two water supplies: (i) well-watered conditions; and (ii) rainfed conditions. Leaf water potential, relative water content, assimilation rate, stomatal conductance, transpiration rate and water use efficiency were assessed at seven-day intervals during the experimental period. Both water regime and ecotype were found to play some role in the ecophysiological responses of *D. glomerata* to water shortage. Under the conditions of moderate water stress of the present experiment, the forest ecotype showed a drought resistance behaviour, maintaining its photosynthetic efficiency. On the contrary the grassland ecotype showed a drought avoidance behaviour, limiting transpirational loss through stomatal closure, negatively affecting its photosynthetic capacity.

Keywords. Water potential – Water use efficiency – Stomatal conductance – Drought.

Réponses écophysologiques de deux écotypes de *Dactylis glomerata* L. en conditions de déficit hydrique

Résumé. Le développement de variétés nécessitant moins d'apports en eau est une priorité des programmes de recherche sur les céréales, d'autant plus si les prédictions de changements climatiques sur la région méditerranéenne s'avèrent exactes. Dans cette étude, les réponses adaptatives de deux écotypes de *Dactylis glomerata* L., l'un provenant d'une forêt de sapins, et un second d'une prairie adjacente, à un déficit en eau, sont étudiées. Les plants sont mis en pots et lors de la période de croissance l'apport en eau est soit abondant soit analogue à celui fourni par les eaux de pluie. Le potentiel hydrique foliaire, le taux d'humidité relative, le taux d'assimilation, la conductance stomatique, le taux de transpiration, et l'efficacité de l'utilisation de l'eau sont mesurés à 7 jours d'intervalle au cours de la période d'étude. Le régime d'alimentation en eau et l'écotype ont tous les deux des effets sur les réponses écophysologiques de *D. glomerata* sur la réduction de l'apport en eau. Sous les conditions modérées de stress hydrique proposées pour cette étude, l'écotype forestier présente une résistance à la sécheresse permettant alors de maintenir l'efficacité de la photosynthèse. A l'inverse, l'écotype de prairie réagit à la sécheresse en limitant les pertes liées à la transpiration foliaire par une fermeture des stomates, réduisant donc l'efficacité de la photosynthèse.

Mots-clés. Potentiel hydrique – Efficience de l'utilisation de l'eau – Conductance stomatique – Sècheresse.

I – Introduction

Dactylis glomerata L. is a widespread perennial grass species, well adapted to dry conditions (Volaire and Thomas, 1995). Its populations thrive in a variety of environments and differ in their

ability to grow and survive under water deficit conditions (Volaire, 1991). Generally, the species resistance to water stress is attributed both to avoidance and to tolerance mechanisms (Volaire, 1995; Volaire and Thomas, 1995). However, populations from dry sites often use the avoidance mechanism (Volaire *et al.*, 2005). In Greece, populations of *D. glomerata* are found in very different bioclimatic zones. Therefore, studying the physiological performance of different ecotypes of this species, subjected to identical environmental conditions, becomes important. The aim of this paper was to study if the ecotype affects the ecophysiological responses of *D. glomerata* under well-watered and rainfed conditions.

II – Materials and methods

The experiment was conducted in the farm of the Aristotle University of Thessaloniki, Northern Greece (longitude: 40° 34', latitude: 23° 43'), at an altitude of 10 m above sea-level. The climate of the area could be characterized as Mediterranean semiarid with cold winters. The mean annual precipitation is 443 mm and the mean annual temperature is 15.5°C. Monthly precipitation (mm) and average temperature (°C) during the experimental period are presented in Table 1.

Table 1. Monthly precipitation and average temperature over the experimental period

	January	February	March	April	May	June	July
Precipitation (mm)	96	11	51	14	44	59	12
Temperature (°C)	6.2	6.0	9.6	13.3	20.3	23.3	26.6

In autumn of 2008, vegetative tillers from 40 randomly selected individual plants of *Dactylis glomerata* were collected from a fir forest and from an adjacent open grassland at Pertouli area (mountains of Central Pindus) at an elevation of 1200-1600 m. Approximately five single tillers of each plant were initially planted into plastic pots, filled with organic matter. The pots were placed in a greenhouse for two months. Eight plants were randomly selected from each ecotype (fir forest and open grassland) and two similar in growth tillers from each plant were transplanted to 16 plastic pots (30 cm diameter, 30 cm depth) filled with soil from the farm. Plants were subjected during the growing season (April-June) to two water regimes: (i) well-watered conditions (up to field capacity) (WW); and (ii) rainfed (RN) conditions. The pots were arranged in a randomized complete-block design with four replications. Four ecophysiological measurements during the experimental period were obtained on clear sunny days around solar noon (12.00-14.00 h) at approximately seven-day intervals. All measurements were obtained on a sample of four mature and intact, fully expanded, upper leaves of each ecotype. Leaf water potential (Ψ) was obtained with the pressure chamber technique (Koide *et al.*, 1991). Relative water content (RWC) was determined on 4 mm discs from leaves similar in age and orientation, and from the same plant to those used for the Ψ determination, according to the procedure described in Iannucci *et al.* (2002). At the same time assimilation rate (A), transpiration rate (E) and stomatal conductance (g_s) were measured with a portable infrared gas analyser system (LCpro+, ADC Bioscientific; Field *et al.*, 1991). Water use efficiency (WUE) was calculated as the ratio of A/E.

One-way analysis of variance was conducted in order to indicate any significant differences between treatments at the end of experimentation and the Duncan's multiple range test at $p < 0.05$ was used to detect differences among means (Steel and Torrie, 1980). Non-linear regressions of varying Ψ in respect to RWC were fitted per ecotype and irrigation treatment using a quadratic function at $p < 0.05$. All statistical analyses were performed with the use of the SPSS statistical software v. 17.0 (SPSS, Chicago, IL, USA).

III – Results and discussion

The two ecotypes presented lower values of g_s , RWC and Ψ under rainfed conditions (Table 2). However, during the experimental period both ecotypes followed the same pattern of changing their Ψ in relation to RWC under well watered conditions (Fig. 1). The same pattern was also observed in the forest ecotype under rainfed conditions with lower values of RWC and Ψ , indicating no limitation of transpirational demands. Indeed, the forest ecotype showed higher values of transpiration rate compared with the grassland one, independently of the water regime used (Table 2). In addition, under rainfed conditions the assimilation rate of the forest ecotype, though lower, was not significantly different from the assimilation rate of both ecotypes under well watered conditions.

Table 2. Mean values (n=4) of physiological parameters of two ecotypes (F: forest and G: grassland) of *Dactylis glomerata* under well watered (WW) and rainfed (RN) conditions at the end of the growing season

Physiological parameters	Treatment				p
	F-WW	G-WW	F-RN	G-RN	
Water potential (MPa)	-1.80 a	-1.67 a	-2.33 b	-2.27 b	***
Relative water content (%)	94.3 a	92.6 a	81.2 b	82.3 b	**
Transpiration rate (mmol m ⁻² s ⁻¹)	1.85 a	1.25 b	1.68 a	1.22 b	*
Stomatal conductance (mol m ⁻² s ⁻¹)	0.08 a	0.07 a	0.05 b	0.04 b	**
Assimilation rate (μmol m ⁻² s ⁻¹)	9.17 a	9.59 a	7.39 ab	5.09 b	*
Water use efficiency	4.91 b	7.96 a	4.35 b	4.19 b	**

a,b: different letters in each column indicate significant differences according to Duncan's multiple range tests. ***p<0.001, **p<0.01, *p<0.05.

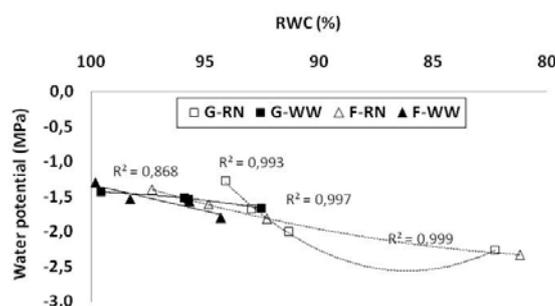


Fig. 1. Relationship between water potential and relative water content (RWC) of two ecotypes (F: forest and G: grassland) of *Dactylis glomerata* under well watered (WW) and rainfed (RN) conditions (n=4).

On the contrary, the grassland ecotype under rainfed conditions responded more conservatively to water shortage maintaining its RWC with decreasing Ψ (Fig. 1). This was probably achieved by stomatal closure (Table 2), induced by hydraulic or chemical signals (Jones, 1998). The closure of the stomatal apparatus resulted also in the reduction of the plant's photosynthetic capacity (Table 2). Instead, plants of this ecotype, when well watered, had significantly higher values of WUE compared to the other treatments, attributed to the lower values of transpiration rate (Table 2).

It has been previously reported that plants from the same bioclimatic zone as the ones used in this study have similar characteristics with populations of temperate zones (Volaire, 1995; Zaharaki *et al.*, 2008). It seems that the growing environment within the same bioclimatic zone played an important role in the ecophysiological responses of *D. glomerata* to water shortage. Under the moderate water stress encountered in the rainfed conditions, due to the existence of rainfall during the experimental period (Table 1), the forest ecotype showed a drought resistance behaviour, maintaining the efficiency of its photosynthetic machinery, which would probably lead to higher production. Indeed, Abraham *et al.* (data under publication in this issue) found that the total production and the number of tillers of this ecotype were high even under rainfed conditions. On the other hand, the grassland ecotype showed a more drought avoidance behaviour, which could be used as an adaptation mechanism to the more intense drought conditions faced in open grasslands compared to forests, and could be also associated with heavier grazing pressure (Zaharaki *et al.*, 2008). However, these results are preliminary. The forest and grassland genotypes need to be tested for more years and under more severe water stress, as the response of the forest ecotype is possible to be a result of phenotypic plasticity and not adaptation (Perez de la Vega, 1997).

IV – Conclusions

Our preliminary results indicate that the ecotype had some important role in the ecophysiological responses of *D. glomerata* to water shortage. Under the conditions of moderate water stress of the present experiment, the forest ecotype had a drought resistance behaviour, maintaining its photosynthetic efficiency. On the contrary, the grassland ecotype showed a drought avoidance behaviour, limiting transpirational losses through stomatal closure, negatively affecting its photosynthetic capacity. From this point of view, selecting plants growing in the forest understorey in a breeding program could possibly lead to the development of productive varieties under water shortage.

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