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MORPHOLOGICAL ADAPTATION OF LOLIUM RIGIDUM GAUDIN TO DIFFERENT CONDITIONS OF THE MEDITERRANEAN SEMI-ARID ENVIRONMENT

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SUMMARY - A field evaluation, aimed at comparing several morphological traits of one cultivar ("Wimmera") and a Sardinian genotype ("Nurra") of annual ryegrass, was carried out in Northern Sardinia. The two accessions were compared over two years in a field experiment, following the UPOV rules for the identification of new cultivars. The distinctness of "Nurra" for flag leaf, tiller, spike and spikelet length was clearly demonstrated. Also, some "Nurra" plant adjustments in water-limited environmental conditions in late spring (elongation of spikelets and awns, late heading, increase of awn presence, higher fertility) seem to be an adaptive response of a drought-tolerant morphotype. The results of this experiment contribute to the agronomic valorisation of "Nurra" ryegrass as a valid drought-tolerant material suitable for introduction into the semi-arid pasture seed market.

Key words: Lolium rigidum, "Nurra", drought-tolerant morphotype, adaptation.

INTRODUCTION

Lolium rigidum Gaudin plays a potentially important role in the qualitative and quantitative improvement of marginal pastures of the Mediterranean climatic areas (Kemp, 1974; Bullitta, 1976; Stephen et al., 1977; Spackman and Cook, 1979; Murphy and Whiteman, 1981; Roggero et al., 1990; Roggero et al., 1993; Franca et al., 1995; Franca et al., 1996). The only cultivar available on the market is "Wimmera", an old Australian cultivar selected for its high forage yield (Mackay, 1982; Kloot, 1983). Further selection has been made from "Wimmera" and, as a result, another variety "Wimmera 62" has been released in California, USA (USDA, 1973). This scarce availability of L. rigidum varieties may be related to the declining importance of the species for the seed industry. In Australia, several constraints such as resistance to selective and non-selective herbicides (Gill, 1996) and the toxicity phenomenon involving annual ryegrass (ARGT) (Riley and Ophel; 1992; Riley and Gill, 1994), have been the major causes negatively affecting the development of new cultivars.

The demand for annual ryegrass seeds is currently high in the Mediterranean regions, for its use in mixtures with annual self-reseeding pasture legumes and as a pure stand, for creating a 3 to 4-year persistent self-reseeding meadow serving a double purpose (hay/pasture) (Caredda *et al.*, 1997). It is hard to quantify the amount of *L. rigidum* seeds traded on the international market, because it is often associated with *L. multiflorum*, also named "annual ryegrass" in US and Australian markets (Bravi *et al.*, 1994).

In the last decade, the CNR – ISPAAM has carried out several collections and field evaluations of Sardinian natural populations of *L. rigidum*, in order to introduce new varieties well adapted to semi-arid Mediterranean conditions. These materials have

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shown great variability for important ecological and agronomic traits (Franca *et al.*, 1993, Bullitta *et al.*, 1997). In particular, one of these populations, named "Nurra", originated from the Nurra Plain region in the northwest of Sardinia, has shown better agronomic performances (re-growth ability, high forage and seed production and specific drought resistance mechanisms) under water-limited regimes, compared to the cultivar "Wimmera" (Franca *et al.*, 1996, Franca *et al.*, 1997; Ledda *et al.*, 1997; Franca *et al.*, 1998).

The aim of the present research was to assess the morphological traits of "Nurra" and "Wimmera" annual ryegrasses, as indicated by the International Union for the Protection of New Varieties of Plants (UPOV, 1990), for the identification of new cultivars, and to check the homogeneity and stability of such traits over two years.

MATERIALS AND METHODS

A two-year field trial was carried out in 1995-96 and 1997-98 at the Ottava Research Station of the University of Sassari in the north-west of Sardinia (Italy). The soil was shallow and calcareous with a N content = 2 kg⁻¹ (Kjeldhal) and P₂O₅ content = 0.053 g kg⁻¹ (Olsen). The climate of the area is Mediterranean semi-arid, characterised by mild winters and dry summers and with an average annual rainfall of 547 mm, concentrated in autumn and winter.

In both years, the Sardinian ecotype "Nurra" and the Australian cultivar "Wimmera" were compared in a randomised complete block design with five replicates. In November, the seeds germinated in open air in Jiffy pots and, after a month, 15 plants per plot were transplanted into the field with a 1 x 1 m spacing. Before the transplanting, 75 kg ha⁻¹ of N and 150 kg ha⁻¹ of P₂O₅ were applied.

As suggested by the guidelines for conducting the tests for distinctness, homogeneity and stability (DHS) of the International Union for the Protection of New Varieties of Plants (UPOV, 1990), in both years the following measurements on each plant were made:

- at inflorescence emergence: natural plant height, tiller length and flag leaf length;
- at complete elongation of the inflorescence: spike length and number of spikelets per spike.

Tiller and flag leaf length were measured on three randomly selected tillers per plant; spike length and number of spikelets per spike were determined on five inflorescences per plant.

Throughout April 1998, three sub-phases of the beginning of the heading stage were identified:

1st: first spikelet emerged in each plant;

- 2nd: at least three spikes with one emerged spikelet;
- 3rd: more than three spikes with at least three emerged spikelets.

On four different dates, the number of plants belonging to each sub-phase was counted.

Moreover, four spikes per plant were chosen and then, on three spikelets (apical, medium and basal) per each spike, the spikelet and the glume length and the average length of the awns, if present, were measured.

STATISTICS

The distinctness of the morphological traits in the two accessions was estimated by processing the data by χ^2 and *t* Student (SAS System, 1987).

The coefficients of variation (C.V.), calculated for each year within accessions (n = 60), was used to check the homogeneity of the morphological variables.

The comparison of the morphological traits over the two years, made throughout a combined ANOVA, was used to check the stability of the traits, after verifying the homogeneity of variances with the Bartlett's test (Gomez and Gomez, 1984); the "year" was considered as a random factor and "Reps within year" was used as the error term to test its significance.

RESULTS AND DISCUSSION

Climatic data

Remarkable differences were recorded between years (Figure 1): in the 2^{nd} year, total rainfall was more than 160 mm higher than in the 1^{st} year, but with an irregular distribution of rains that caused an anticipated water deficit after late April, approximately 45 days before the beginning of the water deficit in the 1^{st} year. The fluctuation in weather over the two years of the trial allowed the identification of the most reliable morphological traits for differentiating between *L. rigidum* accessions.

Plant morphology

Plant height differed only in the 1st year ($P \le 0,05$): on average, "Nurra" plants were 15 cm taller than "Wimmera" (Tab. 1). In the 2nd year, no significant difference was found between accessions; a reduction of the natural plant height for both accessions, even with similar values of tiller length, was recorded, due to a slight tiller lodging at inflorescence emergence. In both years, significant differences were found between the accessions in terms of flag leaf and tiller length. "Nurra", on average, showed flag leaves and tillers,

respectively 33% and 20% longer than "Wimmera". In terms of homogeneity, the flag leaf length showed C.V. always higher than 20% (in both years and for each accession), while natural plant height and tiller length resulted sufficiently homogeneous traits, with C.V. between 12 and 18%.

Spike morphology and fertility

In both years, significant differences between the accessions were found for spike length, number of spikelets spike⁻¹, number of flowers spikelet⁻¹ and number of seeds spikelet⁻¹ (Tab. 2). Also, the spikelet length/glume length ratio differed in both years (respectively for $P \le 0.05$ and $P \le 0.01$). On average, the "Nurra" spike was more than 6 cm longer than "Wimmera", with about 4 spikelets spike⁻¹, 2-3 flowers spikelet⁻¹ and 1-2 seeds spikelet⁻¹ more. The spikelet length differed only in the 2^{nd} year ($P \le 0.01$). The glume length did not differ between the accessions, ranging from 11.4 mm to 12.5, in the 1st and 2nd year respectively. In the 2nd year, "Nurra" showed higher flower fertility, with 7 seeds per spikelet, against 5 seeds in "Wimmera".

Among the measured traits, the number of flowers spikelet⁻¹ and seeds spikelet⁻¹ presented the highest C.V. in both accessions (> 25%), while the lowest variability was ob-

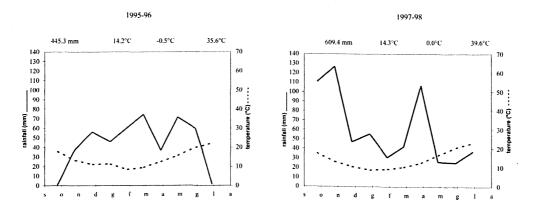


Fig. 1 - Climate diagrams for the experimental site in the two years (Walter and Lieth, 1967). The intersection between the rainfall and temperature lines indicates, respectively at early June and late April, the beginning of the water stress period.

Tab. 1 - Morphological characteristics at inflorescence emergence: averages, t Student's test results and coefficients of variation (C.V.).

Year	Accession	Natural plant height (cm)	Flag leaf length (cm)	Tiller length (cm)
1995-96	Nurra	84	15	80
	<i>C.V.</i>	12	22	13
	Wimmera	69	12	67
	C.V.	15	23	13
	t	*	*	*
1997-98	Nurra	66	17	78
	C.V.	18	27	11
	Wimmera	58	12	64
	<i>C.V</i> .	16	21	12
	t	n.s.	**	**

served for spike length and spikelet length, with C.V. lower than 15%.

The similarity of the C.V. estimated for the morphological traits between accessions and years explains how their variability may be more related to species characteristics than to variety or environmental conditions.

Stability

Table 3 shows the results of the combined ANOVA over the two experimental years. The lack of stability between years in plant height has already been explained by the lodging of the tillers at inflorescence emergence. The length of flag leaf, tiller and spike, together with the number of spikelets spike⁻¹, did not differ significantly; such traits seemed to be less sensitive to the environmental conditions, showing a high stability. The glume length was a stable trait within species, but tended to increase when the anticipated water deficit occurred.

Both accessions were similarly influenced by the climatic differences between the two years, as shown by the non-significant interaction "year *x* accession".

Awn frequency and awn length

The frequency and the length of the awns (Tab. 4) showed a decreasing trend from the

Tab. 2 - Spike morphology at inflorescence emergence: averages, *t* Student's test results and coefficients of variation (C.V.).

Year	Accession	Spike length (cm)	Spikelets spike (n.)	Spikelet length (mm)	Glume length (mm)	Spikelet/ glume ratio	Flowers spikelet ⁻¹ (n.)	Seeds spikelet ⁻¹ (n.)
1995-96	Nurra	29.2	20.6	14.8	11.8	1.26	7	5
	C.V.	15	18	14	15	12	23	31
	Wimmera	23.2	17.3	13.1	11.4	1.20	5	4
	C.V.	15	17	15	15	13	28	30
	t	**	**	n.s.	n.s.	*	**	*
1997-98	Nurra	30.9	21.3	18.5	12.5	1.56	9	7
	C.V.	12	18	12	18	17	19	22
	Wimmera	24.6	17.2	16.2	12.5	1.31	6	5
	C.V.	14	12	15	13	13	25	26
	t	***	*	**	n.s.	**	***	**

n.s. = not significant; * = significant for P \leq 0.05; ** = significant for P \leq 0.01; *** = significant for P \leq 0.001.

Tab. 3 - Stability of plant morphological traits: results of the combined ANOVA over the two years of experimentation.

Variable Source	d.f.	Natural plant height	Flag leaf length	Tiller length	Spike length	Spikelets spike ⁻¹	Spikelet length	Glume length			Seeds ¹ spikelet ⁻¹
Year	1	***	n.s.	n.s.	*	n.s.	***	**	***	***	***
Reps. within year	8										
Accession	1	**	***	***	***	***	***	n.s.	**	***	**
Year * Accession	1	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
Error	8										

apical to the basal spikelets. In both years, "Nurra" presented significantly higher awn frequency than "Wimmera". In "Nurra", the average awn length differed between the years, showing higher values in 1997-98. The Australian accession showed a negligible variation between years, in awn frequency and length.

Heading stage

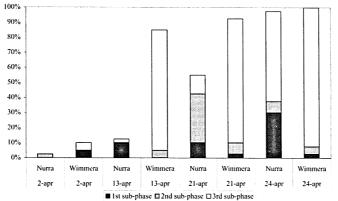
The two accessions differed in heading time (Fig. 2). With the exception of the last observation date (24th of April), when in almost all the plants the heading had started, "Nurra" showed a lower total (1st + 2nd + 3rd sub-phases) heading percentage than "Wimmera"; in mid April, in particular, the Sardinian accession, presented about 72% less headed plants than the Australian variety, although occasional rains and unusual wet conditions prevailed. Furthermore, "Wimmera" plants seemed to be characterised by a shorter mean duration of heading, as showed by the reduced development time of the three sub-phases considered: in this accession, the percentage of plants at the 3rd sub-phase increased from 5% to 80% in 11 days (from the 2nd to the 13th of April), while "Nurra", at he last observation date still presented 30% of plants at the 1st sub-phase.

"Nurra" confirmed the delay in the heading time compared to "Wimmera" (Franca *et al.*, 1996; Franca *et al.*, 1997) and, as a consequence, a longer maintenance of forage palatability for the grazing animals, due to the delay in the lignification of the tissues that occurs at the beginning of the reproductive stage. This characteristic of "Nurra" may have an important agronomic effect, as

Tab. 4 - Awn frequency and length in the apical, central and basal spikelets (counted on 300 spikes per acces-	
sion): X ² and t Student test results.	

Year	Accession	Apical spikelets		Central s	pikelets	Basal spikelets		
		Frequency (%)	Length (mm)	Frequency (%)	Length (mm)	Frequency (%)	Length (mm)	
1995-96	Nurra	34	2,2	23	1,8	9	1,7	
	Wimmera x ²	6 ***	2,5	2 ***	2,8	0 ***		
	t		n.s.		*		—	
1997-98	Nurra	64	3,5	58	3,0	38	2,3	
	Wimmera x ²	9 ***	2,5	6 ***	2,3	3 ***	1,1	
	t		*		n.s.		n.s.	

n.s.= not significant; * = significant at P \leq 0.05; *** = significant at P \leq 0.001;

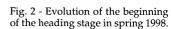


it may allow late grazing, while not compromising seed production.

CONCLUSIONS

Climatic variability seems to influence the occurrence of morphological adaptations in both "Nurra" and "Wimmera" annual ryegrasses: with a dry late spring and when the anticipated water deficit occurs, all L. rigidum plants lengthen the flag leaf and increase spike fertility. However, under these same conditions, the presence of awns and their elongation, seems to have particular relevance only in "Nurra" plants. This trait was previously indicated in cereals as an ultimate source of energy for seed formation and maturation processes in water-limited conditions (Schaller et al., 1972, Johnson et al., 1975) and, therefore it could be considered a further trait for defining a L. rigidum drought-resistant morphotype, in addition to the ones previously indicated by Franca et al. (1998).

Even in different experimental conditions, the results obtained in this investigation confirm "Wimmera" as a strictly "drought escaper", while "Nurra" shows again "drought avoidant" traits, with higher seed production capability and later heading time than the Australian material (Franca *et al.*, 1998).



For its adaptation and persistence under semiarid conditions and for the previously reported productive performances, "Nurra" represents a valid alternative to the Australian cultivar.

The results highlight the importance of the selection of native ecotypes for Mediterranean farming systems, compared to extra-European pasture cultivars, which are not always suitable for our semi-arid Mediterranean environments.

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