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*RESEARCH, REVIEWS, PRACTICES,
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Comparison of Selected Ecotypes
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(*Dactylis glomerata* L.)
Under Pinewoods in Spain

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ABSTRACT. Cocksfoot (*Dactylis glomerata* L.) entries, eight natural populations, and two commercial checks were sown under maritime pine (*Pinus pinaster* Ait.) and under red pine (*P. sylvestris* L.) canopies, as well as in an open field area in Galicia, NW Spain. Natural populations yielded less biomass and had a higher crude protein content under pine canopies than in an adjacent open field stand. In the first two years under *P. pinaster*, population Mg348 yielded, in general, significantly more dry matter than the commercial varieties and more than control plots, which were fertilized but not sown. In contrast, two years after sowing, control plots invaded by spontaneous grasses yielded significantly more than several cocksfoot entries under red pine and in the unshaded stand. Therefore, ecotypic selection in cocksfoot is an alternative to the use of commercial varieties in temporal pastures under *P. pinaster*. In the long term, however, cocksfoot does not persist significantly more than spontaneous grasses. doi:10.1300/J064v29n02_03 [Article copies available for a fee from The Haworth Document Delivery Service: 1-800-HAWORTH. E-mail address: <docdelivery@haworthpress.com> Website: <<http://www.HaworthPress.com>> © 2006 by The Haworth Press, Inc. All rights reserved.]

KEYWORDS. Orchardgrass, pasture yield, *P. pinaster*, *P. sylvestris*, silvopastoral systems

INTRODUCTION

The shade from trees in silvopastoral systems potentially decreases the persistence and production of pasture species, and some cocksfoot cultivars have better tolerance to shade at low photosynthetically active radiation (Devkota et al., 1998). Development of shade-tolerant pasture under pinewoods results in more forage, with better nutritive value that may reduce fire risk more than a pyrophilous understory (Rigueiro, 1985; Koukoura et al., 1999). On the other hand, the introduction of shade-tolerant perennial grass and legume species can improve silvopastoralism. Cocksfoot is a forage grass with some shade-tolerant ecotypes that originate in temperate woodlands, like subspecies *aschersoniana* (sometimes growing in *Pinus cembra* taiga: Türesson, 1929), subsp. *izcoi* from Galicia (northwest Spain) sometimes growing in oakwoods (Lindner and García, 1997), and coastal ecotypes in Galicia, seen by the authors growing spontaneously under *P. pinea*. Since cocksfoot is also an important forage, it has been used for grazing purposes under pinewoods in Spain (Piñeiro and Pérez, 1988; Silva-Pando et al., 1998),

Chile (Ovalle and del Pozo, 1995, cited by Koukoura and Papanastasis, 1996), and Greece (Papanastasis et al., 1995).

This study compares the establishment, dry-matter yield, and survival of different cocksfoot entries under three environments, two pine species and an open field. The objectives were (1) to determine if yield, growth and survival of natural populations justifies the use of ecotype selection in cocksfoot, specifically for pinewoods, as an alternative to the use of commercial varieties, and (2) to verify that, on fertilized soil under different pine canopies, sown cocksfoot yields more than spontaneous grasses.

Preliminary results until November 1998 were presented in an International Symposium (García et al., 1999).

MATERIALS AND METHODS

Field Experiments

Eight natural populations of *Dactylis glomerata* subsp. *izcoi*, seven tetraploids—station code Mg followed by an entry number, and one diploid—station code Mg340, were collected in Galicia. Some plants of two populations—codes Mg137 and Mg175 were collected under oak canopies. All populations were sown on 1 m² plots, on 6 May 1998, in a pinewood in Monfero-A Coruña, Galicia (Spain), together with two commercial cultivars: “Artabro” from Galicia variety, and “Baraula” from the Netherlands but sown commonly in the North of Spain.

The pinewood was 55 years old, located at 650 m altitude, with an average trunk diameter ranging 25-27 cm and an average tree height ranging 11-13 m. The soil was an acidic clay-loam over slate. The climate is Atlantic and the site had mean annual temperatures ranging 10-14°C and total annual rainfall of 1500-1800 mm.

The experimental area is an 11-hectares fenced stand, including different environments covered with two pine species and with treeless areas inside. The environments were: *P. sylvestris* canopy with a density of 700 trees ha⁻¹ and 21% light transmission, *P. pinaster* canopy with 833 trees ha⁻¹ and 36% light transmission, and a stand without trees considered as 100% light transmission.

The experimental layout was a split-plot design with environments as the main plot factor. The ten entries were sown on each environment, in three parallel completely randomized blocks facing northeast. The soil was harrowed one month before sowing. In the open field, gorse vegetation was cut before harrowing. Fertilization was applied to each plot, the

day before sowing, by supplying 72 kg P ha⁻¹, 150 kg K ha⁻¹, 41 kg N ha⁻¹, 28 kg Ca ha⁻¹, and 13 kg Mg ha⁻¹. Sowing density was 30 kg of seed ha⁻¹. In each replicate, one control plot was fertilized but not sown. A second dose of N, Ca and Mg fertilization was applied after the harvest in April 1999, to ensure nutrient maintenance in the soil. Data presented are cocksfoot establishment, average cocksfoot height, percentage of soil cover, production of aerial biomass, and crude protein content. Establishment was visually scored ranging from zero as no emergence to seven as complete establishment. Average cocksfoot height was measured in cm. Percentage of soil cover was visually scored. Production of aerial biomass was determined as dry-matter yield, drying separated cocksfoot in the samples for 24 h at 80°C, expressed as kg ha⁻¹. In the control plots, spontaneous grasses were cut and weighed. Crude protein content was calculated multiplying by 6.25 nitrogen percentage, determined by semimicro Kjeldahl method. Characters, which could only be determined under *P. pinaster* were dry-matter yield on November 1998 and *in situ* photosynthesis fixation rate of fully expanded leaves on September 1999 (with LICOR 6400, adjusting IRGA CO₂ concentration at 400 ppm, and LED light source at 800 μmol m⁻² s⁻¹). Establishment, height, and soil cover were evaluated in the first two months after sowing (June and July 1998). Biomass was harvested on September 1998, November 1998 (only in *P. pinaster* plots), April 1999 and April 2000, but crude protein analyses were made only in April 1999 and April 2000 samples. In January 2002 and March 2004, soil cover percentage was visually evaluated to quantify cocksfoot persistence in time.

Statistical Analyses

In order to standardize data distributions, soil cover percentage scores were transformed by the arcsin of the square root (Papanastasis et al., 1995) and establishment scores were transformed by the square root. In spite of the high coefficients of variation showed by dry-matter yield, data have not been transformed by the logarithm, because this distorts the interaction term (Li, 1969), losing its biological sense. For the detection of differences between environments and cocksfoot entries, a split-plot analysis of variance (ANOVA) with missing values (where entries are the secondary factor) was applied to the data, excluding control plots. Significantly, different entry means were compared by Duncan's multiple range test, including control plots to verify the effect of fertilization alone, using the error mean square of the global split-plot ANOVA.

RESULTS

Cocksfoot began to emerge three days later under *P. sylvestris* (21 May 1998) than in the other stands (18 May 1998).

The split-plot analysis of variance detected significant differences between cocksfoot entries in establishment, average plot height in July 1998, and soil cover and dry-matter yield in April 2000 (data not shown).

Means comparison test (Table 1) shows that Mg387 was the tallest entry and the one which covered most of the soil in 1998 and 2002. The diploid Mg340 had the poorest establishment and soil cover in July 1998, but its soil cover remained stable on average while the commercial variety Baraula had the poorest soil cover in 2002 and 2004 (Table 1). Control plots had the lowest establishment of spontaneous grasses and the lowest soil cover in 1998, 2002, and in the no-tree stand in 2004. These grasses were mainly *Agrostis curtisii* in the no-tree stand, *Agrostis capillaris* in both pine stands, and *Pseudarrenatherum longifolia* under *P. pinaster*.

There were significant differences between stands in average cocksfoot plant height, dry matter yield, and crude protein content. In the no-tree stand, cocksfoot was taller, yielded more in April 1999 and had

TABLE 1. Mean establishment scores, canopy heights, and soil covers for ten cocksfoot entries and an unsown control.

Entry	Establishment score #	Height (cm)	% Soil cover (not transformed)	
	June 1998	July 1998	July 1998	January 2002
Mg387	3.2a	28.8a	52.7a	66.0a
Artabro	3.1ab	25.5abc	45.3ab	37.3b
Mg348	3.0ab	24.2abc	45.9ab	42.8b
Mg96	2.8ab	27.8ab	43.8abc	37.0b
Mg305	2.6ab	23.5abc	38.4bc	40.9b
Mg175	2.5ab	20.1c	34.4bcd	36.4b
Mg196	2.4abc	22.8bc	36.7bcd	36.0b
Baraula	2.0abc	25.7abc	30.7cd	0.6c
Mg137	1.8bcd	23.4abc	35.7bcd	38.0b
Mg340	1.3cd	22.3bc	30.1cd	30.1b
Control	0.9d	23.3abc	23.2d	0.0c

#Stand establishment (not transformed), was scored on a scale of 0 to 7 with 7 representing complete establishment.

Numbers within columns followed by different letters are significantly different at $\alpha \leq 0.05$ according to Duncan's multiple range test.

lower protein content in 1999 and 2000 than under both pine species (Table 2). The stands of *P. pinaster* and *P. sylvestris* showed no significant differences in cocksfoot dry weight in April 1999 nor in crude protein content in April 1999 and 2000 (Table 2).

Dry matter yield in September 1998 and in April 2000 showed a significant entry \times stand interaction (genotype \times environment interaction). Again, cocksfoot entries yielded more in the no-tree stand than under pines (Table 3). In September 1998, every entry (including the control) yielded more under *P. pinaster* than under *P. sylvestris*. However, in April 2000, Artabro, Baraula, Mg175, Mg387, and the control yielded more under *P. sylvestris* than under *P. pinaster*. In this harvest, control plots were the most productive in the no-tree stand and under *P. sylvestris*. Under *P. pinaster*, Mg348 and Mg96 were the most productive, significantly more than the control. Baraula was the least productive in the three stands. Although in September 1998 there were no global differences between cocksfoot entries, there were significant differences within each stand: Mg348 and Mg96 were again the most productive under *P. pinaster*, significantly more than the control (Table 3), and they continued to be so in November 1998. Mg348 yielded significantly more than Artabro and Baraula under *P. pinaster* in September 1998, November 1998, and April 2000.

Mean crude protein content of grass in control plots was not significantly different from that of cocksfoot plots on each year: 13.6% versus 13.4% in 1999, $t_9 = 0.3$ ns; and 10.7% versus 12.8% in 2000, $t_7 = 1.3$ ns (fewer degrees of freedom in 2000 due to higher variance in the control).

Soil cover in 2004 also showed a significant entry \times stand interaction. Now the open field had less grass soil cover than both pine stands. The

TABLE 2. Means of height, dry weight, and crude protein content of ten cocksfoot entries under two pine canopies and a treeless stand.

Environment	Height (cm)	Dry weight (kg ha ⁻¹)		Crude protein (%)	
	July 1998	April 1999	April 1999	April 1999	April 2000
Open field	32.4a	37.6a	11.8b		10.9b
<i>P. pinaster</i>	24.2b	11.3b	13.9a		13.5a
<i>P. sylvestris</i>	17.5c	3.2b	14.6a		13.9a

Numbers within columns followed by different letters are significantly different at $\alpha \leq 0.05$ according to Duncan's multiple range test.

TABLE 3. Means of character showing significant environment \times entry interaction for ten cocksfoot entries and an unsown control.

Entry	Yield (kg.ha ⁻¹) Sep 1998			Yield (kg.ha ⁻¹) Apr 2000			Yield (kg.ha ⁻¹) Nov 1998		% Soil cover (not transformed) Mar 2004	
	No-tree	<i>P. pinaster</i>	<i>P. sylvestris</i>	No-tree	<i>P. pinaster</i>	<i>P. sylvestris</i>	<i>P. pinaster</i>	<i>P. sylvestris</i>	No-tree	<i>P. pinaster</i>
Artabro	950a	139g	66a	319cd	56cd	183ab	249bcd	2.0ab	17.3bc	78.3a
Mg175	943a	157fg	12de	474bcd	27d	64b	175cd	6.7a	8.3bc	28.3b
Mg387	872b	194e	31b	599abc	181ab	209ab	311abcd	1.7ab	26.7abc	85.0a
Mg348	715c	286b	10def	618abc	190a	119ab	562a	11.7a	20.0abc	56.7ab
Mg196	661d	249c	7f	456bcd	114abcd	93b	422abc	0b	15.0bc	55.0ab
Mg137	609e	168f	7f	458bcd	96abcd	70b	298abcd	1.7ab	31.7abc	20.0bc
Mg305	585e	199de	9ef	899ab	174abc	62b	303abcd	0b	66.7ab	56.7ab
Mg96	542f	406a	30b	271cd	186ab	119ab	470ab	0b	26.7abc	66.7ab
Control	442g	19i	18c	1030a	71bcd	306a	129d	3.3ab	73.3a	65.0ab
Baraula	370h	114h	13d	37d	4d	21b	183cd	6.7a	0c	0c
Mg340	317i	219d	9ef	424bcd	106abcd	85b	261bcd	0b	42.5abc	55.0ab

Numbers within columns followed by different letters are significantly different at $\alpha \leq 0.05$ according to Duncan's multiple range test.

exception was Baraula, which disappeared in both pine stands but was the most successful in the no-tree stand (together with Mg175). Mg196, Mg96, and Mg340 disappeared in the no-tree stand and had their maximum soil cover under *P. sylvestris*. Mg305 also disappeared in the no-tree stand, but had its maximum soil cover under *P. pinaster*, like Mg137 and the control plots. Artabro, Mg175, Mg387, and Mg348 had their maximum soil cover under *P. sylvestris*.

DISCUSSION

Isolated Measurements

In November 1998, biomass could not be harvested under *P. sylvestris* nor in the no-tree stand, because of the entrance of two calves into both environments. A visual estimate of the amount of forage eaten was higher in the no-tree stand than under *P. sylvestris* (mean values of 3 vs. 0.2). Goto et al. (1982) have also found lower values of cocksfoot palatability under *P. sylvestris* at 50% sunlight, than in unshaded conditions.

Environments and the Sowing of Cocksfoot

We found emergence three days later under *P. sylvestris* (21% sunlight) than in the other stands. Park et al. (1986b) similarly reported that the emergence of grasses was eight days earlier in full sunlight than under pine trees (50% sunlight).

Cocksfoot was shorter under *P. pinaster* (64% shade) than in the no-tree stand, in contrast with the average cocksfoot height found by Park (1986b) with taller values under 50% shade than under 25% or 75%.

Cocksfoot production under *P. pinaster* and *P. sylvestris* canopies was 30% and 8%, respectively, of that of unshaded stand (Figures 1, 2, and 3). Silva-Pando et al. (2002) found similar results in an adjacent stand with a cocksfoot-clover mixture. Photosynthesis fixation rate measured in September 1999 under *P. pinaster* had a mean value of $4.51 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$, which represents 16% of the maximum value obtained in nonlimited conditions under full sunlight by Peri et al. (2003) fixed at $27.4 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$. This fact shows the “shade tolerance” of cocksfoot, because with photosynthesis fixation decreased to 16%, it was able to produce 30% of the herbage dry matter of the full sunlight stand. Joshi et al. (1999)

FIGURE 1. Cocksfoot plots in an open field stand.



found that cocksfoot's dry-matter production was halved under *P. radiata* (18% photosynthetically active radiation) compared with open pasture.

In April 2000, two years after sowing, control plots were the most productive in the open field stand and under *P. sylvestris*. This year was particularly humid, and ferns (*Pteridium aquilinum*) reached 2 m in height and 90% of ground cover in *P. pinaster* stands, increasing the shade effect, while no ferns were present in open field and *P. sylvestris* stands. Nitrogen content was higher under pine trees than in the no-tree stand, probably because of shade and to a pine effect on pastures described by Davis (1995), who suggests that pines incorporated into farming systems may be able to increase nitrogen availability to pasture species. Burner (2003) obtained higher crude protein content under *Pinus taeda* than in the unshaded control. Papanastasis et al. (1995) found that nitrogen content of understory vegetation, including sown cocksfoot, decreases when *P. pinaster* is thinned. Lindner (1976) and Park et al. (1988) also found more nitrogen content in cocksfoot with increasing artificial shading. However, Joshi et al. (1999) found less nitrogen content in cocksfoot under *P. radiata* and Peri et al. (2001) observed that crude protein values were not influenced by shade (50-60% light intensity).

FIGURE 2. Cocksfoot plots in a *Pinus pinaster* stand.FIGURE 3. Cocksfoot plots in a *Pinus sylvestris* stand.

Along the first two years, the unsown control plots were colonized mainly by *Agrostis curtisii* in the open field stand and by *Agrostis capillaris* under pines. Both species are annual grasses which germinate in spring. Thus, they were absent in January 2002. However, in April 2000, control plots were the most productive in the no-tree stand and under *P. sylvestris*. Therefore, in the case of *P. sylvestris*, harrowing and fertilization was enough to get grass production in spring. Shrub species (*Ulex europaeus*, *U. gallii*, *Erica cinerea*, *E. mackaiana*, and *Daboecia cantabrica*), herbaceous species as *Potentilla erecta* and bryophytes were present four years after sowing in the open field stand. In 2004, the full plot area was covered by a 50 cm tall, dense layer, which obstructed any grass seed emergence. In *P. pinaster* and *P. sylvestris* stands, spontaneous grass species, mainly *Agrostis capillaris*, grow in competition with the survivor cocksfoot. In the open field, the plant community, which dominated before the trial, re-established itself. In the *P. sylvestris* stand, cocksfoot survived better: The deeper root system of old pines does not compete with cocksfoot and shade maintains air and soil-surface moisture, even in summer, favoring cocksfoot over other species. The mean values of powdery mildew found under *P. sylvestris* were higher than under *P. pinaster* and in the no-tree stand (García et al. 1999), suggesting increased humidity in the shade environment, as Park et al. (1986a) had found under pine trees giving 50% shade. An intermediate situation between the unshaded stand and *P. sylvestris* cover occurred in the *P. pinaster* stand.

Variation in Cocksfoot, Its Use Under P. pinaster

Significant differences between cocksfoot entries are mainly genotypical and they were found in yield in April 2000, while no significant differences were found in photosynthesis fixation rate in the stand under *P. pinaster* (data not shown). Yield differences between cocksfoot cultivars were also found by Koukoura and Papanastasis (1996) and Devkota et al. (1998). In the present study, the interaction environment by entry in yield indicates that cocksfoot selection must be applied on each environment. Under *P. pinaster* (36% light), Mg348 and Mg96 were the most productive entries. Koukoura and Papanastasis (1995) also found that cocksfoot establishes well under *P. pinaster*, when light intensity is >20% of the total radiation. However, in the present study, in 2004 control plots had the highest soil cover in the *P. pinaster* stand. Baraula and the diploid population Mg340 were, in general, the less productive cocksfoot entries. Lindner (1976) and Lumaret et al. (1987) have also

found significant differences in yield between diploid and tetraploid cocksfoot grown in shaded pots.

Sibbald (1999) wondered if the best forage species and varieties were being recommended for silvopastoral systems. The conclusion of the present study is that the sowing of cocksfoot under *P. pinaster* in Galicia is justified as a temporal pasture. In these conditions, ecotype selection can be used as a breeding method in cocksfoot, since the two commercial varieties, the foreign Baraula and the autochthonous Artabro, produce less than Mg348 in the first two years. However, in the long-term, cocksfoot persistence was not greater than the colonization of spontaneous grasses in any of the three stands.

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