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**GENETIC VARIATION AND GENOTYPE BY ENVIRONMENT INTERACTION OF  
CROWN RUST RESISTANCE IN ANNUAL RYEGRASS**

**(*LOLIUM MULTIFLORUM* LAM.)**

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**Abstract**

The genetic variation and genotype by environment interaction of crown rust resistance of annual ryegrass (*Lolium multiflorum* Lam.) was evaluated at two locations in Argentina, as part of the breeding programme in progress at our Institute. Forty half sib families originated from commercial tetraploid cultivars, were grown from seeds in a greenhouse during autumn 1999. At the stage of tillering initiation forty eight plants/family were randomly selected and transplanted as spaced plant trial in a randomized design with three replicates, at the two locations. All plants were scored for a range of morphological and phytopathological attributes. The severity of crown rust and the winter dry matter production were assessed on each plant. Crown rust resistance at both evaluation dates, was highly heritable and not affected by environment interaction. By contrast, winter dry matter showed a large family by location interaction and a high environmental influence. The results of the present study indicated that important progress in crown rust resistance in annual ryegrass can be achieved through phenotypic recurrent selection.

**Keywords:** crown rust resistance, *Lolium multiflorum*, genotype by environment interaction.

## **Introduction**

One of the most common and serious diseases of Italian ryegrass (*Lolium multiflorum* Lam.) is crown rust (CR), caused by the fungus *Puccinia coronata*. Many authors have shown that high levels of CR in ryegrass can have a substantial effect on green matter yield, rather than in dry matter (Lancashire and Latch, 1966) and in forage quality (Thomas, 1997), diminishing the milk and meat production. In addition to this, it has caused rejection of diseased herbage in grazing trials. In Argentina, many foreign cultivars have consistently shown high susceptibility to *P. coronata*, and there is evidence of serious constraints on forage yield (Andrés and Bertin, 1998). The behavior of these materials is probably due to lack of adaptation to local environmental conditions, where no attention has been paid to disease problems. The objectives of this study were (1) to evaluate the resistance to CR of cultivars of Italian ryegrass; (2) to study the genotype – environment (location) interaction on winter growth and CR resistance; (3) to select genotypes for the breeding programme in progress at INTA. The final aim of this programme is to provide varieties of Italian ryegrass, with reasonable levels of resistance to CR.

## **Material and Methods**

Trials were carried out at two locations Pergamino (33° 56' S, 60° 33' W) and Concepción del Uruguay (32° 29' S, 58° 20' W) that represent the middle and Northern area of the Humid Pampas, Argentina. Forty half sibs families originated from genotypes out of the following commercial tetraploid cultivars: Blizzard, Comet, Grandesa, Hercules, Magnum, Max and Tama, were grown from seeds in a greenhouse during autumn 1999. At the stage of tillering initiation, forty-eight plants per family were transplanted into individual

pots to vegetative increase the material. Then, they were transplanted at both locations with a spatial arrangement of 0,50 m within row and 1,0 m between rows. The trials were designed as 3-replicated randomised complete blocks, with 8 plants per replicate and per family. All plants were scored for a range of morphological and phytopathological attributes. Winter growth activity was evaluated by cutting aerial biomass (10 cm height) at the end of the winter and dry matter production per plant was registered. The severity of CR was assessed on each plant as the proportion of leaf area with symptoms, using a local scale from 0 (0% affected tissue) to 5 (100% affected tissue) (Andrés, 1999). Assessments were carried out twice in the spring of 1999 (early expression: October; late expression: November), after natural infection was detected at the field. Statistical analyses were performed on each attribute by using the SAS programme (SAS Institute Inc., 1989). Analyses of variance were carried out on plot mean basis and variance components were calculated according to Nguyen and Sleper (1983). Data on CR visual scoring was transformed ( $\sqrt{x+1}$ ) prior to analyses. Narrow-sense heritabilities on a phenotypic family mean basis averaged over replications and locations were estimated as:  $h^2_{PFM} = \frac{\sigma^2_F}{(\sigma^2_F + \sigma^2_{F.L}/l + \sigma^2_e/l.r)}$ , where  $\sigma^2_F$ = HS family variance,  $\sigma^2_{F.L}$ = family by location interaction variance, and  $\sigma^2_e$ = environmental variance due to interplot variability. In order to compare cultivar CR reaction, family data was pooled according to cultivar of origin, and an unbalanced ANOVA (GLM, SAS) analysis was performed.

## **Results and Discussion**

Mean, range, variance components and narrow-sense heritability on a phenotypic mean basis of measured variables are presented on Table 1. As expected, winter growth, according to its polygenic character and high environmental influence, showed a large family by location interaction and environmental variance components, resulting in a low heritability

estimate. In contrast, crown rust resistance, at both evaluation dates, was highly heritable and not affected by interactions with the environments. The present state of knowledge indicates that in some populations resistance to CR in ryegrass is under polygenic inheritance and that in others it is controlled by major genes (Kimberg, 1999). Narrow-sense heritabilities estimates on CR resistance in ryegrass varies between 0.22 and 0.68 (Kimberg, 1999). The results of this study showed much higher values than those reported in the literature. One reason may be that the HS families evaluated in this study were generated out of several populations (cultivars) that have showed marked differences in CR resistance (see below). Another likely reason is that CR resistance was measured using visual scores that were well correlated with spore production and pustule per unit of leaf surface (Kimberg, 1999), but did not account for the within class variability. The results did not show an important, although significant, family by location interaction which probably indicates that no abiotic environmental effects were important in the expression of the resistance and/or similar race/s of the pathogen were present at both locations. However, other researchers have found significant genotype by environment (year and location) interactions and have stated the need of multilocation evaluation for CR resistance breeding (Kimberg, 1999). When data was pooled according to original cultivars that generated the HS families, large differences in CR resistance were detected (Table 2). Although the families studied represented only a sample of the whole variability present in the cultivars, the cultivar ranking obtained in this study was in agreement with that obtained in dense stand trials (De Battista, unpublished). Finally, the results of the present study indicated that important progress in crown rust resistance in annual ryegrass can be achieved through phenotypic recurrent selection. Multilocation evaluation is necessary for other polygenic characters, such as dry matter yield, that are involved in the plant breeding programme in progress at our Institue.

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**Table 1** – Mean, range, variance components and narrow-sense heritability on phenotypic mean basis for winter growth and crown rust resistance in annual ryegrass evaluated at two locations.

Variable	Mean	Range	$\sigma^2_F$	$\sigma^2_{F.L}$	$\sigma^2_e$	$h^2_{PFM}$
Winter growth (g/plant)	39.87	2.1-115.6	10.1	19.05	14.62	0.231
Crown resistance early (0-5)*	1.57	0.00-3.75	0.0729	0.0072	0.0030	0.877
Crown resistance late (0-5)*	3.41	0.00-5.00	0.1428	0.0044	0.0036	0.947

$\sigma^2_F$  = family variance;  $\sigma^2_{F.L}$  = family x location variance;  $\sigma^2_e$  = environmental variance;  $h^2_{PFM}$  = narrow-sense heritability

\* SCALE = 0 (0% affected tissue) to 5 (100% affected tissue)

**Table 2** - Crown rust resistance scores among cultivars.

Cultivar	Crown resistance early*	Crown resistance late*
Hercules	0.46 c	1.42 d
Blizzard	0.63 c	1.98 c
Magnum	0.70 c	1.80 c
Grandesa	1.93 b	4.04 b
Comet	1.80 b	3.90 b
Max	2.41 a	4.82 a
Tama	2.54 a	4.92 a

Within columns means followed by different letters differ  $p < 0.05$ . Duncan's test

\* SCALE = 0 (0% affected tissue) to 5 (100% affected tissue)