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Agromorphological and grain quality characterisations of northern Spanish wheats under low-nitrogen conditions

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Abstract – Fifteen wheat landraces were evaluated in a low-nitrogen system (50 kg·ha⁻¹·year⁻¹ of N) at Mabegondo Agronomic Centre during two years in a randomised complete block design with two replicates of 50 plants. Plots consisted of single rows 2 m long. Two commercial wheat cultivars (Marius and Soissons) were also planted. The entries (landraces and cultivars) were characterised for seven qualitative and 15 quantitative traits. This data set was reduced to four principle components that cumulatively explained 80% of the total variance. Cluster analysis was useful in identifying three groups of landraces with differentiated characters. In general, wheat landraces performed as well as commercial cultivars for grain yield and grain quality characteristics, except for Zeleny values which were low in most of the landraces. Conservation of these resources, if possible in situ, should be a priority objective.

Triticum aestivum L. / agromorphological and grain quality evaluation / landrace / low-nitrogen / northern Spain

Résumé – Caractérisation agronomique et qualité du grain de blé du nord de l'Espagne sous conditions de fertilisation azotée réduite. Quinze variétés de pays de blé ont été évaluées sous une fertilisation azotée faible (50 kg N·ha⁻¹·an⁻¹) au Centre de Recherche Agronomique de Mabegondo pendant deux ans dans un dispositif en deux blocs complets randomisés où la parcelle élémentaire était un rang de 2 m. Deux cultivars commerciaux de blé (Marius et Soissons) ont été aussi évalués. Sept caractères qualitatifs et 15 caractères quantitatifs ont été considérés. Cet ensemble de données a été réduit à quatre composantes principales qui expliquent 80 % de la variance totale. La classification ascendante hiérarchique a permis l'identification de trois groupes différenciés de variétés de pays. En général, les variétés locales de blé ont eu des performances agronomiques similaires aux cultivars commerciaux à l'exception de

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faibles valeurs du test Zélény pour la plupart des variétés locales. La conservation, si possible in situ, de telles ressources nous paraît un objectif prioritaire.

Triticum aestivum L. / évaluation agromorphologique et qualité du grain / fertilisation azotée réduite / nord d'Espagne / variété de pays

1. Introduction

The surface areas dedicated to the cultivation of wheat in Galicia and in Asturias are 30 000 and 180 hectares respectively [10], they are part of almost all the traditional rotations, especially in Galicia. This difference is because in Asturias at present, wheat is not an important crop due to the specialisation of farms in forage crops for animal production (milk and beef). The collapse of traditional farming systems and abandonment of agrarian activities is perhaps the main threat to these wheats.

However, there is an important market at the regional level for bread made with the flour of wheat landraces that promotes to some extent its cultivation. The cultivation of these landraces shows disadvantages that relate to the high lodging incidence due to their long culm and because of their low grain yield in high fertilisation conditions. For these reasons, commercial wheat cultivars are increasingly in use [7].

The importance of northern Spain for wheat genetic resources led the International Board of Plant Genetic Resources to declare it as a zone of priority for the collection of wheat destined for genebanks [2]. The main objectives of genetic resource management are to conserve genetic diversity and stimulate its use [9]. The description of agronomically important and useful characters is an important prerequisite for effective and efficient use of germplasm collections in breeding programs [3]. In spite of the importance of the Spanish landraces only limited agromorphological characterisation data are available [5, 12, 13].

The objective of this study was to estimate the agromorphological and grain quality diversity of northern Spanish wheat landraces. This effort was motivated by the fact that autochthonous materials are at risk of being lost.

2. Materials and methods

The 15 northern Spanish wheat landraces studied originated from two regions: eleven landraces from Galicia and four from Asturias (Fig. 1). These landraces are still cultivated in these regions. This germplasm is conserved at the Mabegondo Agronomic Centre. Two commercial wheat cultivars (Marius and Soissons) were chosen as controls due to the fact that they are two of the most cultivated wheat cultivars in Spain and that they are used as official controls in the Spanish Wheat Evaluation Program. The study was conducted over two successive years (1997/1998, 1998/1999) at one location of Northwestern Spain (Galicia). The site was at Mabegondo Agronomic Centre (43° 15' N, 8' 18' W) near the coast (100 m.a.s.l.) on a silt loam soil. Soil was sampled on September 1997 to a depth of 15 cm to determine initial soil nutrient status. Analysis was done by the Galician Agrarian Laboratory.

The crop cycle was from November to the end of July. In both seasons, each entry (landraces and cultivars) was planted in two replicates in a randomised complete block design. Plots consisted of single rows 2 m long. Rows were spaced 0.5 m apart in both years. This wide spacing was chosen to prevent possible negative interactions between rows due to potential lodging. Fifty seeds were planted per row to facilitate individual plant observation. Fertilisation practices were 50 kg·ha⁻¹ of nitrogen applied at the end of tillering and 150 kg·ha⁻¹ of P₂0₅ and K₂0. Traditional fertilisation practices in the cultivation of wheat landraces does not include any mineral nitrogen application.



Figure 1. Geographical origin of 15 wheat landraces and the site of characterisation (Mabegondo).

There was no application of any fungicide or growth regulator.

The entries were characterised for seven qualitative (1 to 7) and 15 quantitative traits (8 to 22). The values for traits 1 to 5, 9 to 13 and 15 were calculated as the mean of five sounded spikes per row, (1) glume beak length (1 short to 5 long), (2) glume beak shape (1 straight to 5 geniculate), (3) glume shoulder width (1 narrow to 5 broad), (4) glume shoulder shape (1 sloping to 5 elevated with second point present), (5) spike density (1 lax to 5 dense), (6) stem rust (Puccinia graminis) incidence on the row (1 low to 5 high), (7) lodging incidence on the row (1 low to 5 high), (8) heading date of the row (days past 1st January), (9) plant height (cm) at maturity from soil to apex of a spike, not including the awns, (10) number of spikelets per spike including sterile, (11) rachis length (mm), (12) glume length (mm), (13) glume width (mm), (14) maturity date (days past 1st January), (15) number of grains per spike, (16) mean grain length of 25 grains per row (mm), (17) mean grain width of 25 grains per row (mm), (18) grain yield of the row $(g \cdot m^{-2})$, (19) thousand grain weight (g), (20) test weight $(g \cdot L^{-1})$, (21) grain protein content as N concentration multiplied by 5.7(%), and (22) the Zeleny value (ml).

For the quantitative characteristics studied, univariate normality was checked on their residues. Their shapes and the Shapiro-Wilk statistics showed normal distributions. The following model of analysis of variance was applied:

 $X = Mu + Ye + bl + Ye \cdot bl + En + Ye \cdot En + Err$

where Mu = overall average; Ye = year effect; bl = block effect; $Ye \cdot bl = year \cdot block interaction$; En = Entry; $Ye \cdot En = year \cdot Entry interaction$; Err = residual error.

The year effect was considered random and the signification of the Entry effect was tested using the Year · Entry interaction as error term.

The effect of the competition between tall (landraces) and small (cultivars) wheats on grain yield and on yield components (grains/spike, spikelets/spike and the thousand grain weights) was taken into account by using one covariate based on the mean difference in height between a row and its two adjacent neighbours. The mean difference in heading date was also used as a second covariate to see if there was an effect of the different heading dates on grain yield and yield components [6].

For the qualitative-scored characteristics the non-parametric Kruskal-Wallis test was used to test the existence of statistically significant differences between entries.

Multivariate relationships among landraces were revealed with a principal component analysis (PCA) using a correlation matrix derived from the significant quantitative and qualitative characters. The original variables were reduced to four independent linear combinations, principle components (PC) of the variables, with eigenvalues greater than 1. These PCs were used as the input for an ascendant cluster analysis to detect groups of similar agromorphological and grain quality types. Ward's clustering algorithm, which minimises within-cluster variance summed over all variables was used. Statistical analyses were computed using SAS [14].

3. Results

Soil testing at the commencement of the experiment showed a pH of 5.3, cation exchange capacity of 14.9 cmol(+)·kg⁻¹, and organic matter content of 4.1%. Potassium, Ca and Mg were present at levels of 0.4, 4.8 and 0.4 cmol(+)·kg⁻¹, respectively. Olsen P content was 30 ppm and Ammonium acetate K content was 162 ppm.

The covariate calculated as the difference between the height of a row and the mean height of its two adjacent neighbours was statistically significant for grain yields and for the number of spikelets per spike. For these characters the adjusted means will be given. The effect of the covariate calculated as the difference of heading date had no statistically significant effect on grain yield and yield components.

Significant differences (P < 0.05) between entries were obtained for all characters except spikelets per spike, grains per spike and grain yield (Tab. I). On average, wheat landraces performed as well as commercial cultivars, except for Zeleny values. Landraces presented higher stature and longer rachis and glumes than cultivars. For the qualitative characteristics, non-parametric Kruskal-Wallis tests showed significant differences between entries (landraces and cultivars) (P < 0.001) for all the characteristics, except the glume beak shape, stem rust and lodging incidences. Landraces in general showed more lax spikes and longer glume beak than cultivars.

PCA showed that the four first principal components (eigenvalues greater than 1) accounted for 87% of the total variance. Correlation coefficients between the first four PCs and the original characteristics were computed to detect which of the characteristics more strongly contributed to the PCs. PC1 explained 47% of the total variance and was positively correlated with plant height, rachis length, glume width, protein content and glume beak length. It was negatively correlated with Zeleny value and spike density. PC2 accounted for 21% of the total variance and was positively correlated to glume width, grain length, grain width and thousand grain weight. PC3 (11% of the total variance) was mainly positively correlated to glume shoulder width and glume shoulder shape and negatively correlated to test weight. Finally, PC4 accounted for 8% of the total variance and was positively correlated to heading and maturity dates.

Hierarchical clustering analysis performed on the first four principal components leads to the dendrogram presented in Figure 2. This dendrogram was cut at the four cluster level describing 64% of the phenotypic variation among entries. Cluster 1 consisted of the two commercial cultivars, Marius and Soissons. They presented the highest Zeleny values, short compact spikes, low plant height, short glume and glume beak and low protein concentration. Cluster 2 contained two Galician (Monterroso and Veiga) and one Asturian (Narcea) landraces. All were early to head and mature. They had narrow glumes, short and narrow grains, low thousand grain weight and were intermediate in spike length and spike density.

Cluster 3 grouped 11 landraces, had long lax spikes, wide glumes, long and wide grains, and with high thousand grain weight. They were intermediate to head and mature. Cluster 4 consisted of only one landrace from Galicia (Xinzo) late to head and mature. It was poorly adapted to the Mabegondo environment, which contributed to its low test weight and thousand grain weight. It presented narrow glumes, wide and pointed glume shoulders and short and narrow grains with short lax spikes.

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characters. F level of signi significantly	ralues (ficance differen	for quai given.*, t at 5% a	ntitative **,** s accordin	traits) significa g to the	of the er ant at 0.(e Bonfer	atry effe 05, 0.01 roni T te	ct and C and 0.0 ests.	Chi-squa 01 leve	are valu Is respe	es of th ctively.	le Krusl Means	kal-Wa in the a	lis test same rov	(for the v follow	qualitati 'ed by d	ve trai ifferen	ts) with tetters	their were
Quantitative Characters	F values	Ordenes	Coris- tanco	Fene	Callobre	Ponte- ceso	Narcea	Limés	Oscos	Castro	Llacín	Veiga	Villalba	Xinzo	Monte- rroso	Mabe- gondo	Soissons	Marius
Heading date Plant height Spikelets/spike Rachis lenoth	6.7*** 26.7*** 0.7NS	148bcd 143.3abcd 20.7 121 1ab	149bc 142.7abcd 21.6 127 1ab	154a 137.0cd 21.1 107.6cde	147bcd 145.8abc 19.8 173.4ab	148bcd 143.6abcd 21.2 173.0ab	140ef 140.6bcd 20.7	146cd 135.1d 1 19.7 121 2ab	147cd 43.8abcd 21.2 115 7bcd	148bcd 140.7bcd 22.4 124.0ab	146cd 144.6abcd 21.5 129.6a	136f 135.6d 19.7 108.6cde	151abc 151.1a 21.8 118 1bc	152ab 140.9bcd 19.8 107 3e	140ef 150.0ab 20.9	148bcd 146.5abc 19.8 115 3bcd	143d 80.3e 17.4 82 4a	138f 84.7e 18.0 97 3f
Glume length Glume width Maturity date	6.6** 7.3*** 9.8**	13.3def 13.3def 4.6ab 201ab	122.140 14.9abc 4.6ab 205a	13.2def 13.2def 4.1fg 206a	14.6abcd 4.5bcd 206a	13.7cde 4.4bcde 205a	12.1f 12.1f 4.3cdef 184de	14.7abcd 4.8a 198abc	13.0ef 13.0ef 4.5bcd 201ab	15.3ab 15.3ab 4.6bc 193bcd	122.04 13.4def 4.4bcde 196abc	15.5a 15.5a 4.2efg 179e	113.6cde 13.6cde 4.5bcd 205a	13.3def 4.0g 205a	11.9f 11.9f 4.3def 190cd	13.9bcde 4.5bcd 206a	02.75 10.2g 3.9g 200ab	10.2g 10.2g 4.3cdef 192bcd
Grains/spike Grain length Grain width	1.8NS 7.5*** 3.9***	46.5 7.6abcdef 3.6abc	46.9 7.7abcde 3.6abc	41.6 7.5abcdef 3.7ab	44.6 7.3cdef 3.8ab	43.9 7.5abcdef 3.7ab	43.9 7.1def 3.5abc	49.0 8.3a 3.6abc	47.7 7.8abcde 3.8a 767 8	52.6 7.9abcd 3.7ab	48.4 8.2ab 3.8a 257 3	44.5 7.2cdef 3.3bc	47.7 7.8abcde 3.7ab	37.7 7.5abcdef 3.5abc	46.6 7.3bcdef 3.5abc	43.3 8.1abc 3.7ab	45.5 6.7f 3.2c 132 7	40.5 6.9ef 3.4abc
Thousand Grain weight Test weight Protein content Zeleny value	4.5** 2.5* 3.6**	52.0ab 52.0ab 76.9ab 12.6ab 19.0cde	52.7ab 74.3b 12.9a 18.0cde	50.3ab 77.9ab 12.9a 16.0cde	<i>57.4</i> a <i>77.7</i> ab 12.4ab 21.5bcde	53.7ab 77.3ab 12.6ab 23.5bcd	43.4ab 75.9ab 12.1ab 15.5cde	54.2ab 74.3b 11.8ab 17.0cde	52.9ab 76.5ab 12.6ab 22.5bcd	53.9ab 74.3b 11.4ab 18.5cde	57.7a 74.7ab 11.8ab 12.0de	39.4b 76.3ab 12.0ab 16.0cde	56.2a 77.8ab 12.1ab 14.0cde	41.4ab 72.2b 11.8ab 10.0e	45.6ab 76.8ab 12.9a 21.0bcde	54.4ab 78.8ab 12.6ab 25.5abc	48.1ab 82.0a 111.0ab 36.0a	54.3ab 77.3ab 10.5b 31.0ab
Qualitative Characters	Chi-square Values																	
Glume beak lenght Glume beak shape Glume shoulder width	127.5*** 24.2NS 48.4***	3.1 1.6 2.4	3.4 1.9 2.1	3.1 2.1 2.2	3.3 1.7 2.1	3.1 1.6 2.3	2.7 1.6 2.9	3.1 1.7 2.6	3.1 2.2 2.3	3.1 1.5 2.5	2.9 1.8 2.5	3.5 1.8 2.6	3.1 1.8 2.4	3.2 1.5 2.6	2.6 1.6 2.0	3.1 1.4 2.5	2.0 1.6 2.1	1.7 1.7 2.5
Glume shoulder shane	38.5**	1.4	1.2	1.2	1.1	1.2	1.8	1.8	1.3	1.4	1.5	1.5	1.3	1.4	1.1	1.4	1.1	1.4
Spike density Stem rust incidence Lodging incidence	127.4*** 21.8NS 20.9NS	1.1 3.2 2.5	1.1 3.7 3.0	1.2 2.7 1.0	$1.0 \\ 3.5 \\ 3.0 \\$	1.2 4.0 1.0	1.4 3.7 3.0	1.0 3.5 1.0	1.1 3.5 1.5	1.1 3.2 1.5	1.1 3.2 2.0	1.2 3.7 2.5	1.1 3.0 2.0	1.6 3.5 2.0	1.2 3.2 2.5	1.1 3.0 2.0	2.3 2.0 1.0	$1.9 \\ 1.5 \\ 1.0$

Table I. Means of 15 landraces and two commercial cultivars (Marius and Soissons) of Triticum aestivum L. for 15 quantitative and 7 qualitative



Figure 2. Hierarchical classification dendrogram of 15 wheat landraces and two wheat cultivars.

4. Discussion

This study has permitted the characterisation of some northern wheat landraces multiplied and conserved until now by farmers of Galicia and Asturias, outside a specific program of conservation. These landraces are original in relation to the improved commercial cultivars.

Grain yield was not significantly different among entries. It is assumed that with respect to wheat cultivars, northern wheat landraces exhibit more vigorous growth under adverse growth conditions [8]. The conditions of low-nitrogen fertilisation of our experiment may result in conditions similar to those of marginal areas and this may explain the similar grain yields obtained in wheat landraces compared with cultivars. Covariate adjustment of grain yield by the mean height difference of genotype neighbours was an effective method of reducing bias of genotype means in single-row plots supporting findings from other authors [6].

Protein content was similar in landraces and commercial cultivars. Since wheat end-use quality is strongly influenced by environment, specially as related to grain protein content, the low nitrogen dosage of this study probably had an influence on the protein content. The bread making quality of flour is influenced both by protein content and protein type but for a given protein content, wheat quality largely depends on the nature of the gluten protein composition [4]. The lower Zeleny values of some landraces compared with cultivars showed a probable low gluten strength of the flour. Indeed Zeleny is generally considered as an indirect measure of gluten strength [1].

As generally known from field data, northern wheat landraces mature later than commercial wheat cultivars [8]. Earliness of wheats, measured as days to heading, may have a negative influence on the time of harvest in the climatic conditions of the North of Spain with abundant rain. The time of harvesting in northern Spain is heavily dependent on the climatic conditions. Nonetheless, at the beginning of August, the crop is ready to be harvested. Wheat landraces did not show very high lodging scores. This is probably due to the moderate nitrogen fertilisation applied and the wide spacing between rows. One main goal of breeding wheat landraces should be the reduction of their height to minimise the role of lodging under various conditions of cultivation.

Interest in wheat landraces has recently increased due to their resistance to several diseases [11]. Our results showed no significant differences between entries for stem rust incidences in two years of evaluation.

The results of our work showed the interest of some wheat landraces from Galicia and Asturias

for low-nitrogen fertilisation conditions because they performed as well as two of the most used wheat cultivars. Today, in northen Spain finding small fields of wheat landrace cultivation that has survived associated with traditional agriculture is still possible. However, the changes occurring in these rural communities has led to the abandonment of traditional agrarian practices and, along with it, also the interest in these crops and their cultures. Efforts should be made urgently to safeguard this heritage before it is irretrievably lost.

When evaluated in large numbers, quantitative morphological traits appear to provide a good source of distance information for measuring phenotypic similarity as an estimate of genetic similarity. In addition, they quantify traits of potential interest to breeders. With the aim of revealing other sources of polymorphism, this study should be continued with genetic markers in the form of isozymes, storage proteins, and direct DNA markers.

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