Homogeneity of wheat varieties developed by the use of double-haploids[1]

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SUMMARY – The use of the haplo-diploidy for the development of new cultivars in autogamous species has as a consequence that the evaluated lines for selection must be homocigotic. On the contrary, certain residual segregation between the generator lines is foreseen in conventional programmes. Our group is involved in breeding programmes of durum and bread wheat. In this study, lines of both wheat species have been evaluated for the composition of the endosperm storage proteins previously to be released as cultivars. This analysis has the double finality of evaluate the potential gluten quality together with their homogeneity. Our analyses indicate that double-haploid lines present higher homogeneity than the lines obtained by conventional programmes.

Introduction

One of the limitations for the cultivar registration and identification in autogamous plants is the residual segregation, which is consequence of the moment where the decision of selecting one line is taken. Probably, in this moment, a certain level of heterozygosis is still present in the plants. This heterozygosis will give rise to a residual segregation that can be translated in heterogeneity of the resulting line.

One of the additional advantages of the system of breeding by haplo-diploidy is to avoid this problem, since in this case the generator lines come from the duplication by endomitosis of a haploid cell, which consequently is totally homozygotic.

Given the well-known potential of storage proteins, and in particular of the gliadins to detect polymorphism, the objective of the present work was to use this protein fraction to compare the degree of heterogeneity of lines in evaluation for registration obtained by both systems.

Material and methods

AGROVEGETAL S.A. is breeding for new bread wheat varieties, using segregating material from the International Wheat and Maize Improvement Center (CIMMYT) and also haplo-diploid material from a research program developed jointly with researchers of the Institute of Sustainable Agriculture (CSIC) and the Department of Genetics of the University of Córdoba.

In the present study, gliadins of twelve lines of bread wheat that AGROVEGETAL has under evaluation as candidates for registration have been analysed. These twelve lines have been cultivated in multiplication plots with the same handling system. Six of them (THA1415, THA1417, THA1420, THA1432, THA1438 and THA1450) have been the result of the process of selection conducted in CIMMYT material, while the others (THA1456, THA1460, THA1467, THA1481, THA1490 and THA1461) come from the program of haplo-diploids. In each case, ten individual ears were taken.

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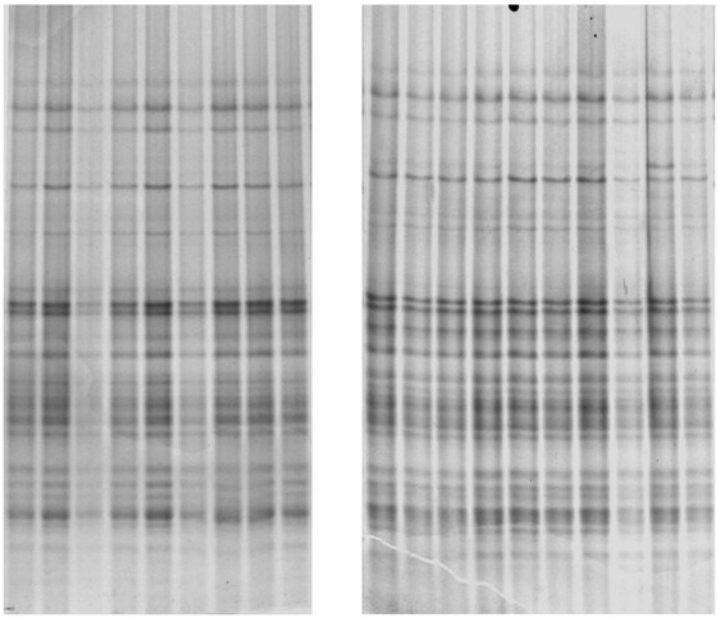
Gliadins were extracted with a 1.5 M dimethylformamide aqueous solution and fractionated by A-PAGE al 8.5% (C=2.67%) with low catalyst levels (ferrous sulfate and hydrogen peroxide) for increasing the gel firmness (Khan et al., 1985). Electrophoresis was performed at 25mA/gel at 18°C for 45 min after the tracking dye (methyl violet) migrated off the gel. Gels were stained overnight with 12% (w/v) trichloroacetic acid solution containing 5% (v/v) ethanol and 0.05% (w/v) Coomassie Brilliant Blue R-250. De-staining was carried out with tap water.

Results and discussion

The study of the corresponding gels revealed that each one of the twelve lines has a characteristic gliadins pattern, having confirmed therefore the potential of this technique for the varietal identification. Both homogenous (Figs 1A and 1B) as heterogeneous materials (Figs 2A, 2B and 2C) were found.



A B

Fig. 1. THA1456 (A) and THA1450 (B).

The difference between both types of heterogeneous materials is the origin of the mentioned heterogeneity, since the analysis of the heterogeneous patterns allows distinguishing whether this heterogeneity is fruit of later mixtures of material in the process of selection as generator lines, or if it is due to processes of residual selection as previously described. In Figs 2A (line THA1490 – haplo-diploid) and 2B (line THA1432 - obtained by conventional selection) there is a dominant pattern which is the characteristic one of the respective lines, and contamination by mixture that present very different patterns from the characteristic one. On the contrary, in the heterogeneity of the line THA1415 (Fig. 2C), obtained by conventional methods, it is observed as the heterogeneity of limits some bands, being the rest of the homogenous. This result is typical of the residual segregation.

Conclusions

In conclusion the gliadin analysis has high potential to be used in varietal identification and as tool to detect heterogeneous materials. In this regard, it is important to emphasize the necessity to establish new systems of homogeneity control to detect the habitual process of mixture in the handling of materials destined to the reproduction. On the other hand, the residual segregation is a source of heterogeneity of the breeding materials obtained by conventional procedures that is not present in haplo-diploid materials.

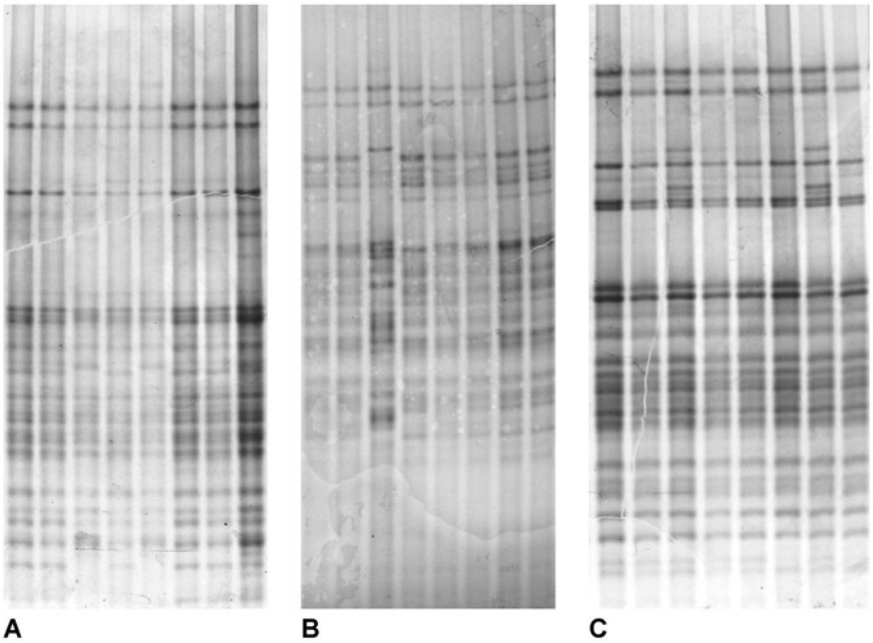


Fig. 3. THA1490 (A), THA1432 (B) and THA1415 (C).

References

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