

Study of genetic diversity of the wheat leaf rust fungus *Puccinia triticina* in Morocco using AFLP

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Introduction :

Leaf (or brown) rust, caused by *Puccinia triticina* Eriks. or *Puccinia tritici-duri* V. Bourgin is an important disease of wheat worldwide that causes enormous losses in Morocco. The disease spreads by means of urediniospores and overwinters on *Thalictrum speciosissimum* L. (in the Ranunculaceae) as a commonly alternative host of *P. triticina* in the temperate zones. In the Mediterranean region, *Anchusa italica* Retz. (in the Boraginaceae) was described as the second host for leaf rust of wheat. The objective of this study was to examine the molecular genetic diversity of *P. triticina* populations in Morocco using the amplified fragment length polymorphism (AFLP) technique and to determine how the variation is partitioned within and between different geographic locations, using Abda-Doukala, Chaouia-Tadla, Gharb-Saïss, and Tangerois north as a case study.

Materials and methods:

Wheat leaves bearing uredinia of leaf rust (*P. triticina*) were collected in 2005 and 2006 throughout fields of wheat in Morocco. The surveyed region covered four agro-ecological areas: Abda-Doukkala, Chaouia-Tadla, Gharb-Saïss and Tangerois north.

DNA of 38 *Puccinia triticina* isolates was extracted from urediniospores using a cetyltrimethyl ammonium bromide (CTAB) protocol with one chloroform step.

The Genetic variability and collection structure of the wheat leaf rust fungus *Puccinia triticina* were investigated by Amplified fragment length polymorphism markers. A set of five AFLP primers combinations Pst/Mse was used and the DNA bands were developed using a silver nitrate stain.

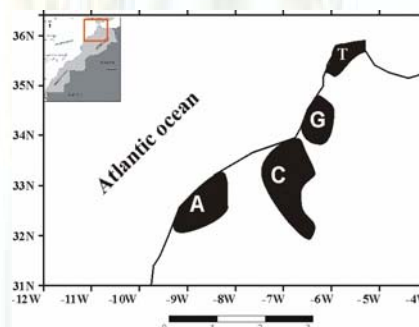


Fig.1 : Agro-ecological areas of cereal production in Morocco showing the origin of the *Puccinia triticina* isolates collected in 2005 and 2006. Area T = Tangerois north, Area G = Gharb-Saïss, Area c = Chaouia-Tadla, Area A = Abda-Doukkala.

Results :

A total of 253 loci was scored for the five AFLP primer combinations. Genetic diversity in terms of percent polymorphic loci (Table 3) was lowest for Tangérois isolates (55.34%) and highest for Chaouia-Tadla and Gharb-Saïss (88.14%). Nei's diversity index and Shannon index were highest for Gharb-Saïss isolates and lowest for Tangérois isolates. Cluster analysis of AFLP markers indicated that isolates from Abda-Doukkala were the most closely related to each other, followed by isolates from Gharb-Saïss, however they tend to cluster with isolates of Chaouia-Tadla as confirmed by the lowest calculated Nei's genetic distance between the two populations (Nei_D = 0.038). This seems consistent with the fact that the two populations are close geographically. The findings of cluster analysis were confirmed with the analysis of two and third dimensional space in Principal Component Analysis. Thus we can conclude that the collection structure occurred with regard to their geographical origin.

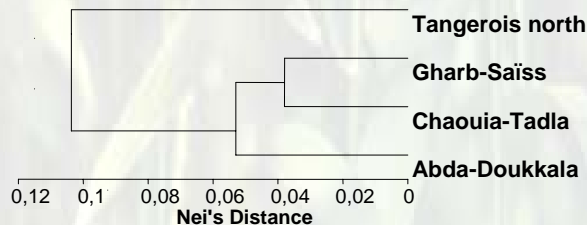


Fig.2 : Graphical representation of genetic distance relationships among four collections of *Puccinia triticina*.

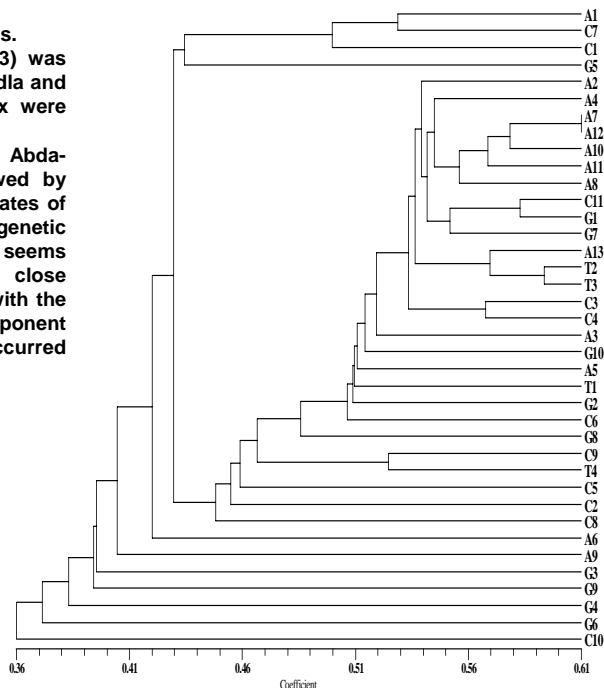


Fig.3 : Dendrogram showing similarities among isolates of *Puccinia triticina* based on amplified fragment length polymorphism (AFLP) analysis (data from five primer combinations combined, based on NTSYS-pc single link cluster analysis and Jaccard's coefficient. T = isolates from Tangerois north, G = isolates from Gharb-Saïss, C = isolates from Chaouia-Tadla, A = isolates from Abda-Doukkala.