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Short Note: Genetics of Chestnut (Castanea sativa Mill.)

II. Uniformity of Isozyme Phenotypes in grafted Orchards

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(Received 12th September 1986)

Summary

Some enzyme systems (LAP, GOT, SKDH, IDH, 6-PGDH, MDH, GDH and G-6-PDH) were studied in bud tissues of European chestnut by means of starch gel electrophoresis. Individual trees of a grafted stand were collected in central Italy. Results reveal the uniformity of the enzyme phenotypes for all of the enzyme systems analyzed. Individual trees can therefore be considered as ramets of the same clone.

Key words: isozymes, grafting, clone, *Castanea*.

Zusammenfassung

Einige Enzymsysteme (LAP, GOT, SKDH, IDH, 6-PGDH, MDH, GDH und G-6-PDH) sind mittels Stärkegelelektrophorese im Knospengewebe von *Castanea sativa* analysiert worden. Einzelne Bäume sind in einem gepfropften Bestand in Mittel-Italien beerntet worden. Ergebnisse zeigen die Einheitlichkeit der Enzym-Phänotypen aller untersuchten Systeme. Die analysierten Einzelbäume können daher als Ramets eines einzigen Klons betrachtet werden.

Introduction

European chestnut (*Castanea sativa* Mill.) is a species which is naturally widespread over the hill and mountain altitude of the entire Italian territory. Because of its economic value, it has always been cultivated and bred for the quality and quantity of the wood and the fruit.

The so-called "varieties" have been developed through grafting and asexual propagation. No certain information is available on the genetic identity of the varieties. The specification of the cultivated varieties is very difficult, because they are usually described and classified only according to their geographic origin. Different varieties, having distinct origin and diverse local names, could be derived from a single variety, which has differentiated morphologically due to environmental factors (Paglietta and Bounous, 1979).

In order to identify (or differentiate) the varieties, numerous studies based on the analysis of morphological characters of the fruit have been carried out (Antonaroli *et al.*, 1983, 1984; Bassi and Sbaragli, 1984; Bassi and Marangoni, 1984; Borghetti *et al.*, 1983). Studies on the genetic structure of chestnut varieties are still lacking. Preliminary studies based on observations of isozymes as environmentally independent traits have been reported by Sawano *et al.* (1984) and Fineschi (1986), but no information has been available on the genetic control of isozyme systems in chestnut species.

The question of whether different individuals belonging to the same variety are genetically identical or not will be discussed in the present study. In fact, it is possible that only a single tree bearing particularly large fruits has been utilized to graft other trees and has therefore founded a so-called "variety". If all trees of a grafted population turn out to possess identical zymograms, they can be considered as individuals of the same clone. This requires that

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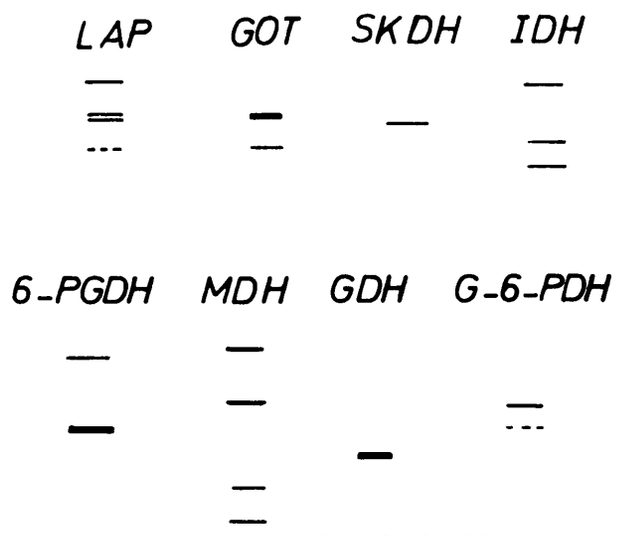

Fig. 1. — Enzyme phenotypes shown by the eight systems.

enzyme phentypes be expressed by the genotypes and not be environmental impacts. Furthermore, the enzyme phenotypes must be controlled by polymorphic enzyme gene loci which show segregation in the case of sexual reproduction. The uniclonal composition could be one explanation for the increased sensitivity of grafted populations to chestnut blight as compared to wild populations.

The purpose of the present study was to verify the uniformity of individual trees with respect to a number of enzyme systems.

Material and Methods

A small stand (around 1 ha) of about 100 grafted trees was selected in a chestnut population in central Italy (Umbria). A sample of 30 individuals chosen at random was examined for the systems LAP (leucine aminopeptidase, E.C. 3.4.11.1), GOT (glutamate oxaloacetate transaminase, E.C. 2.6.1.1.), SKDH (shikimic dehydrogenase, E.C. 1.1.1.25), 6-PGDH (6-phosphogluconate dehydrogenase, E.C. 1.1.1.44), IDH (isocitric dehydrogenase, E.C. 1.1.1.42), MDH (malate dehydrogenase, E.C. 1.1.1.37), GDH (glutamate dehydrogenase, E.C. 1.4.1.2) and G-6-PDH (glucose-6-phosphate dehydrogenase, E.C. 1.1.1.49).

Starch gel-electrophoresis was performed to test the isozyme systems in bud tissues during the winter of 1984. Electrophoretical methods have been previously described (Fineschi, 1986).

Results and Discussion

Enzyme phenotypes of the 30 individual trees are surveyed in *Figure 1.* The results clearly indicate the uniformity of the isozyme phenotypes in the different systems for all of the individuals tested.

In spite of the lack of information on isozyme variability between chestnut individuals and populations, our first results on several samples of wild trees from several different areas in central Italy show a remarkable enzyme polymorphism, both for the cited enzyme systems and for additional systems which we are currently investigating (Fineschi *et al.*, unpubl.). Therefore, the electrophoretic uniformity detected by the analysis of grafted trees cannot be explained by a low degree of variability in this species.

Although no information is available on the genetic control of enzyme systems in chestnut, these analyses suggest that the LAP system could be controlled by three independent gene loci and the MDH, 6-PGDH and IDH systems by more than one locus. Therefore, the uniformity of the phenotypes of the grafted trees investigated does not reflect only the condition of a single gene locus. These results are confirmed by another study (Villani *et al.*, 1986) carried out in winter of 1986, in which bud tissues from about 20 grafted individual trees were analyzed for several enzyme systems. Furthermore, our first attempts on the differentiation of some chestnut "varieties" (Villani *et al.*, unpubl.) reveal that uniformity occurs not only between individuals of the same "variety" but also between "varieties" known to be different and having different names and geographic origin.

These results suggest that grafting practice could have transformed natural forests into clonal stands. Higher degrees of dieback detected in grafted stands as compared to wild populations could be one consequence of this alteration.

Acknowledgements

This study was carried out at the Abteilung für Forstgenetik und Forstpflanzenzüchtung der Universität Göttingen, and was supported by a grant from the National Research Council of Italy (CNR).

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