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Genetic control and modifications of peroxidase pattern in two species of *Nicotiana* and their interspecific hybrid

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Summary. The peroxidase patterns of *Nicotiana suaveolens*, *N. glutinosa* and their interspecific hybrid have been studied, and their possible genetic control analyzed. Modifications of the isozymatic pattern along the plant have been observed.

The peroxidases represent a wide group of isozymes present in plants generally, showing inter and intraspecific variability. This variability is also shown during plant development². Almost all the genetic studies carried out with peroxidases coincide in showing their monomeric character and monogenic control³⁻⁸ and in showing the existence of null alleles^{3,8}. The only exception observed is in rice⁹, in which a dimeric behaviour might be indicated.

In the present work, the study of the patterns of the leaf peroxidase isozymes of *Nicotiana suaveolens* × *N. glutinosa* hybrids and both parental species has been carried out, in order to analyse their genetic control, and also to detect the possible variation of the pattern along the plant.

Material and methods. Flowering plants of *Nicotiana suaveolens* (su) and *N. glutinosa* (glu) as well as their

interspecific hybrid (su × glu) were used. The seeds of the *Nicotiana* species were supplied by the 'Centro Tecnológico del Tabaco', Sevilla (Spain) (su) and the 'Hortus Botanicus Bergianus', Stockholm (Sweden) (glu). The analyses were carried out on crude extracts of leaves taken at different plant levels; level 1 – the basal rosette, level 2 – intermediate, level 3 – floral branches. Peroxidase isozymes were studied using Tris-citric acid 0.015 M, pH = 7.75 as the gel (12% starch) buffer and NaOH-boric acid 0.3 M, pH = 8.6 as the electrode buffer. The gels were stained using the method described by Shaw and Cohen¹².

Results and discussion. Anodal peroxidases (APX) as well as cathodal peroxidases (CPX) were observed. In level 1 (basal rosette) the APX pattern showed a total of 6 bands (Nos 6, 7, 8, 9, 10 and 11) for glu and su × glu, and 5 bands

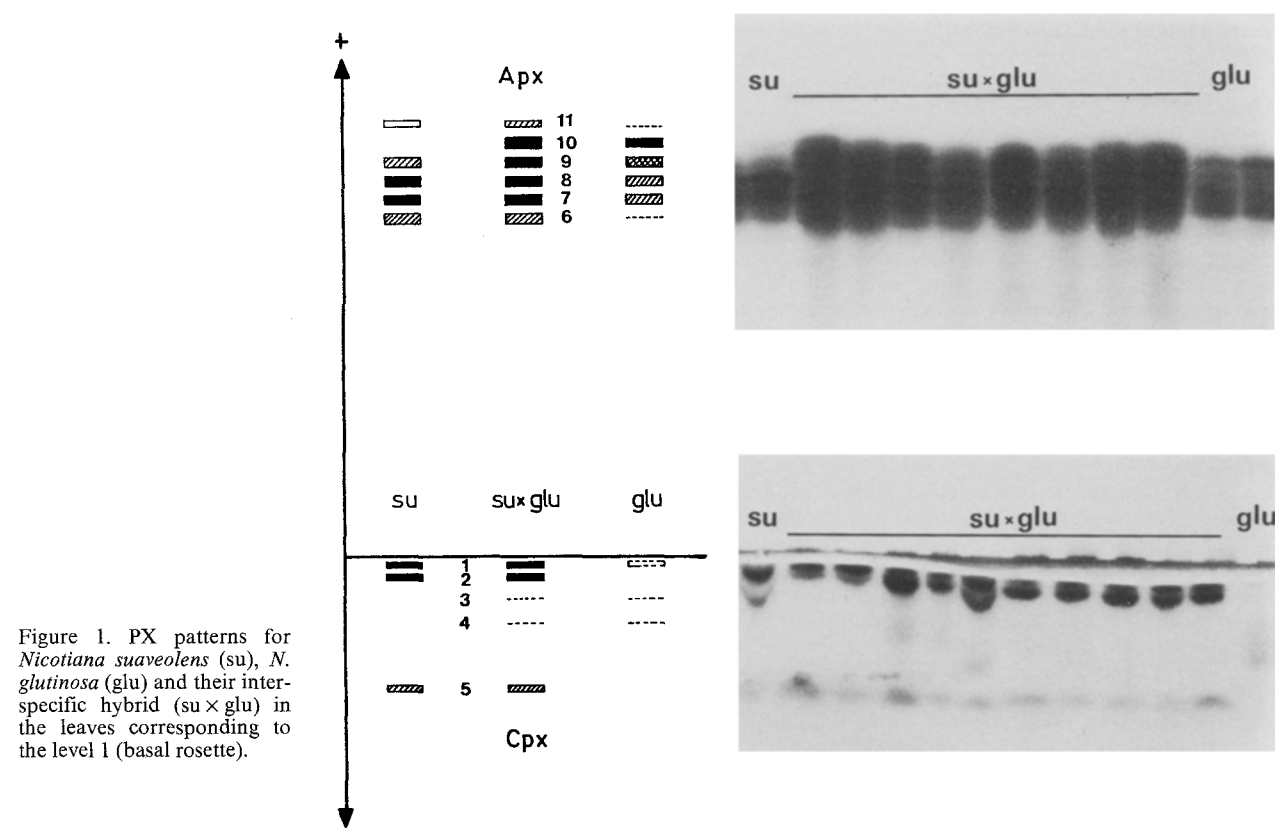


Figure 1. PX patterns for *Nicotiana suaveolens* (su), *N. glutinosa* (glu) and their interspecific hybrid (su × glu) in the leaves corresponding to the level 1 (basal rosette).

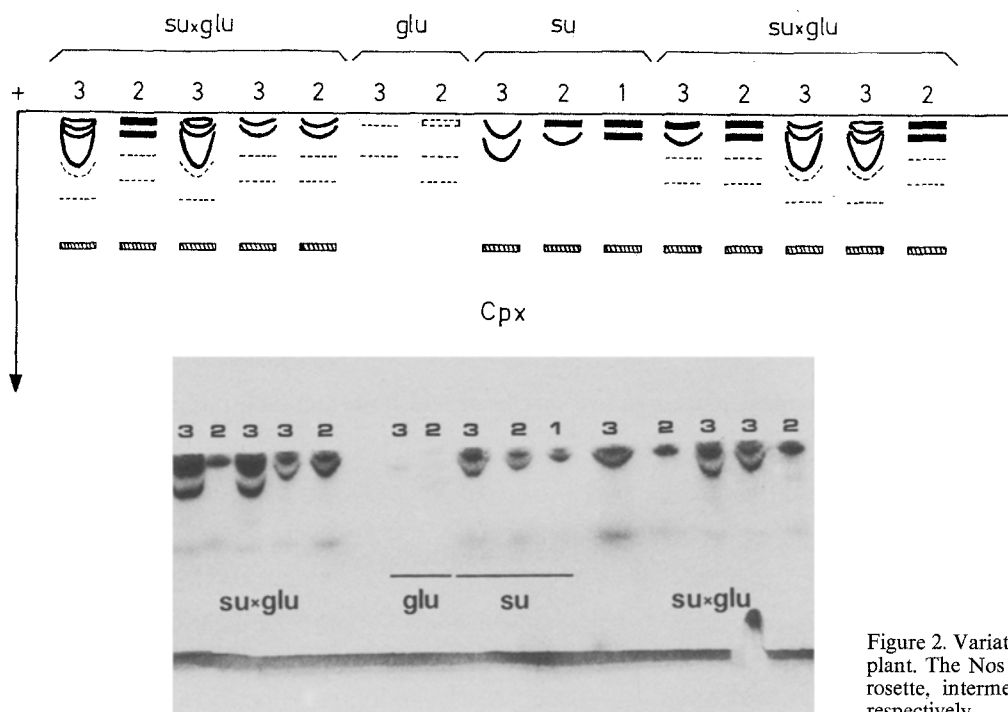


Figure 2. Variation of CPX pattern along the plant. The Nos 1, 2 and 3 refer to the basal rosette, intermediate, and flowering levels, respectively.

(Nos 6, 7, 8, 9 and 11) for *su* (fig. 1), these observations being coincidental with those of Sheen⁶ for the same species, the band number 10 of *su* being the only exception because in our case it was not detected. The hybrid plant reunites the bands of both parental species, showing monomeric isozyme behaviour which has previously been described not only in the genus *Nicotiana*^{4,6}, but in other species^{3,5,7,8}, because no hybrid isozymes were detected. Taking into account the monogenic control of each peroxidase isozyme in most of the plants studied, we assumed that at least 6 loci could be involved in the genetic control of the APX, with a fixed allele in each case, and with a null allele fixed in the locus responsible for the band number 10 in *su*.

In level 1 (basal rosette), the CPX pattern showed a total of 5 bands (Nos 1, 2, 3, 4 and 5) for *su* × *glu*, 3 bands (Nos 1, 2 and 5) for *su* and 3 bands (Nos 1, 3 and 4) for *glu* (fig. 1). The only difference from the pattern described by Sheen⁶ was seen in the band number 5, which he did not observe. Again, we observed monomeric isozymes, and a monogenic control of these can be assumed. We can put forward 2 alternative explanations: 5 loci could be implicated, each with a fixed allele, or only 4, if the bands No. 1 and 2 of *su* are controlled by 2 alleles of the same locus. In the last case *su* must be a fixed heterozygous species in spite of its autogamy, this being possible because it is an allopolyploid species, and, as Adams and Allard¹³ pointed out, allopolyploidy can maintain a 'fixed heterozygosity' in homozygous individuals because different alleles are fixed in different genomes.

The CPX pattern in the levels 2 (intermediate) and 3 (flowering) showed modifications of bands numbers 1 and 2 (fig. 2). The 3rd level leaves showed a distorted mobility, and half-moon shaped bands were noticed. They could be a consequence of post-translational modifications of the polypeptides already present. Thus, modifications of the PX pattern have been described in *Lycopersicon*¹⁰ and the existence of a gene which modifies the translational products of other ones has been postulated. There are other systems, such as the fraction-1 protein, in which post-transcriptional or post-translational modifications have also

been postulated¹¹. In our case we consider the possibility of the existence of some substances present or not in the different levels of the plant, which could modify the mobility of the No. 1 and 2 CPX peroxidases. Another possible explanation could be the existence of an hormonal gradient along the plant. It is known that IAA modifies the peroxidase pattern¹⁴ by the elimination of some isozymes. In our case we can assume that if the auxine concentration is higher in the upper part of the plant¹⁵, this level will be the one sharing most modification in the peroxidase pattern. We did not find band elimination but we did, however, find mobility modification of the peroxidase isozymes. No modifications of the isozymatic APX pattern along the plant have been observed.

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