Vitis vinifera - a chemotaxonomic approach: Pollen wall proteins

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Summary: The electrophoretic pattern of the pollen wall proteins from clones of cv. Nebbiolo grown in different areas show a geographical clustering. Vermentino, Pigato and Favorita are found virtually identical both by morphological and biochemical criteria.

Key words: pollen, cell wall, protein, Vitis vinifera, variety of vine, clone, ampelography, Italy.

Introduction

Pollen wall proteins as extracted by 0.2 M glycine give on pH 3.5-10 focusing range complex banding patterns with tens of components clustered in the acidic to neutral portion of the gradient (Cargnello *et al.* 1988). For cvs Cabernet and Merlot, the IEF pattern has been shown to be specific to individual clones and independent from the environment as well as from the growing conditions (Cargnello *et al.* 1988; Tedesco *et al.* 1989). The above approach was thus applied when addressing two problems in *Vitis* systematics.

Materials and Methods

For each sample 0.2 M glycine buffer extracts were obtained with 1 : 20 (w/v) ratio and adjusted to ca. 20-30 μg/lane.

Isoelectric focusing under native conditions of the pollen wall extracts was performed on a non-linear pH 3.5-10 gradient. Proteins were stained with silver nitrate according to Merril *et al.* (1981). Zymograms were developed for esterase (Coates *et al.* 1975), phosphoglucomutase (Spencer *et al.* 1964), acid phosphatase (Swallow and Harris 1972), alcohol dehydrogenase (Smith *et al.* 1971) and peroxidase (Taketa 1987).

After reduction with 2-mercaptoethanol, the subunits of the storage protein from the seed endosperm were resolved on a pH 4-6 gradient in presence of 8 M urea (Gianazza *et al.* 1989).

Results

Cultivar Nebbiolo is widely grown in the north-eastern part of Italy (Piemonte through Lombardia). When the banding patterns of the pollen wall proteins for clones from different areas were compared, a geographical clustering was obvious: Sassella, Valgella and Inferno from Valtellina gave identical bands, as did Bolla, Michet, Lampia and Rosé from Asti. Grumello was found to be more similar to the former group, Gattinara to the latter (Fig. 1). The same relationships were observed when the subunits of the seed storage proteins were analyzed by IEF in presence of 8 M urea after reduction with 2-mercaptoethanol (Fig. 2).

Cvs Vermentino, Pigato and Favorita are not easily distinguished by ampelographic criteria. Likewise, the banding patterns of their pollen wall proteins are very similar, with some quantitative variations only for the minor components (Fig. 3 A). Virtually identical results for the three samples are also obtained for the seed components – storage proteins (Fig. 3 B) as well as a number of

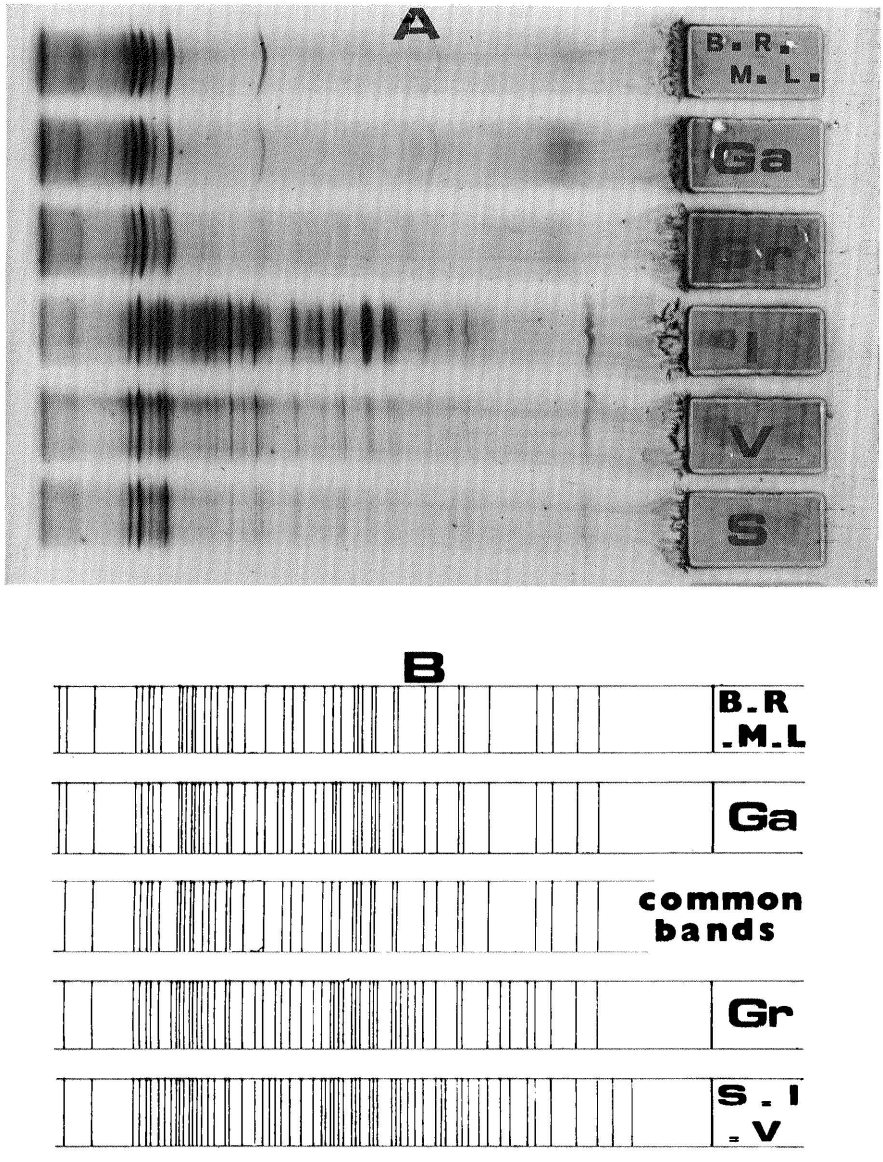

Fig. 1: IEF pattern of the pollen wall proteins o a non-linear pH gradient (range 3.5-10) under native conditions. Samples: Nebbiolo cultivars. Sassella (S), Valgella (V), Inferno (I), Grumello (GR), Gattinara (GA), Bolla (B), Rosé (R), Lampia (L), Michet (M), Barolo (BA). – A) Original gel. B) Schematic drawing.

enzymes (Fig. 3 C-G). These findings are in agreement with the hypothesis that the three clones do not correspond to distinct cultivars but to different names given to a single cultivar in different areas.

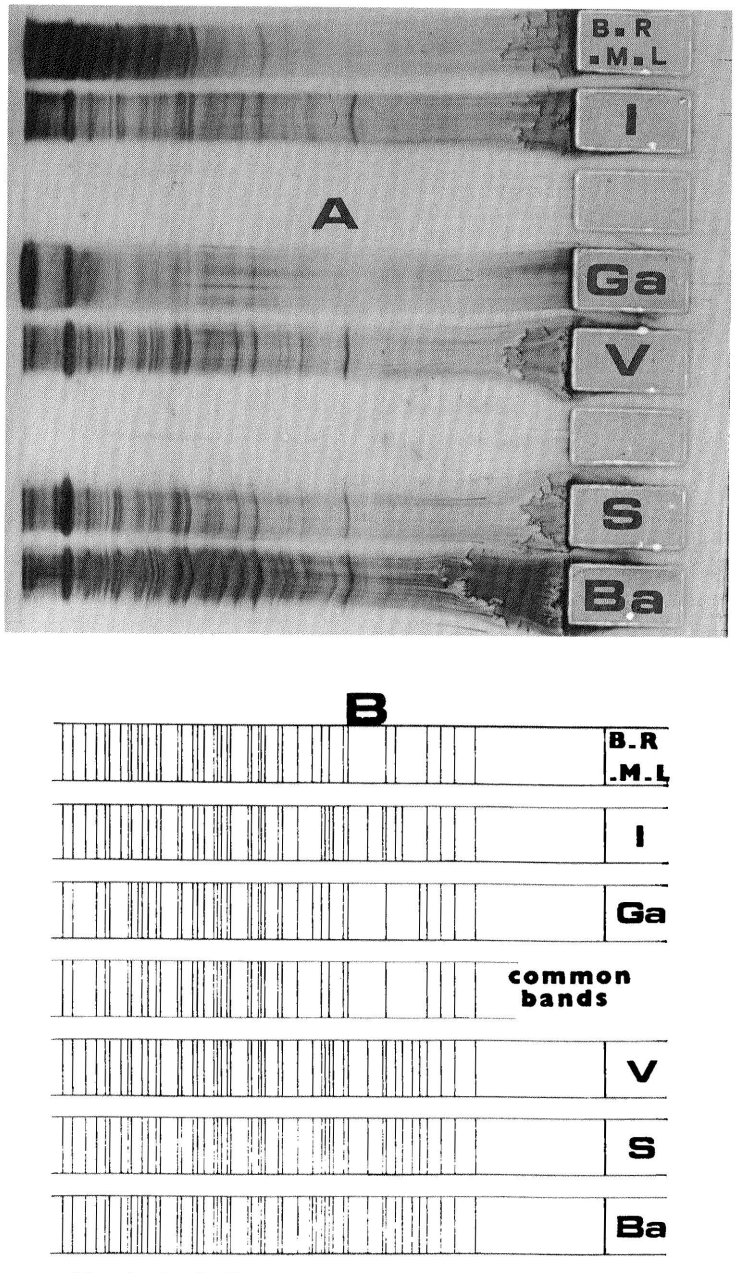

Fig. 2: IEF pattern of the subunits of pollen wall proteins on a non-linear pH gradient (range 3.5-10) in 8 M urea. Sample abbreviations as in Fig. 1. – A) Original gel. B) Schematic drawing.

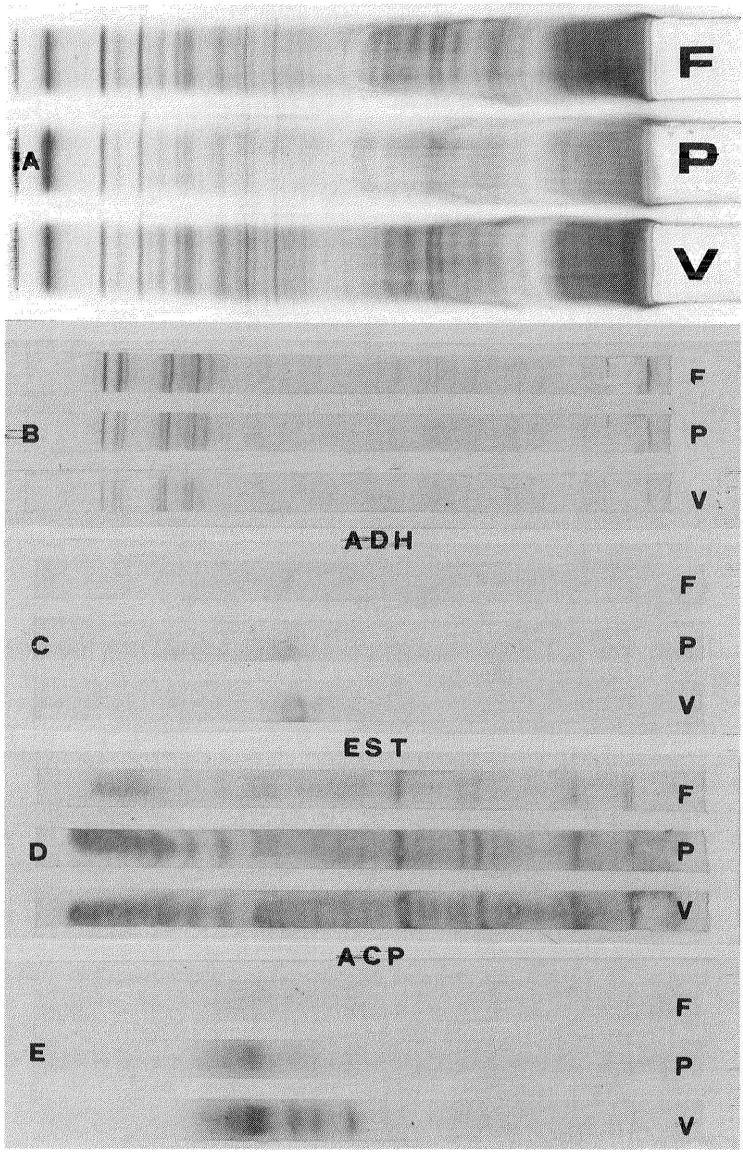

Fig. 3: Samples: Favorita (F), Pigato (P), Vermentino (V). – A) IEF pattern of the pollen wall proteins on a non-linear pH gradient (range 3.5-10). – B) IEF pattern of the subunits of seed proteins on a non-linear pH gradient (range 4-6) in 8 M urea. – C-G) IEF pattern of isoenzymes for seed extracts: C) Alcohol dehydrogenase (ADH) on pH range 4-10. d) Esterase (EST) on pH range 4-10. E) Acid phosphatase (ACP) on pH range 4.5-7. F) Peroxidase (POD) on pH range 4-10. G) Phosphoglucomutase (PGluM) on pH range 4-10. (Continued overleaf)

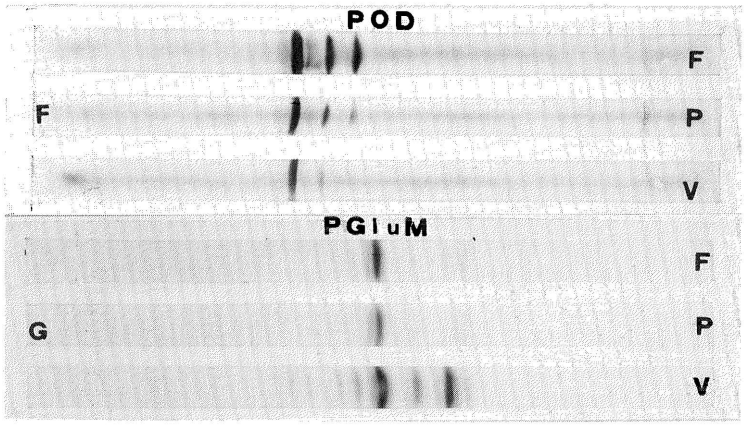

Fig. 3 (continued)

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