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## Studies on the embryo sac and on the stigmatic receptivity of *Vitis* cultivars with different productivity (Picolit giallo and Verduzzo friulano)<sup>1)</sup>

by

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### Untersuchungen über den Embryosack und die Bestäubungsreife der Narbe bei Rebsorten unterschiedlicher Ertragsleistung (Picolit giallo und Verduzzo friulano)

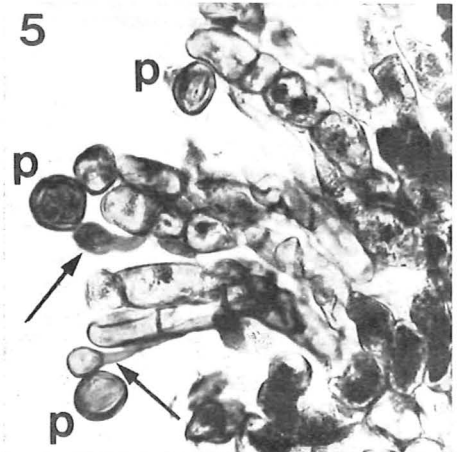
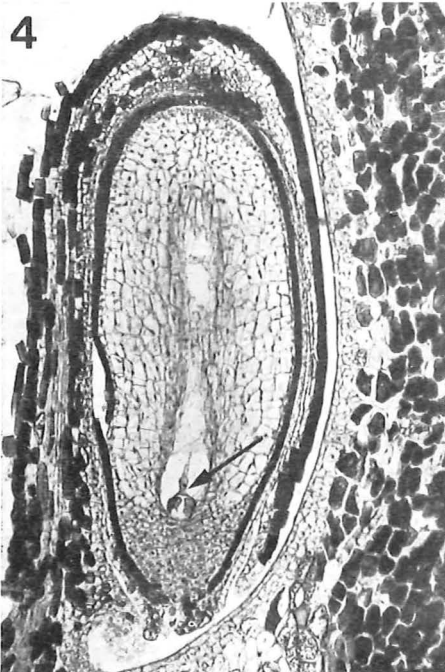
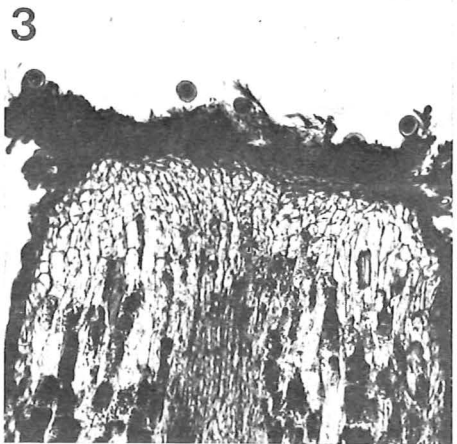
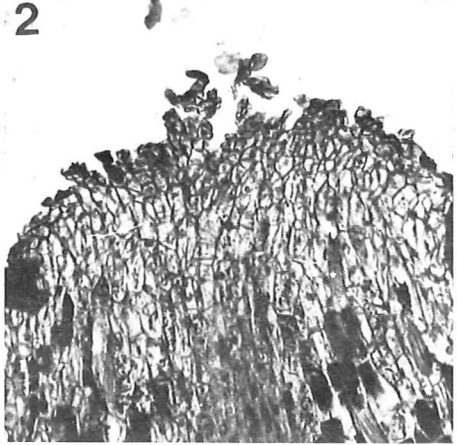
**Zusammenfassung.** — Die Rebsorte Picolit besitzt acolporate, d. h. ungefurchte und porenlose Pollenkörner, wodurch die Pollenkeimung verhindert wird. Da sich indessen die beiden untersuchten Picolit-Klone und ebenso die Haupt- und Geiztriebe eines Weinstockes in ihrer Ertragsleistung unterscheiden, wurden weiterführende Untersuchungen über die Entwicklung des Embryosackes und die Bestäubungsreife der Narbe angestellt. Die Befunde bei Picolit wurden mit dem Verhalten der normaltragenden Sorte Verduzzo verglichen. Die Samenanlage ist bei dem nahezu normaltragenden Picolit-Klon im Gegensatz zu dem schwachtragenden Klon normal entwickelt. Außerdem bestehen bei den Narben beträchtliche Unterschiede hinsichtlich der Dauer der Bestäubbarkeit. Dies könnte eine weitere Ursache für die unterschiedliche Ertragsleistung der untersuchten Rebsorten sein.

### Introduction

In previous studies (LOMBARDO *et al.* 1976, 1978 and in press) we have observed that pollen grains of *Vitis* are usually tricolporate showing three furrows with germinative pores; in contrast, all the examined pollen grains of Picolit possess a continuous sporopollenin wall, uniformly thick, without furrows and germinative pores; these features have been related with the low productivity of some clones of this cultivar (LOMBARDO *et al.* 1978). Nevertheless, since the productivity of Picolit varies among different vineyards and in the same plant between main branches and feathery, we have thought that other factors could influence the rate of fertilization of its clones. Presumably some of these factors could be represented by a different length of stigmatic receptivity, or by an anomalous development of the ovules, or by a restricted life period of the female gametophyte (MARRO and LALATTA 1978).

In the present paper we report our observations on the development of the female gametophyte and on the stigmatic receptivity of flowers of Picolit taken from clones characterized by a high and a low productivity. From this last clone we have also studied pistils taken from flowers of the feathery. The data obtained have been compared with the behaviour of the embryo sac and of the stigmatic surface of Verduzzo, a cultivar showing normal productivity.

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### Material and methods

We have examined pistils taken from flowers of main branches of: a) the high producing cultivar Picolit 31 A.A.U.<sup>1)</sup>, b) the low producing clone Picolit F.<sup>2)</sup>, c) the normal producing cultivar Verduzzo; and pistils from flowers of feathers of Picolit F.

The flowers were picked in subsequent times; the first samples were drawn at the time of the natural fall of the calyptra, the following after 1, 2, 4, 6, 10 d.

The samples have been fixed in Nawaschin, dehydrated in tertiary butanol and embedded in paraffin. 10  $\mu$ m thick longitudinal sections were dyed in Delafield hematoxylin and examined with a light microscope.

### Results

**Verduzzo:** At the fall of the calyptra all the ovules of the examined pistils appear normally shaped and contain normally developed embryo sacs (Fig. 1). At this time the stigma shows well developed distinct papillae; nevertheless, the papillae degenerate quite rapidly, so that the style apex already appears bare the second day after the fall of the calyptra (Fig. 2) or covered by a dark matrix enclosing the stigmatic papillae (Fig. 3). The amount of stigmata, showing degenerated papillae, increases rapidly and 4 d after the fall of the calyptra, the pistils of all the flowers have lost receptivity.

**Picolit 31 A. A. U.:** At the fall of the calyptra the pistil shows normally shaped ovules, all with evident and well developed embryo sacs. (Fig. 4). The stigmata

Fig. 1: Ovule of Verduzzo at the fall of the calyptra. In the middle of the nucellus the embryo sac appears well developed; some of the nuclei of the female gametophyte are clearly visible.  $\times 300$ .

Fig. 2: Stigma of Verduzzo, 2 d after the fall of the calyptra, already deprived of papillae.  $\times 100$ .

Fig. 3: Stigma of Verduzzo, 2 d after the fall of the calyptra, the papillae being no more distinct and enclosed in a dark matrix.  $\times 100$ .

Fig. 4: Ovule of Picolit 31 A.A.U. at the fall of the calyptra, containing a well developed embryo sac with an evident egg apparatus (arrow).  $\times 100$ .

Fig. 5: Stigmatic part of Picolit 31 A.A.U. at the fall of the calyptra; some germinating pollen grains (p) with evident pollen tubes (arrows) adhere on the stigmatic papillae.  $\times 350$ .

Abb. 1: Samenanlage der Sorte Verduzzo beim Abwerfen der Calyptra. In der Mitte des Nucellus ist ein wohlentwickelter Embryosack mit deutlich sichtbaren Zellkernen vorhanden.  $300 \times$ .

Abb. 2: Narbe der Sorte Verduzzo 2 d nach dem Abwerfen der Calyptra; die Papillen haben sich schon abgelöst.  $100 \times$ .

Abb. 3: Narbe der Sorte Verduzzo 2 d nach dem Abwerfen der Calyptra; Die Narbenpapillen sind nicht mehr erkennbar, da sie in eine dunkle Masse eingeschlossen sind.  $100 \times$ .

Abb. 4: Samenanlage der Sorte Picolit 31 A.A.U. beim Abwerfen der Calyptra. Der wohlentwickelte Embryosack enthält einen deutlich sichtbaren Eiapparat (Pfeil).  $100 \times$ .

Abb. 5: Narbenausschnitt der Sorte Picolit 31 A.A.U. beim Abwerfen der Calyptra. An den Narbenpapillen haften keimende Pollenkörner (p) mit deutlich erkennbaren Pollenschläuchen (Pfeile).  $350 \times$ .

<sup>1)</sup> These grapevines derive from the clone 31 A.A. and are cultivated in the wine farm G. B. Cragnoini near Cividale del Friuli (Udine) see LOMBARDO *et al.* 1978).

<sup>2)</sup> These plants are grown in the wine farm L. Felluga near Oleis (Udine).

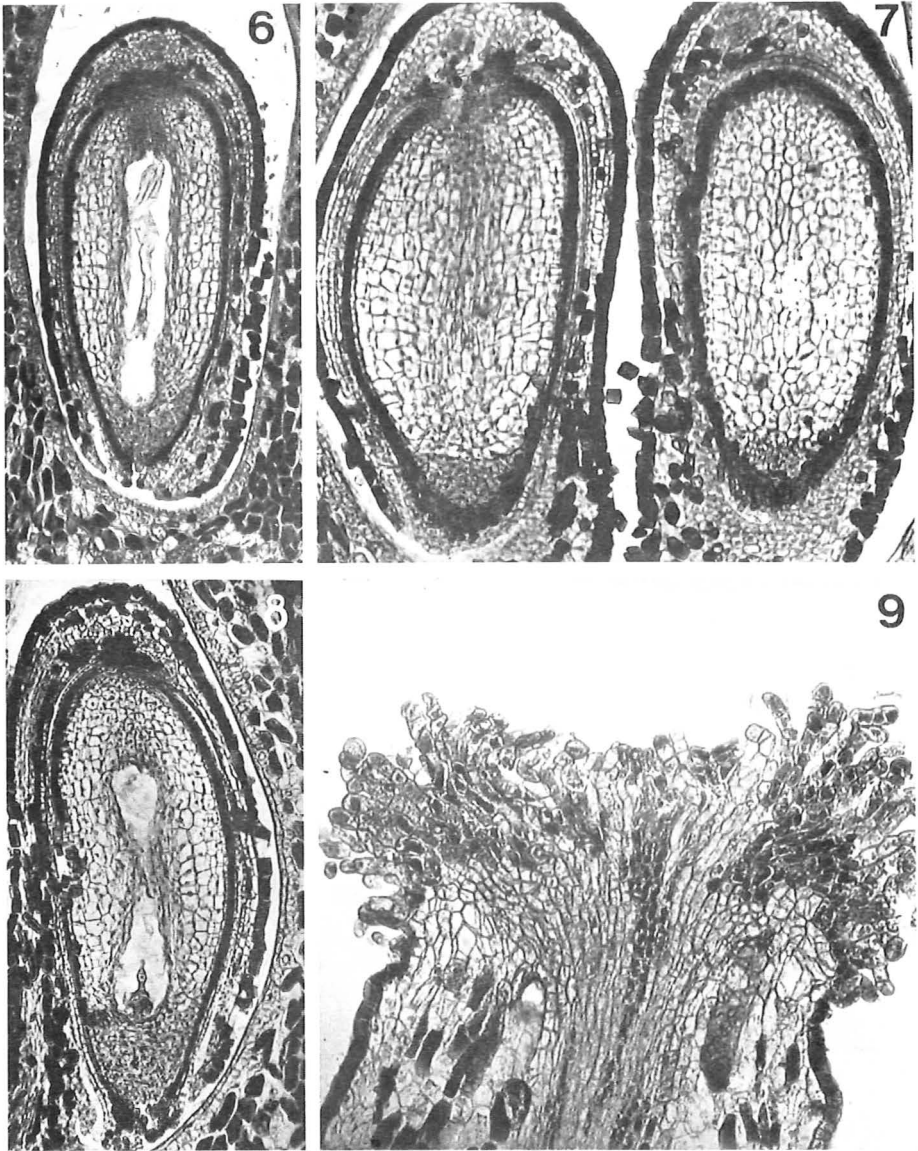


Fig. 6 and 7: Ovules from a main branch of Picolit F. at the fall of the calyptra, one containing a normally developed embryo sac, the others with a compact nucellus without embryo sac.  $\times 80$ ,  $\times 100$ .

Fig. 8: Ovule from a feather of Picolit F. at the fall of the calyptra. The nucellus contains a well developed embryo sac with an evident egg apparatus.  $\times 100$ .

Fig. 9: Stigma from a feather of Picolit F. at the fall of the calyptra showing distinct fan-shaped papillae.  $\times 100$ .

Abb. 6 und 7: Samenanlagen eines Haupttriebes der Sorte Picolit F. beim Abwerfen der Calyptra. Die erste Anlage enthält einen gut entwickelten Embryosack; die beiden anderen zeigen einen kompakten Nucellus ohne Embryosack.  $80 \times$ ,  $100 \times$ .

possess normally developed and fan-shaped papillae, sometimes in contact with germinating pollen grains (Fig. 5). The papillae persist unaltered until the 6th d; later on they begin to degenerate. On the 10th d, the stigma has lost functionality since its papillae appear sticky, adhering one another and enclosed in a filamentous matrix.

**Picolit F.:** At the fall of the calyptra the flowers of the main branches have pistils containing approximately 80% normally developed ovules only and nearly 20% ovules showing a nucellus without any embryo sac (Fig. 6 and 7). The stigmatic papillae appear normally developed and fan-shaped; very few pollen grains adhere to their surfaces. This condition persists until the 4th d, when the stigma begins to degenerate and its papillae appear sticky, adhering one another and enclosed in a dark matrix.

At the fall of the calyptra the flowers of the feathers possess pistils containing normally developed ovules, everyone having an evident embryo sac (Fig. 8). The stigma shows distinct fan-shaped papillae; few pollen grains are to be seen adhering to them (Fig. 9). This condition persists unchanged at least up to the 10th d.

### Discussion

We have already observed that pollen grains of Picolit show a continuous sporopollenin wall, without furrows and germinative pores as it usually occurs in pollen grains of *Vitis* (LOMBARDO *et al.* 1976, 1978 and in press). This anomalous structure of the pollen grains of Picolit does not represent the only reason of the low productivity of this cultivar. In fact, remarkable differences appear in the productivity of main branches and feathers as well as in the yield of various vineyards.

The present studies show that another limiting factor of the productivity of Picolit could be the anomalous development of the ovules. As a matter of fact, in flowers grown on main branches of Picolit F. nearly 20% of the ovules do not overcome the merely vegetative stage, according to the lack of meiosis and gametophyte formation. Such ovules that appear deprived of the embryo sac at the fall of the calyptra do not develop it even in the following days when the stigma could be still receptive. It is not possible to ascertain why a number of ovules do not fully develop; nevertheless, some physiological derangement could occur also related to the ratio of vital pollen grains present in the anthers. We are carrying out further investigations about this problem.

Another very important factor related to the productivity of various fruit trees (MARRO and LALATTA 1978) is the life period of the embryo sac and the relationship between this vitality and the length of the stigmatic receptivity. With reference to the first problem our observations do not allow to find any remarkable differences between the examined clones of Picolit. Regarding the receptivity period of the stigma, our data show that if the pollination is intense, through a great number of vital and germinable pollen grains, as it normally occurs in Verduzzo and in all the grapevines with hermaphrodite flowers, the stigmatic papillae degenerate in few

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Abb. 8: Samenanlage eines Geiztriebes der Sorte Picolit F. beim Abwerfen der Calyptra. Der Nucellus enthält einen wohlentwickelten Embryosack mit deutlichem Eiapparat. 100 ×.

Abb. 9: Narbe eines Geiztriebes der Sorte Picolit F. beim Abwerfen der Calyptra. Die fächerartig angeordneten Narbenpapillen sind deutlich zu erkennen. 100 ×.

days. On the other hand, when we lack pollen germination or this one is extremely reduced, as it occurs in Picolit or when the pollination is less massive than that of self-fertile flowers, the stigmatic papillae maintain their normal structure even for several days. We are still carrying out experiments on the effects of castration; we could already ascertain that, if the stamens are early removed to prevent self-pollination, the stigmatic papillae persist vital for a longer period also in Verduzzo. Moreover, our observations have shown that the clone of Picolit, characterized by a productivity close to normality (Picolit 31 A.A.U.), has a more prolonged stigmatic receptivity than the clone at low productivity (Picolit F.). In this clone the higher productivity observed in flowers grown on the feathers seems also related to a longer receptivity of the stigmata, that maintain indeed a normal aspect even 10 d after the fall of the calyptra.

Further investigations are needed to ascertain whether pollination by germinable or non-germinable pollen grains could influence the receptivity of the stigma; nevertheless, it is already possible to say that in plants characterized by a low productivity the stigmatic receptivity period under the same conditions appears reduced when compared with that of plants with high productivity.

### Summary

The cultivar Picolit is characterized by acolorated pollen grains. Nevertheless, since the productivity of Picolit varies between the two examined clones and between main branches and feathers of the same plant, we extended our researches to the development of the embryo sac and to the length of the stigmatic receptivity of such clones. The results have been compared with the behaviour of Verduzzo, a cultivar with normal productivity. The development of the ovules is normal in the clones of high productivity, while it appears anomalous in that of low productivity; besides, remarkable differences appear among the various stigmatic receptivities. This fact could be a further reason for the different rates of productivity of the examined grapevines.

### Literature cited

- LOMBARDO, GIULIANA, CARRARO, LUISA, CARGNELLO, G. and BASSI, MARIA, 1976: Ultrastructure of pollen of *Vitis vinifera* L. cv. "Picolit giallo" and its behaviour in experiments of self- and cross-pollination. *Vitis* 15, 73—81.
- —, CARGNELLO, G., BASSI, MARIA, GEROLA, F. M. and CARRARO, LUISA, 1978: Pollen ultrastructure in different vine cultivars with low productivity. *Vitis* 17, 221—228.
- —, CARRARO, LUISA, CARGNELLO, G. and GEROLA, F. M.: Pollen morphology of Picolit grown in different Italian regions. In press.
- MARRO, M. and LALATTA, F., 1978: Osservazioni sulla durata e vitalità del sacco embrionale del melo in rapporto alla allegazione. *Atti del Seminario sulla Fertilità della Pianta da Frutto*, Bologna, 15 dic. 1978, 637—642.

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