



## Title: "A technique for screening grape germplasm for resistance to *Meloidogyne incognita*"

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Breeding programs aiming at developing new nematode-resistant rootstocks are important because current rootstocks with adequate nematode resistance tend to have other undesirable viticultural characteristics (excessive vigor, excessive potassium uptake, poor resistance to phylloxera, rooting difficulty, inadequate zinc absorption). In this paper the authors develop a technique for evaluating resistance to root-knot nematode (*Meloidogyne incognita*) in grape seedlings that will promise to fastforward what used to be a very slow process.

- Until now, to screen grape progeny for potential nematode resistance, seedlings from rootstock crosses had to be allowed to grow for several years in order to provide sufficient cuttings for planting in a test vineyard. Subsequently, plants had to be allowed up to four months of nematode feeding, after which roots were examined and individual nematode eggs -or nematode juveniles- were painstakingly counted under a microscope. Lower viability numbers were interpreted as higher resistance.
- Since root galls (large masses of undifferentiated root tissue containing nematodes) are one of the main symptoms of root-knot nematode infection, counting the number of galls was originally proposed as a means to estimate resistance. But varieties that do not form galls, yet may be highly infected, have since been detected. So it soon became apparent that *galling ability* could not be used as an indication of nematode resistance. Instead, measuring actual nematode *reproduction ability* (for example, number of eggs) seemed unavoidable.
- The authors observed that infected vines that do not form galls actually showed very obvious masses of eggs that could be seen without a microscope. Perhaps these egg masses—they reasoned-correlate with the degree of nematode reproduction and could be used to evaluate resistance.
- To test this theory, the authors infected cuttings of both nematode-resistant cultivars (*Vitis* hybrid Harmony) and nematode-susceptible cultivars (*Vitis vinifera* cv. Colombard) with a population of root-knot nematodes that they had been maintaining in tomato plants. They did this by chopping up the tomato roots, extracting the juvenile nematodes from the roots, and placing them in the soil next to the potted test cultivars. A few weeks after inoculation, they counted both the **individual eggs** (traditional method) and the "**masses of eggs**" (new proposed method) to see if a good correlation existed between the two. To facilitate the counting of the egg masses, they stained the roots first with a specific dye (eosine).

- Besides testing **cuttings** of full-grown plants, the authors also tested young grape **seedlings**. The seedlings tested originated from both susceptible open-pollinated varieties, as well as different crosses between susceptible and resistant rootstocks.
- When the authors compared number of eggs and number of "egg masses", they found a nearly perfect correlation (r=0.96), both when they used cuttings and when they used seedlings. Only one cross (Harmony x 3309C) had a poor correlation, which they attributed to the small number of egg masses. As expected, and confirming the good nematode resistance of the Harmony cuttings, none of the vines of this variety showed any egg masses. Similarly, Colombard susceptibility was confirmed by the presence of significant egg masses (80-200) in every cutting examined.

The value of this paper is to provide a fast and simple technique to evaluate actual nematode infection (not their symptoms) in grapevine roots. By validating their technique for use with immature seedlings, not only mature cuttings, the authors are providing a valuable tool that will be of great help in future breeding efforts. They can now cross rootstocks with high resistance to nematodes with rootstocks of desirable viticultural properties and use this simple staining technique to screen for the best nematode resistance among the progeny, without waiting for those seedlings to grow. A technique seemingly only of interest to the researchers, but that benefits the growers in the long run.

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