## LETTER TO THE EDITOR

## Thermotherapy sanitation of two grapevine cultivars

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Two grapevine cultivars Müller-Thurgau and Portugieser Blau were selected for a thermotherapy at the onset of sanitation program in Czech Republic. These cultivars represent tradition and a good quality in wine making. Currently, the area in Czech Republic planted with Müller-Thurgau and Portugieser Blau is 1975.7 and 676 ha, respectively.

From these cultivars we selected 5 promising clones that produced stable yields and a good quality wine. Examination of the 5 clones for the presence of viral infections revealed the presence of grapevine leafroll-associated virus 1 (GLRaV-1), arabis mosaic virus (ArMV), grapevine fanleaf virus (GFLV), grapevine fleck virus (GFkV), and rupestris stem pitting-associated virus (RSPaV). All tests were done using RT-PCR. The primers for ArMV and GFLV were used according to our previous work (3), and primers for GLRaV-1, GFkV, and RSPaV were used according to our study on multiple virus infection (4).

Cuttings taken from the mother plants were used for growing of new plantlets. After proper rooting and growing of the new shoots in pods, the new plants were placed in a climate chamber for thermotherapy that was conducted for 45 days at 37°C. The light conditions were 16/8 hours day/night. This procedure resulted in the onset of a new growth from the axillar buds. This effect usually called rejuvenation

created a good chance for the initiation of propagation in vitro. Topical and axillar meristematic tissues were taken from the new shoots and cultivated in vitro. The optimal composition of cultivation medium differed for the cultivars tested. Müller-Thurgau grew best on C2D medium (1), while Portugieser Blau preferred modified WPM medium (6). Both media contained 20 g/l sucrose. Agar (Duchefa) concentration was 6 g/l (pH = 5.6). After cultivation of the explants, the new plantlets were separated, rooted, and cultivated in a greenhouse at non-sterile conditions. The surviving plants were tested by RT-PCR for the presence of viruses detected before sanitation. After thermotherapy, the examination of plants was repeated annually for 3 years (Table 1). Only one plant of each cultivar was found to be free of all viruses after the thermotherapy: one Müller-Thurgau cultivar, clone MT 26/19, and one Portugieser Blau cultivar, clone PM 11/48.

As indicated in Table 1, the effectiveness of virus removal by thermotherapy was low. RSPaV was especially resistant to the thermotherapy procedure. Problems with the removal of RSPaV during the sanitation procedure were reported earlier (2, 5). RSPaV is a phloem-associated virus and such viruses are often resistant to the thermotherapy procedure (7). On the other hand, nepoviruses such as the GFLV and ArMV were relatively successfully removed.

In summary, our study documented the relatively low efficiency of thermotherapy in the sanitation of grapevine infected with several viruses.

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**Abbreviations:** GLRaV-1 = grapevine leafroll-associated virus 1; ArMV = arabis mosaic virus; GFLV = grapevine fanleaf virus; GFkV = grapevine fleck virus; RSPaV = rupestris stem pitting-associated virus

Table 1. Thermotherapy of the grapevine cultivars

Cultivar	Clone	No. of tested plants	Detected viruses before thermotherapy	Detected viruses after thermotherapy	No. of sanitized plants
Müller-Thurgau	MT 23/37	10	GFLV, GLRaV-1, RSPaV	GFLV, RSPaV	0
	MT 26/19	25	GFLV, GLRaV-1, RSPaV	GLRaV-1, RSPaV	1
	MT 33/16	15	GFLV, GLRaV-1, RSPaV	GLRaV-1, RSPaV	0
Portugieser Blau	PM 11/48	15	ArMV, GLRaV-1, RSPaV, GFkV	RSPaV	1
	PM 30/40	10	GFLV, GLRaV-1, RSPaV, GFkV	GLRaV-1, RSPaV, GFkV	0

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