

# Certification programme for production of virus-free propagating material of grapevine and its results in Hungary

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**Summary:** In Hungary, detection of virus and virus-like diseases of grapevine began in 1960's at the Research Institute for Viticulture and Enology by János Lehoczky and his colleagues. At present, sixteen virus and virus-like diseases of *Vitis vinifera* are known to occur in Hungary.

Regular virological screening of grapevine varieties started in 1972. The present system of screening (visual selection, indexing, ELISA) has been established using methods with continuous improvement according to recommendations of international organizations.

In the first year symptomless grapevine plants are selected and marked during surveys carried out twice in the vegetation period: at about flowering and in the second half of September. At the first selection time plants are sampled for ELISA.

In the spring of the second year, overwintered canes are checked by woody indexing on 8 indicator species in the field.

In the third and fourth years the nursery is evaluated twice again. At the end, the marked grapevine plants, giving negative results on all indicators in every case, are considered virus-free.

In autumn of the fourth year, the virus-free material is planted out under screenhouse and also in a special mother block (nuclear stock) for maintenance and propagation.

Mother blocks of virus-free scion varieties have been established on 2 ha and those of rootstock varieties on 0.5 ha planted with the following number of varieties included in the national list: 71 European scion – and 12 rootstock varieties or variety candidates/clones. It is necessary to increase the area of Pre-base, Base and Certified stocks exclusively with tested virus-free (clean) material.

**Key words:** grapevine, certification, virological screening methods, virus-free varieties

## Introduction

In Hungarian viticulture the use of clones developed from cultivated grapevine varieties is well justified in order to establish true to type, healthy and productive plantations. The use of virus-free propagating material is an important factor to improve quality and volume of grape production.

In Hungary the detection of virus and virus-like diseases of grapevine began in 1960's in the Research Institute for Viticulture and Enology by János Lehoczky and his colleagues. At present, sixteen virus and virus-like diseases of *Vitis vinifera* are known to occur in Hungary (Lehoczky et al., 1992; Lázár et al., 1993; Kölber et al., 1997). Some of these viruses, for example fanleaf and leafroll cause significant yield loss and/or lower fruit quality. Other virus diseases, for example, Rugose wood complex can provoke ultimately death of grapevine stocks. A few viruses are latent. Little is known about their effects on grapevine, however occurrence of these diseases is quite frequent, so they may have high economic importance.

Fanleaf virus (Figure 1) and the related strains (Yellow mosaic and Veinbanding) are most widespread and present

in all vine-growing regions of Hungary. Other nepoviruses: Arabis mosaic, Chrome mosaic, Bulgarian latent, Tomato black ring are not so frequent. Symptoms of Enation, Yellow mottle, Line pattern were observed only in one or two cases in the grapevine-growing regions. Rugose wood complex



Figure 1 Fanleaf – chronic symptoms on FS-4 indicator



Figure 2 Kober stem grooving – basipetal pitting and grooving on the surface of woody cylinder of T.K. 5BB indicator (left the healthy control)



Figure 4 Phytoplasma symptoms on *Vitis vinifera* Chardonnay



Figure 3 Fleck – clearing of the veinlets of *Vitis rupestris* St. George indicator

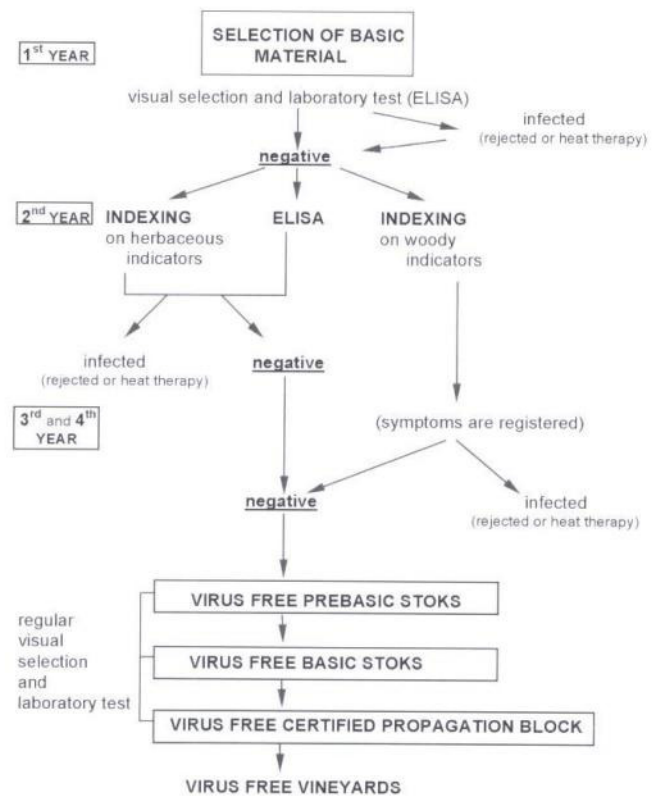


Figure 5 Procedure of virological screening of grapevine in Hungary

(Figure 2), Leafroll and Vein mosaic are widely distributed in almost all the main grapevine-growing regions of Hungary, affecting the major table and wine varieties. Fleck (Figure 3) and Vein necrosis can be often found on the indexed varieties and rootstocks, with incidence varying from 50 to 80%. Yellows disease was observed in several grapevine-growing regions of Hungary and identified as phytoplasma (Figure 4) belonging to subgroup 16 SrI-G (stolbur and related phytoplasmas).

Regular virological screening of grapevine varieties started in 1972 (Lehoczky et al., 1992). The present system of screening (Figure 5) has been established using methods (visual selection, biological indexing, ELISA) with continuous improvement according to recommendations of international organizations (Anonymous, 1992; Neszmélyi et al., 1996).

## Material and method

In the first year, symptomless grapevine stocks are selected and marked. Surveys are carried out twice in the vegetation period (at about flowering and in the second half of September). At the time of the first selection, sampling is performed for ELISA. Since 1985 ELISA has been routinely applied for the detection of 7 viruses/strains: GFLV, GFLV-YM, GFLV-VB, ArMV, GCMV, ToBRV, AIMV. Since spring 1993 raspberry ringspot and strawberry latent ringspot viruses have also been serologically screened. In November canes of symptomless and ELISA-negative plants are collected for further investigations and stored in plastic bags at 2-3 °C in cold room.

**Table 1** Woody indicators for the identification of virus- and virus-like diseases of the grapevine in Hungary

Indicator	Disease identified
1. Siegfriedrebe (FS4 201-39)	Fanleaf and strains Arabis mosaic Bulgarian latent
2. <i>Vitis rupestris</i> St. George	Arabis mosaic Fleck Rupestris stem pitting
3. <i>V. vinifera</i> cv. Pinot noir and other red-berries cv.	Leafroll Chrome mosaic Tomato black ring Arabis mosaic Yellow mottle Line pattern
4. <i>V. vinifera</i> cv. Chardonnay	Fanleaf and strains Yellow mottle Tomato black ring Grapevine yellows
5. <i>V. riparia</i> Gloire	Vein mosaic Bulgarian latent Line pattern
6. <i>V. berl. x V. rip.</i> Kober 5BB	Kober stem grooving
7. LN 33 (Coudere 1613 x <i>V. berl.</i> )	LN 33 stem grooving Corky bark
8. <i>V. rup. x V. berl.</i> 110 R	Enation Vein necrosis

**Table 2** Virus-free grapevine varieties in Hungary until 2002

Varieties for white wine	
Arany sárfehér (2)	Leányka E.99 (5)
Bianca (66)*	Magyar rizling (2)
Chardonnay 75 (42)	Nektár (71)**
Cserszegi fűszeres (2659)**	Olasz rizling B.20 (79)**
Ezerfürtű (11)	Piros veltelíni (8)
Ezerjő (123)**	Rajnai rizling B.7 (1)
Furmint T.85 (258)**	Rajnai rizling Gm.239 (3)
Furmint T.92 (123)**	Rajnai rizling 391 (18)
Göcseji zamatos (64)	Rajnai rizling Bernk.68 (4)
Hárslevelű K.9 (115)**	Rizlingszilváni K.3 (1)
Hárslevelű P.41 (8910)**	Rizlingszilváni (10)
Hárslevelű T.311 (134)**	Rizlingszilváni D.100 (8)
Jubileum 75 (10)	Rozália (1)
Juhfark (2)	Sauvignon blanc (10)
K. 15 (2123)**	Szürkebarát B.10 (8)**
K. 35 (33)	Szürkebarát 34 (1)
Karát (6)	Zala gyöngye (1345)*,**
Kerner (5)	Zengő (10)
Királyleányka 21 (2)	Zéta (4)**
Korai piros veltelíni (9)	Zeus (2)
Kövidinka K.8 (31)	Zöld veltelíni La.10/83 (14)
Varieties for red wine	
Blauburger (2)	Kékoportó Kt.1 (10)
Cabernet franc E.11 (1)**	Medina (69)*
Cabernet sauvignon E.153 (22)	Merlot (9)
Kadarka P.9 (16)	Pannon frankos (39)
Kármin (5)	Pinot noir M.2 (9)
Kékfrankos G.379 (2)**	Pinot noir P.1
Kékfrankos Kt.1 (2)	Turán (1)
Kékmedoc (2)	Zweigelt (123)**
Table grape varieties	
Boglárka (9)	Nero (426)*
Cegléd szépe K.73 (2)	Pannónia kincse K.56 (19)
Favorit (2)	Piros chasselas Fr.36-28 (15)
Fehér chasselas Fr.38-95 (46)	Piros chasselas K.18 (1)
Fehér chasselas K.16 (12)	Pölöskei muskotály (63)*
Hamburgi muskotály (11)	Teréz (4)*
Irsai Olivér K.11 (26)**	
Rootstock varieties	
Fercal (25)	Teleki-Kober 125AA (147)
Georgikon 28 (7)	Teleki-Kober 5BB (F-21)
Teleki 5C (E-20)	Teleki-Kober 5BB Cr.2-K.18 (18)
Teleki 5C Gm.6-K.64 (64)	Teleki-Kober S.O.4 (133)
Teleki 5C Gm.10-K.74 (74)	Riparia portalis (VT)
Teleki 5C Wed.-K.103 (103)	Rupestris du Lot (VT)
Number of other virus-free varieties and clones 42	
Note: * = interspecific varieties; ** = heat-treated varieties	

In the spring of the second year, overwintered canes are checked by woody indexing on 8 indicator species in the field: FS 4, *Vitis rupestris* St. George, *V. vinifera* cv. Pinot noir, *V. vinifera* cv. Chardonnay, *V. berlandieri* x *V. riparia* Kober 5BB, Couderc x *V. berlandieri* LN 33, *V. riparia* Gloire, *V. rupestris* x *V. berlandieri* 110 R. In the present system FS 4 and Chardonnay are regularly used but they will be omitted or only occasionally used in the future (Table 1). Symptoms are recorded in June and September. In the spring of the second year overwintered canes are occasionally checked also by mechanical transmissions onto herbaceous indicator plants: *Chenopodium quinoa*, *C. amaranticolor*, *Cucumis sativus* „Delicates”, *Gomphrena globosa*, *Nicotiana clevelandii*, *N. tabacum* „Samsun”, *N. glutinosa*, *Phaseolus vulgaris* „Beautiful”.

In the third and fourth years the nursery is evaluated twice again. At the end, the marked grapevine plants, giving negative results on all indicators in every case, are considered virus-free. If there are varieties from which it is not possible to select healthy plants, their cuttings are rooted and heat-treated or adapted to *in vitro* culture for the production of virus-free progenies. After heat treatment they are re-tested for viruses and trueness-to-type.

In autumn of the fourth year the virus-free material is planted out under screenhouse (3–4 plants/variety) and also in a special mother block (nuclear stock) (30 plants/variety) for maintenance and propagation. Plants of the nuclear stock (Pre-base) produce propagating material (basic), which will be planted out in propagation stocks. The progeny of basic material originating from the propagation stocks is used for

nursery propagation. Propagating material derived from mother grapevines established in nurseries is delivered to the growers as certified material.

During these propagation steps, visual observation and random tests by ELISA are done to monitor the virus status of the plants. Propagation is performed under strict official control by the national Plant Protection Service. Trueness-to-type is also monitored by the inspectors of the National Institute for Agricultural Quality Control.

## Results

Mother blocks of virus-free scion varieties have been established on 2 hectares and those of rootstock varieties on 0.5 ha including the following number of varieties included in the national list: 71 European scion – and 12 rootstock varieties or variety candidates/clones. Number of other virus-free varieties and clones: 42 (Table 2). There are 167.8 ha of virus-free Pre-base Base and Certified stocks from 71 European scion varieties and 98.4 ha from 12 rootstock varieties (Figure 6). It can be stated that the selection, clone propagation and maintenance of the virus-free stocks intended for propagation and nurseries meet the EU standards. In Hungary the existing area is not sufficient to produce propagating material for the renewal of all plantations. So it is necessary to increase the area of Pre-base, Base and Certified stocks using virus-free (clean) material.

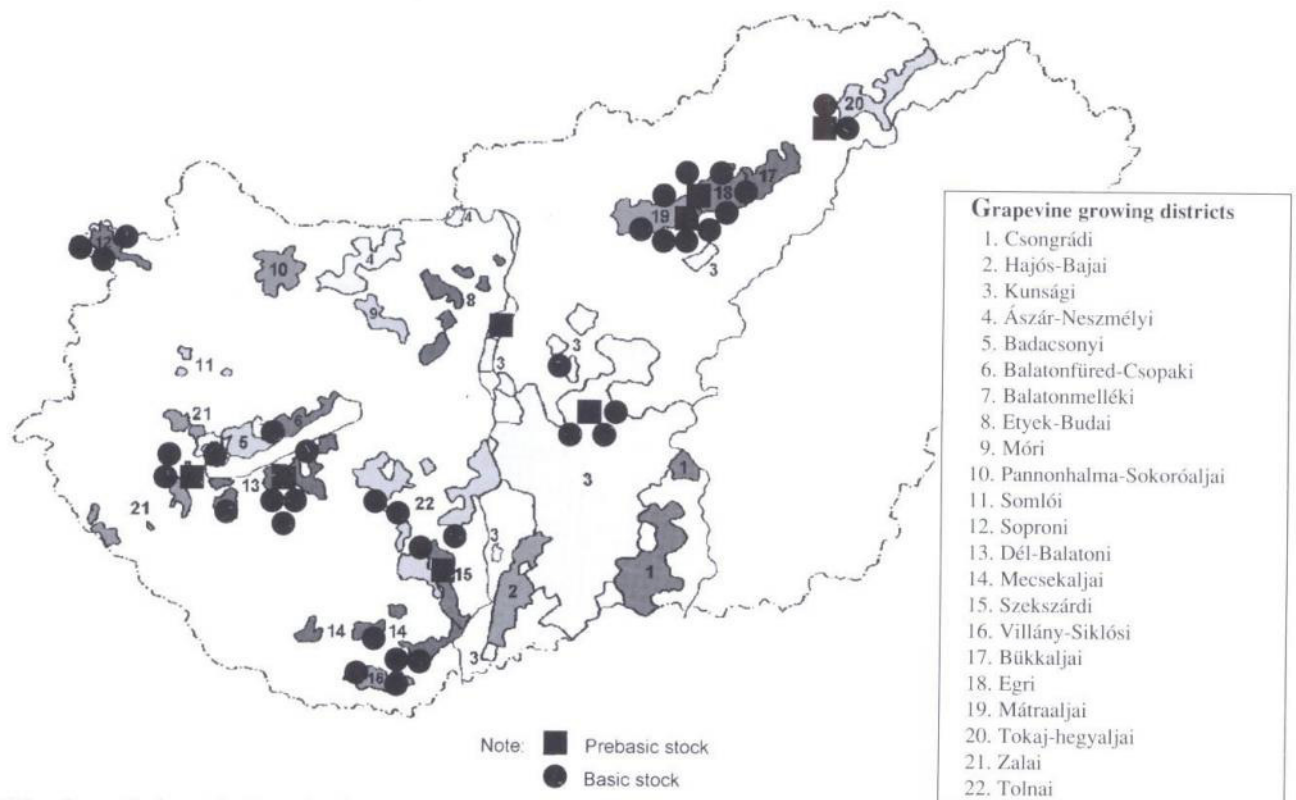


Figure 6 Virus-free prebasic and basic stocks of grapevine in Hungary

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