## Elimination of virus diseases by in vitro culture

G. STAUDT and H.-H. KASSEMEYER

Staatliches Weinbauinstitut Freiburg, Merzhauser Str. 119, D-7800 Freiburg, F. R. Germany

A b s t r a c t: Arabis mosaic virus (ArMV) and raspberry ringspot virus (RRV) are causal agents of grapevine fanleaf disease, one of the most damaging virus diseases of grapevines occurring in Germany. A simple method has been developed to eliminate the viruses using *in vitro* propagation of single nodes.

Nodes from ArMV or RRV-infected vines were micropropagated to complete plants and cultivated in a modified Murashige-Skoog medium. The position of individual nodes on their mother plant was documented. For any new subculture, the plants were dissected to single nodes when they reached a certain length. These were cultivated separately. After three *in vitro* subculturings, the plants were transferred to pots with a substrate containing a disinfected loess/sand mixture and cultivated under greenhouse conditions.

After each *in vitro* subculture and 18 months after the beginning of the *in vitro* culture, the resulting plants were tested using ELISA for the presence of ArMV and RRV, respectively. After the first and second subculture, a certain number of plants already showed no detectable viruses. After 18 months, no correlation could be detected between the origin of the plant from any particular node and virus elimination.

The results suggest that this *in vitro* culture method makes elimination of ArMV and RRV possible within 18 months without any thermotherapy.

## Agrobacterium-mediated transformation of grapevine (Vitis vinifera L.)

 $V.~Guellec^{-1}$ ), C.~David,~A.~Petit,~J.~Tempe~and~M.~Branchard

1) L.A.P., UA 115, Bt 360, Université de Paris-Sud, F-91405 Orsay Cedex, France

A b stract: Genetically transformed grapevine (*Vitis vinifera* L.) roots were obtained after inoculation of *in vitro* grown whole plants (cv. Grenache) with *Agrobacterium*. The strain used contains two independant plasmids: the wild type Ri-plasmid pRi 15834 (agropine type) and a Tiderived plasmid which carries the neomycin phosphotransferase II gene (NPTII) and the nopaline synthase gene. Expression of the NPTII gene can confer kanamycin resistance to transformed plant cells. Axenic root cultures derived from single root tips were obtained. Opine analysis indicated the presence of agropine and/or nopaline in established root cultures. For one of the cultures the presence of Ri and Ti derived DNA was confirmed by Dot-blot hybridizations of genomic root DNA with pRi 15834 TL-DNA and the NPT II gene. Callogenesis was induced by subculturing root fragments on Murashige and Skoog (1962) medium supplemented with benzylamino purine (0.2 mg/l) and indoleacetic acid (0.5 mg/l).