Sanitary certification of grapevine: European network for the establishment of reference protocols for detection of infectious agents

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The infection of most cultivated grapevine varieties by different viruses and virus-like diseases results in severe losses. Today, the only way to control, to some extent, the detrimental effects of grapevine viruses is sanitary selection and certification. When the natural vectors of the viruses are known (nematodes, mealybugs), chemical control of vector populations can more or less efficiently restrict the spread of the viruses.

The objective of sanitary selection is to propagate clones which are not infected by harmful viruses and to protect them as long as possible from infection in the mother blocks for multiplication.

The effectiveness of the sanitary selection relies on the sensitivity and the reliability of the techniques used for the detection of viruses.

The EEC Directive 68/193 on the "marketing of vegetatively propagated material of grapevine", then modified by Directives 71/140 and 77/629, contains indications encompassing the sanitary characteristics of the mother vineyards destined for the production of propagating material. Unfortunately the sanitary provisions of these directives are unsatisfactory and far from ensuring an acceptable sanitary status of propagative material of any category. Furthermore, they are no longer adequate, failing to take into account recent scientific acquirements in grapevine virology. Much progress was made recently in the aetiology, epidemiology and diagnosis of grapevine viruses and virus-like diseases.

The vagueness of the directives in phytosanitary matters reflects in the laws stemming from them that were promulgated in the six EU member states with major viticultural industries, i.e. France, Germany, Greece, Italy, Portugal and Spain. Moreover, enforcement of the directives varies from country to country because of differences in the type and number of diseases regarded as undesirable in national certification schemes, and in the methods for their detection.

These differences are prejudicial to the sanitary standardisation of propagative material of *Vitis* produced in the EU, and impair its free circulation within the Union, much to the concern of producers (nurserymen) and users (growers).

Different meetings, held in Gorizia (Italy) in December 1990, in Valenzano (Italy) in March 1991 and in Colmar (France) in June 1991, were carried out among grapevine virologists and nurserymen in order to evaluate the discrepancies existing in the implementation of grapevine sanitary selection and certification in different States in Europe and to discuss about the perspective of the harmonisation of the rules and of their technical set up in the EU. From the discussions it came out that the European Directive on "marketing of vegetatively propagated material of grapevine" had to be updated and that the virologists had to work on the harmonisation of the protocols used for the detection of viruses and virus-like diseases.

After a meeting of European virologists in Le Grau du Roi (France), a project proposal was submitted to the AIR program of EU. The funding of the project entitled "Sanitary certification of grapevine: European network for the establishment of reference protocols for detection of infectious agents" was decided by the Directorate General for agriculture (DG VI F.II.3) for a period from 1993 to 1996.

The participants to the AIR network were:

- Institut National de la Recherche Agronomique, Colmar, France *(B. Walter) coordinator*
- Etablissement National Technique pour l'Amélioration de la Viticulture, Le Grau du Roy, France, *(R. Boidron)*
- Staatliches Weinbauinstitut, Freiburg, Germany *(H.H. Kassemeyer)*
- National Agricultural Research Foundation, Plant Protection Institute, Volos, Greece *(I.Rumbos)*
- University of Bari, Dipartimento di Protezione delle Piante dalle Malattie, Bari, Italy *(G.P. Martelli)*
- University of Sassari, Istituto di Patologia Vegetale *(U. Prota)*

- Insituto Nacional de Investigacào Agrària. Departamento de Fitopatologia, Oeiras, Portugal *(O.A. De Sequeira)*
- Instituto Nacional de Investigacion y Tecnologia Agraria y Alimentaria, Departamento de Proteccion Vegetal, Madrid, Spain *(J. Fresno-Perez)*
- Centro Investigacion Desarollo Agroalimentario, La Alberca, Murcia, Spain *(V. Padilla)*

The proposed actions were the following:

I) comparison of the ELISA methods used for the detection of nepoviruses, closteroviruses and grapevine fleck virus;

II) comparison of indexing techniques for the detection of leafroll, fleck, corky bark, stem pitting, stem grooving, vein mosaic and vein necrosis;

III) constitution of a central collection of grapevine infected by the concerned viruses and virus-like diseases;

IV) preparatory meeting for a study of methods used for the elimination of viruses and virus-like diseases.

The first part of the work consisted in discussions for comparing in details the ELISA techniques used by the participants, to establish the list of indexing varieties, diseases, grafting techniques and expression of symptoms. The second part consisted mainly in the exchange of materials (ELISA reagents, cuttings of indicators and references) and comparative analyses. The third part was the establishment of the grapevine repository at Colmar.

Further meetings took plase in Sassari (Italy – May 1994), Lisbon (Portugal – June 1995), Murcia (Spain – May 1996) and Freiburg – Colmar (Germany and France – October 1997).

The results obtained in the course of EUGN activities were collected in a book edited by B. Walter, entitled "Sanitary selection of the grapevine. Protocols for detection of viruses and virus-like diseases", Les Colloques n° 86, INRA Editions Paris, 1997.