

Research Note

Occurrence of diverse MLOs in tissues of grapevine affected by grapevine yellows in different countries

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Grapevine yellows (GY) have been reported in many regions of the world. MLO etiology has been evidenced in the case of flavescence dorée (FD) but remains to be fully demonstrated in most other occurrences. In a previous study (DAIRE *et al.* 1993), diseased grapevine samples from France were examined by PCR, using primers designed for the specific amplification of a part of the 16S rRNA gene from most known MLOs; according to the *AluI* restriction profile of the DNA fragment amplified, MLOs can be classified into four groups (AHRENS and SEEMÜLLER 1992). FD was shown to belong to group IV, and occurred in samples from the south of France. In addition, two non-FD MLOs occurred in FD-negative samples: the most frequent type was attributed to bois noir disease (BN) and belonged to group II. In the present survey, we examined samples of diseased grapevine harvested in different countries.

FD previously identified in symptom-bearing Alicante Bouschet and symptomless rootstock 3309 C cultivar harvested in southern France, and BN from Chardonnay harvested in Burgundy were used as references. In Italy, symptom-bearing canes were harvested on San Giovese in Bologna area, Chardonnay in Torino area, Perera, Sauvignon, Chardonnay and Garganega in Udine area, and on Inzolia in Sicily. Chardonnay with symptoms was also harvested in Israel, and diseased Riesling in New York State.

All the procedures for DNA extraction from grapevine, PCR and restriction analysis have been described (DAIRE *et al.* 1992, 1993). The Figure presents the data obtained. Apart for FD-infected French samples (Alicante Bouschet and rootstock 3309 C), only the samples of the four grapevine varieties in Udine, Italy, provided a *AluI* digestion pattern similar to group IV (only Perera and Sauvignon are shown on the Figure). The group II pattern found in Burgundy with the BN-MLO was evidenced in samples from Bologna, Torino and Sicily areas, and in the sample from Israel. A type III pattern was found in the Riesling sample from New York State.

The results presented here bring additional information about the occurrence of MLOs other than FD in GY-affected grapevines, and their relative diversity. Most of the diseased samples were of BN type (group II). The data obtained in Udine (FD-type, group IV) are confirmed by positive results obtained both with FD-specific probes (DAIRE unpublished) and antibodies (KUSZALA unpublished) on the same grapevine samples. A group III type (WX-type) was found in New York. This result is in agreement with the detection of a WX-MLO type in grapevine from Virginia State (DAVIS, personal communication).

The juxtaposition of different MLOs in very close areas, previously observed in France, may be a more general phenomenon. For instance, in New York two different MLOs have been identified in grapevine (group III, this survey) and in vector leafhoppers (FD, MARXNER *et al.* 1993) in the same vineyard; in Udine (Italy), a MLO transmitted from grapevine to periwinkle

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by dodder (FDU, DAVIS *et al.* 1992) belongs to group III (AHRENS and SEEMÜLLER 1992), although only FD-MLO was found in the present study, and FD antigens were found in leafhoppers used for transmission from grapevine to grapevine (OSLER *et al.* 1993).

An exhaustive survey of grapevine yellows in the main viticultural areas of the different countries should lead to a better knowledge of the variety of MLOs which are involved in this group of grapevine diseases. Specific genomic and serological tools, such as have been obtained for FD, will be necessary for etiological and epidemiological studies.

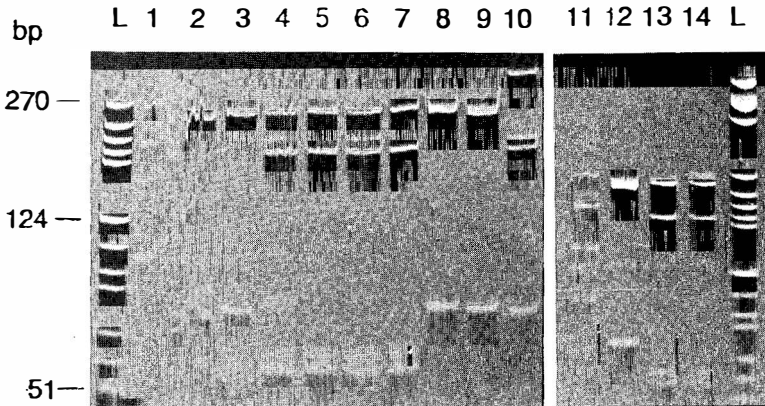


Figure: Polyacrylamide gel electrophoresis of *AluI* restricted MLO 16S rDNA fragments PCR-amplified from GY-affected grapevines. Healthy grapevines: (1), (11) Alicante Bouschet from two different stocks. Weak bands were obtained together with absence of MLO-DNA like profile. FD-infected grapevines from southern France: (2) and (12) Alicante Bouschet from different localities; (3) rootstock 3309 C. BN-infected grapevine: (4) Chardonnay from Burgundy. GY-affected grapevines from other countries: (5) and (6) two different samples of Chardonnay from Torino, (7) San Giovese from Bologna, (8) and (9) Perera and Sauvignon from Udine, (10) Riesling from New York, (13) Inzolia from Sicily, (14) Chardonnay from Israel. L: molecular weight marker.

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