

ASSESSING GENETIC DIVERSITY OF NERO D'AVOLA GRAPEVINE CULTIVAR BY USING SSR MARKERS

P. RICCARDI***, G. LAURIA**, R. GRILLO**, M.C. FIORE**, R. CIFARELLI**,
F. SUNSERI*

*) Dipartimento di Biologia, Difesa e Biotecnologie AgroForestali, Università degli Studi della Basilicata, Via Ateneo Lucano 10, 85100 Potenza

***) Metapontum Agrobios – SS Jonica 106 km 448,2, 75012 Metaponto (MT)

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Grapevine (*Vitis vinifera* L.) is highly polymorphic since more than 7000 cultivars are believed to exist in the world. This variability can be observed at the morphological level as well as at the agronomical and technological level (Alleweldt and Possingham., 1988).

The methods based on morphologic and ampelographic features commonly used for *Vitis vinifera* cultivar identification not always allow the most accurate information because of the interaction of the genotype with the environment (Dettweiler, 1993); on the contrary it is well reported (Imazio *et al.* 2002) that genetic methods (molecular marker characterization) overcome this problematic. With the aim to characterize and clarify the diversity of the Nero d'Avola grapevine cultivar it has been developing a DNA fingerprinting method. For this purpose it has been used different accessions belong the cultivar studied from different Sicilian areas, in particular 15 vineyards from 3 different areas (Caltanissetta, Trapani and Ragusa) were sampled.

Within a wide range of molecular markers we used a PCR-based method named Simple Sequence Repeats (SSRs), also called microsatellites. This work included all steps of characterization from sampling through fingerprinting.

Genomic diversity and differentiation in accessions was analyzed using polymorphism at twenty-four microsatellite loci (VVS2; VVMD5, 7, 27,14,17, 21, 26, 31, 24, 25, 28, 32, 34, 36; VrZAG62, 79, 7, 12, 14, 15, 21, 25, 67). Forward primers of the twenty-four SSR loci were labelled with one of the three unique ABI PRISM fluorescent dyes: 6-FAM, TET, HEX, and the PCR reactions were run on ABI PRISM.

The automated sequence system ABI PRISM combined with fluorescent labelling of expected fragments has been applied as an alternative to radioactivity detection using [³²P] or [³³P]-labelling. This technology provides an automatic and rapid sizing of the fragments through the use of specific internal size standard (GS500-Liz) and allows analysis of fragments obtained by simultaneously multiplex amplification up to three different labeled PCR products in the same reaction. The data collected from each sample were automatically analysed by GeneMapper Analysis Software.

The genetic diversity between accessions were estimated and multivariate analysis methods were applied for the analysis of genetic relationships among the vineyards and the Nero d'Avola areas.

References

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