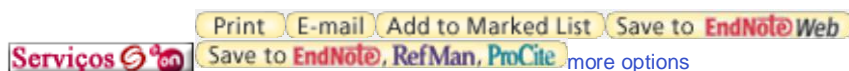


## Multiplex RT-PCR for detection and identification of three necroviruses that infect olive trees



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**Abstract:** An optimized multiplex RT-PCR assay was developed to discriminate three necrovirus (Olive latent virus 1 (OLV-1), Tobacco necrosis virus D (TNV-D) and Olive mild mosaic virus (OMMV)) that infect olive trees. An olive orchard consisting of 54 trees of cv. "Galega vulgar" in the south of Portugal was surveyed. dsRNA fraction was used as template and revealed the 3 viruses, singly or in multiple infections, present in 17 out of 54 trees in the orchard. OMMV was the most frequent occurring in 15 trees, followed by OLV-1 in 12 and TNV-D in 4 plants. The results obtained showed that necrovirus- specific dsRNAs do exist in infected tissues in amounts below the resolution permitted by gel electrophoresis analysis and that the developed multiplex PCR based assay is of much higher sensitivity. The design of the specific primers described enabled, for the first time, to discriminate between OMMV and TNV-D by means of RT-PCR assays, an indispensable tool in identification, epidemiology and survey studies.

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