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Poster Abstract - H.04

VALIDATION OF REAL-TIME PCR METHODS FOR GRAPEVINE DETECTION AND IDENTIFICATION: A PRACTICAL APPROACH

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Genetic engineering of commercial crops has progressed very rapidly in the last decades and recently genetic modification of grape has been taken into consideration as a powerful tool to improve quality of vine varieties and their derived products. In view of the scientific and technical results already achieved and expected in the near future, European Union considered the necessity to introduce specific provisions for risk assessment, labelling and monitoring of genetically modified (GM) vine in the legislation and the marketing of material for the vegetative propagation of the vine (Council Directive 2002/11/EC) and implemented a comprehensive GM food and feed legislation (Regulation EC 1829/2003). The enforcement of this legislative framework entails the development, validation and application of analytical methods for the specific detection and quantification of transformation events of grape varieties. Real-time PCR is currently the most powerful technique for the quantification of specific nuclei acid sequence. As for grapevine, however, this technique is not a routine yet and needs to be improved. Being embarked in projects on grape molecular breeding and GMO detection, we developed a protocol for standardizing and optimizing grape Real-time PCR identifying a valuable endogenous gene to be applied as genotype referee and gene copy number standard. Finally, we developed a practical approach for the validation of our technique, and provided a detailed protocol for the evaluation of detection and quantification limits, linear dynamic range, precision and trueness, specificity and robustness.

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