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The alternative oxidase family of *Vitis vinifera* reveals an attractive model to study the importance of genomic design

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ABSTRACT

'Genomic design' refers to the structural organization of gene sequences. Recently, the role of intron sequences for gene regulation is being better understood. Further, introns possess high rates of polymorphism that are considered as the major source for speciation. In molecular breeding, the length of gene-specific introns is recognized as a tool to discriminate genotypes with diverse traits of agronomic interest. 'Economy selection' and 'time-economy selection' have been proposed as models for explaining why highly expressed genes typically contain small introns. However, in contrast to these theories, plant-specific selection reveals that highly expressed genes contain introns that are large. In the presented research, 'wet'*Aox* gene identification from grapevine is advanced by a bioinformatics approach to study the species-specific organization of *Aox* gene structures in relation to available expressed sequence tag (EST) data. Two *Aox1* and one *Aox2* gene sequences have been identified in *Vitis vinifera* using grapevine cultivars from Portugal and Germany. Searching the complete genome sequence data of two grapevine cultivars confirmed that *V. vinifera* alternative oxidase (Aox) is encoded by a small multigene family composed of *Aox1a*, *Aox1b* and *Aox2*. An analysis of EST distribution revealed high expression of the *VvAox2* gene. A relationship between the atypical long primary transcript of *VvAox2* (in comparison to other plant *Aox* genes) and its expression level is suggested. *V. vinifera* Aox genes contain four exons interrupted by three introns except for *Aox1a* which contains an additional intron in the 3'-UTR. The lengths of primary Aox transcripts were estimated for each gene in two *V. vinifera* varieties: PN40024 and Pinot Noir. In both varieties, *Aox1a* and *Aox1b* contained small introns that corresponded to primary transcript lengths ranging from 1501 to 1810 bp. The *Aox2* of PN40024 (12 329 bp) was longer than that from Pinot Noir (7279 bp) because of selection against a transposable-element insertion that is 5028 bp in size. An EST database basic local alignment search tool (BLAST) search of GenBank revealed the following ESTs percentages for each gene: *Aox1a* (26.2%), *Aox1b* (11.9%) and *Aox2* (61.9%). *Aox1a* was expressed in fruits and roots, *Aox1b* expression was confined to flowers and *Aox2* was ubiquitously expressed. These data for *V. vinifera* show that atypically long *Aox* intron lengths are related to high levels of gene expression. Furthermore, it is shown for the first time that two grapevine cultivars can be distinguished by *Aox* intron length polymorphism.

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