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Charaterization of new MYB genes potentially regulating anthocyanin synthesis in *Vitis vinifera*

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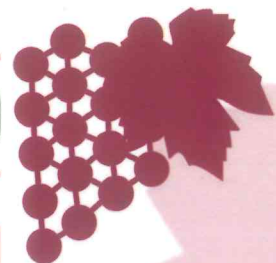
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Third International Symposium

MACROWINE2010

on Macromolecules and Secondary Metabolites of Grapevine and Wines

Torino, Italy, June 16-18 / 2010 - Congress Center Torino Incontra Via Nino Costa 8



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BOOK OF ABSTRACTS

Wednesday, 16 June 2010

9.00 – 9.20 Opening Ceremony

- SESSION I – CHEMISTRY, FUNCTIONALITY AND ANALYSIS OF GRAPE AND WINE MACROMOLECULES AND SECONDARY METABOLITES

Chairperson: Véronique Cheynier, Montpellier, France

9.20 - 10.00 **Keynote Lecture**

CHEMISTRY AND ANALYSIS OF KEY VOLATILE COMPOUNDS OF WINE AND THEIR PRECURSORS IN GRAPE

Philippe Darriet, *University of Bordeaux 2, Villenave d'Ornon, France*

10.00 – 10.40 **Dalmasso Lecture**

WINE OXIDATION: RECENT REVELATIONS

Andrew Waterhouse, *University of California, Davis, USA*

10.40 – 11.00

Characterization and formation kinetic study of phenolic compounds-volatile thiol adducts by chemical and enzymatic oxidation

Maria Nikolantonaki, *Institut des Sciences de la Vigne et du Vin, Villenave d'Ornon, France*

11.00 - 11.30 Coffee break

11.30 – 11.50

Preliminary studies on polyphenol assessment by Fourier transform-near infrared spectroscopy (FT-NIR) in berries

Chiara Pagliarani, *University of Torino, Torino, Italy*

11.50 – 12.10

A metabolomic view of wine micro-oxygenation

Fulvio Mattivi, *Fondazione Edmund Mach, San Michele all'Adige, Trento, Italy*

12.10 – 12.30

Effects of ionic strength and sulfate upon thermal aggregation of grape chitinases and thaumatin-like proteins in model system

Matteo Marangon, *The Australian Wine Research Institute, Glen Osmond, Australia*

12.30 - 12.50

Progress in precision viticulture for the assessment of grape anthocyanins at harvest in Australian vineyards

Marine Le Moigne, *Centre Universitaire Orsay, Orsay Cedex, France*

13.00 - 14.00 Lunch

- SESSION II - BERRY DEVELOPMENT AND BIOSYNTHESIS OF GRAPE SECONDARY METABOLITES

Chairperson: Serge Delrot, Bordeaux

14.00 - 14.40 **Keynote Lecture**

REGULATION OF FLAVONOID BIOSYNTHESIS DURING GRAPE BERRY DEVELOPMENT

Jochen Bogs, *University of Heidelberg, Adelaide, Germany*

14.40 - 15.00

Grapevine anthocyanin transport involves in vivo vesicle-mediated trafficking and the contribution of anthomate transporters and GST

Agnès Ageorges, *INRA - UMR SPO, Montpellier, France*

15.00 - 15.20

Cellular localization of leucoanthocyanidin reductase and anthocyanidin reductase in grape skins of *Vitis vinifera* L. cv. Cabernet-Sauvignon during development

Soizic Lacampagne, *Institut des Sciences de la Vigne et du Vin, Villenave d'Ornon, France*

15.20 - 15.40

Cloning and characterization of putative anthocyanin acyltransferases from grape and *Petunia*

Sofia Provenzano, *University of Torino, Torino, Italy*

15.40 - 16.10 Coffee break

16.10 - 16.30

New MYBS through the glass: a deeper look into the regulatory network of flavonoid synthesis in wine grapes

J. Tomas Matus, *Center for Research in Agricultural Genomics, Barcelona, Spain*

16.30 - 16.50

Study of genetic determinants of flavonol content in grape berry skin

Giulia Malacarne, *Fondazione Edmund Mach, San Michele All'Adige, Trento, Italy*

16.50 - 17.10

Are VvMYB5a and VvMYB5b regulators of grape flavonoid pathway?

Giambattista Tomielli, *University of Verona, Verona, Italy*

17.10 - 17.30

Investigation of VvDXS function and its effects on Muscat flavor levels

Juri Battilana, *IASMA Research and Innovation Centre, San Michele All'Adige, Trento, Italy*

17.30 - 17.50

Microclimate influence on gene expression and metabolite accumulation in grape berries

Christian Kappel, *University of Bordeaux, Bordeaux, France*

18.00 - 20.00 Welcome cocktail

GRAPEVINE ANTHOCYANIN TRANSPORT INVOLVES *IN VIVO* VESICLE-MEDIATED TRAFFICKING AND THE CONTRIBUTION OF ANTHOMATE TRANSPORTERS AND GST

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Anthocyanins are plant secondary metabolites responsible for most of the red to purple pigmentation found in flowers, fruits and leaves. In grape berry, these compounds are accumulated during development from *véraison* to ripening, and are responsible for pigmentation found in red skin berries. Grapevine anthocyanins are 3-monoglucosides, which can be acylated by acetic, p-coumaric or caffeic acid. In the cell, anthocyanins are synthesized at the cytoplasmic surface of the endoplasmic reticulum and are then transported and finally accumulated inside the vacuole. In grapevine, two kinds of molecular actors are putatively associated in vacuolar sequestration of anthocyanins : a Glutathion S-transferase (GST) ⁽¹⁾ and two MATE-type transporters, anthoMATE1 (AM1) and anthoMATE3 (AM3), which specifically mediate acylated anthocyanin transport *in vitro* ⁽²⁾. However, the molecular suite of mechanisms by which anthocyanins are imported into the vacuole remains unclear.

We used Myb-A1 transformed hairy roots ⁽³⁾ as a grapevine model tissue producing anthocyanins and took advantage of the unique autofluorescence of anthocyanins to study their cellular trafficking. In these tissues, anthocyanins were not only visible in the largest vacuoles but also, and more intensively present in several small vesicles, suggesting that these vesicles could mediate anthocyanins to the vacuole. In the cell, these vesicles actively moved alongside the tonoplast, suggesting a vesicular trafficking.

Subcellular homologous localisation assays in anthocyanin accumulating organs revealed that AMs were not only located to the tonoplast but also closely related with these small vesicles while GST was localised in the cytoplasm, and specifically around the nucleus, suggesting a close association with the endoplasmic reticulum.

Furthermore, in hairy roots expressing *AM3* antisense, no such anthocyanin containing vesicles were observed, while in hairy roots expressing *GST* antisense, anthocyanins were accumulated in these vesicles only but not in the vacuole. These observations suggest that in grapevine, AnthoMATE transporters are involved in vesicular transport of anthocyanins while GST could be involved in another yet unknown transport mechanism.

REFERENCE LIST

- [1] Conn et al. (2008) Purification, molecular cloning, and characterization of glutathione S-transferases (GSTs) from pigmented *Vitis vinifera* L. cell suspension cultures as putative anthocyanin transport proteins. *J Exp Bot* **59**: 3621-3634
- [2] Gomez et al. (2009) Grapevine MATEtype proteins act as vacuolar H⁺-dependent acylated anthocyanin transporters. *Plant Physiol* **150**: 402-415
- [3] Cutanda-Perez et al. (2009) Ectopic expression of VmybA1 in grapevine activates a narrow set of genes involved in anthocyanin synthesis and transport. *Plant Mol Biol* **69**: 633-648