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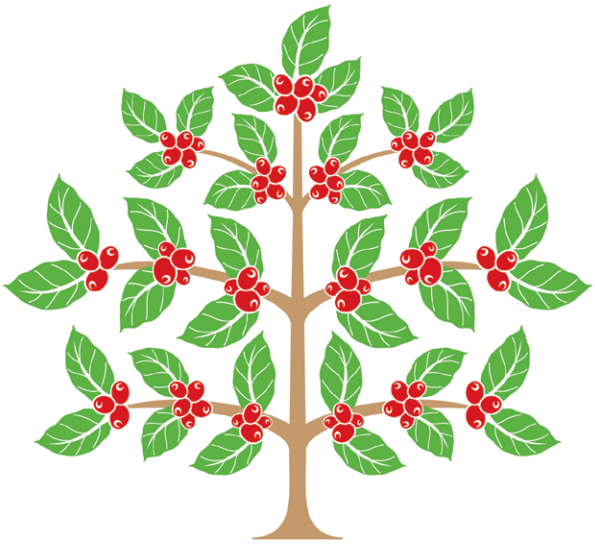

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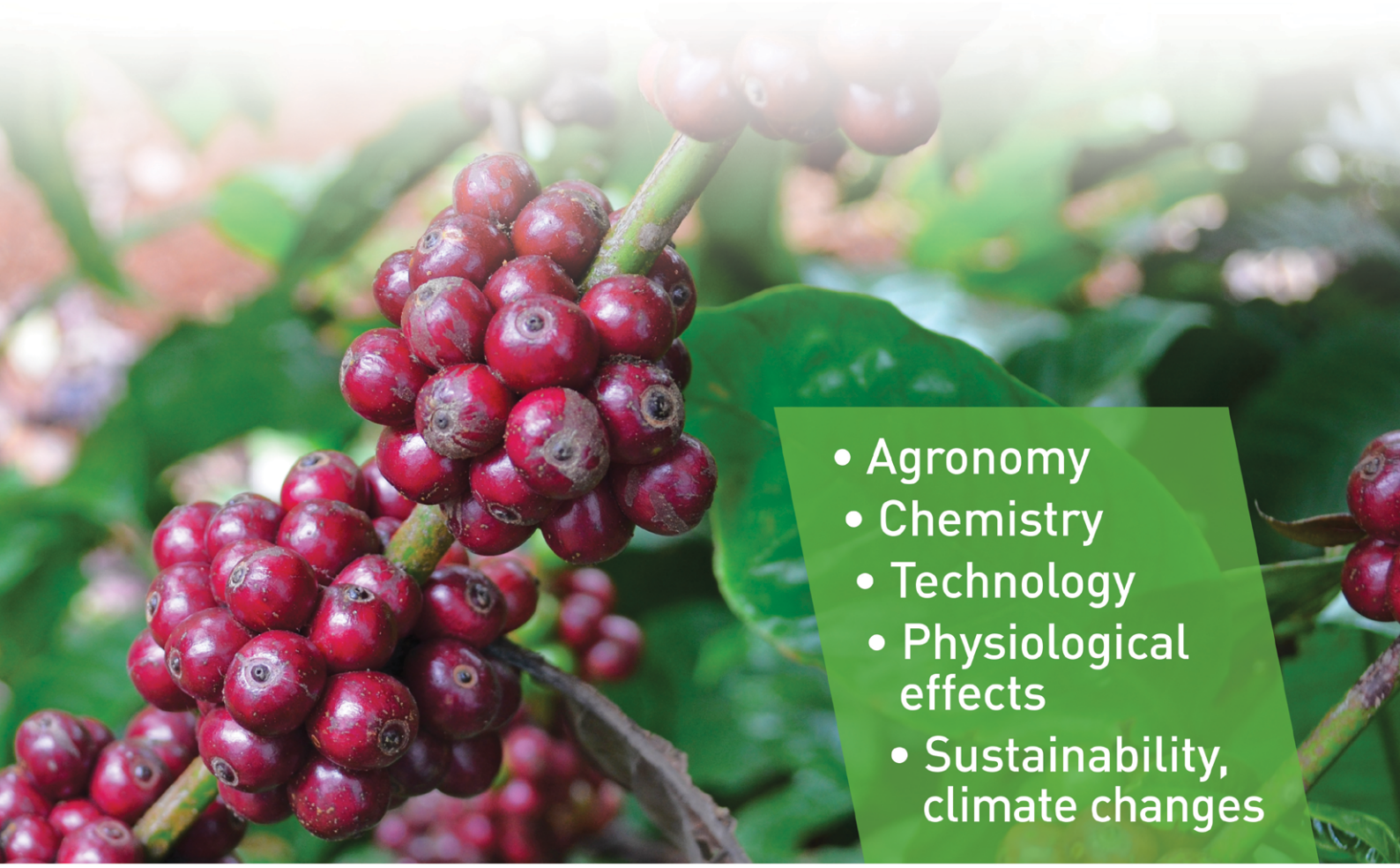
BOOK OF ABSTRACTS



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Unravelling the metabolic and hormonal machinery during key steps of somatic embryogenesis: A case study in coffee

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RATIONALE

Somatic embryogenesis (SE) is one of the most promising processes for large-scale dissemination of elite varieties. However, whatever the species considered, SE research still remains essentially empirical resulting in many drawbacks to fulfil market demands, especially due to an overall slow technical progress over the last 20 years. Knowledge about the molecular events involved in the key steps of the SE process is urgently needed to pilot the optimization of SE protocols. In this study, we took advantage of the latest metabolomics technologies and applied them to one of the most advanced and reliable large-scale SE processes, the one developed for coffee.

METHODS

Sampling covered 15 key developmental stages. Five independent leaf introductions were carried out with more than 4,000 leaf explants and a total of 25 independent cell lines. All obtained cell lines were high-yielding and time-synchronized during embryo regeneration enabling a successful sampling. Primary metabolites, secondary metabolites and phytohormones were quantified using GC-MS, HPLC and UPLC- MS/MS respectively. A robust statistical method was used to identify metabolic pathway changes associated with the main developmental phases and phase switches. Histological analysis and cell imaging were also required to characterize developmental stages and associate metabolic profiles with cell structure organization. Lastly, comparing Arabica embryogenic and non-embryogenic calli enabled the identification of metabolic markers of the embryogenic capacity.

RESULTS

Statistical analysis performed on 104 metabolites revealed that massive re-configuration of metabolic pathways induced SE. During initial dedifferentiation, a sharp decrease in phenolic compounds and caffeine levels was observed while auxins, cytokinins and ethylene levels were at their highest. Totipotency reached its highest expression during the callus stages when a shut-off in hormonal and metabolic pathways related to sugar and energetic substance hydrolysis was evidenced. Abscisic acid, leucine, maltotriose, myo-inositol, proline, tricarboxylic acid cycle metabolites and zeatin appeared as key metabolic markers of the embryogenic capacity. Combining metabolomics with multiphoton microscopy led to the identification of chlorogenic acids as markers of embryo redifferentiation.

CONCLUSIONS & PERSPECTIVES

The present analysis shows that metabolite fingerprints are signatures of cell fate and represent a starting point for optimizing SE protocols in a rational way. These findings should be informative and useful to a wide range of plant species, offering unprecedented perspectives in plant micropropagation.

References:

- Awada et al. 2019 International Journal of Molecular Sciences DOI: 10.3390/ijms20194665