

A gene-focussed approach to reducing cadmium uptake in cocoa

Using a yeast assay to accelerate progress in identifying varieties with low uptake

Cadmium (Cd), is a heavy metal which can enter the human food chain from a number of sources including foods such as rice and vegetables which take up cadmium from the soil. In 2019, the European Union set maximum limits for Cd in a range of foods, including cocoa, to protect the health of consumers. The new regulation has a particular impact on cocoa farmers in some areas of Latin America and the Caribbean where the local soils and planting materials can result in higher levels of Cd in cocoa beans. Since many of the areas affected are known for their production of fine/flavour cocoas, there is a concern that supplies for specialty blends and “single origin” premium chocolates would be particularly affected. Previous research at the Cocoa Research Centre at the University of the West Indies (CRC/UWI) has shown that some varieties take up less Cd than others. Better ways to identify “low uptake” varieties would accelerate progress in developing the rootstocks and varieties urgently needed by farmers.

This research uses a gene-based approach focused on candidate genes associated with the uptake of Cd by roots and its transport into the vegetative parts of the plant. These genes belong to two families: the Natural Resistance-Associated Macrophage Protein (NRAMP) genes that encode metal transporter proteins, and Heavy-Metal ATPase (HMA) genes. Relevant studies from other species has shown that natural genetic variation in these genes is associated with modification of Cd uptake and accumulation. Five NRAMP genes were identified in cacao and their expression studied in various organs and in response to variation in nutrient availability and the addition of Cd. A yeast-assay system was used to study the function of these genes. Yeast can be easily transformed and the effect of the introduced gene(s)

on Cd uptake can be rapidly assessed in a few days whereas testing the genes even in a simple plant system can take several months. Training in this technique was provided by UoR for a visiting scientist from CRC at an early stage of the project. Research undertaken in collaboration with Imperial College, used Cd isotope analysis to indicate that movement of Cd within the plant is primarily unidirectional from roots to leaves, with little evidence for re-distribution, and that lighter isopes are preferentially retained in the roots. When used with the yeast-assay system, isotope studies provided further evidence for the important role that NRAMP5 plays in the pathways for uptake of Cd in cacao whereas the HMA-family appear to contribute to Cd sequestration. Variants of the genes detected in set of nine cacao clones which had been selected based on their contrasting Cd uptake and within-plant movement characteristics are being assessed in both the yeast-assay system and in a simple plant system to further investigate whether they contribute to the varietal differences in Cd uptake and in plant movement.

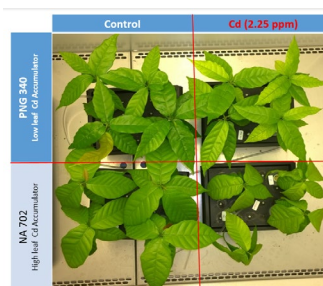


Photo: I. Ullah

Figure 1. Growth of PNG 340, a low cadmium (Cd) accumulating accession, is less affected than NA 702, a high Cd accumulating accession, under 2.25 ppm Cd application compared to control.

Publications:

Ullah, Wang, Eide, Dunwell (2018) Evolution, and functional analysis of Natural Resistance-Associated Macrophage proteins (NRAMPs) from *Theobroma cacao* and their role in cadmium accumulation. *Sci Rep* 8(1).

Moore, Ullah, de Oliveira, Hammond, Strekopytov, Tibbett, Dunwell, & Rehkämper (2020). Cadmium Isotope Fractionation Reveals Genetic Variation in Cd Uptake and Translocation by *Theobroma Cacao* and Role of Natural Resistance-Associated Macrophage Protein 5 and Heavy Metal ATPase-Family Transporters. *Hort Res* 7 (1): 71

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