FOLATE BIOFORTIFICATION; Metabolic engineering strategies and potential for human nutrition

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Biofortification is the enrichment of nutrients in food organisms by means of breeding or genetic engineering. It has advantages over regular fortification, once it is developed, the cost of generation of biofortified foods would be the same as conventional ones, making it sustainable; in addition, the enhanced nutrients are in their natural form. We focused on folate biofortification in tomato fruit. Folates are essential cofactors for the transfer of one-carbon units in organisms, they cannot be synthesized by humans, and folate deficiency is a global problem associated to a number of disorders like neural tube defects, anemia, cardiovascular disease, and some cancers. Folate molecule is composed by a pteridine ring, attached to a *p*-aminobenzoate (PABA) moiety which is glutamylated at different extent. Its biosynthesis is compartmented: pteridines are synthesized in citosol, PABA is made in plastids, and both molecules are condensed and glutamylated in mitochondria. The first strategy for engineering folates consisted on the increment of pteridine flux by overexpressing GTP cyclohydrolase I in citosol, giving this a slight folate increment of 2-fold and massive pteridine accumulation. Then, the second round of engineering was focused on elevate PABA production in plastids by means of aminodeoxychorismate synthase. Combination of the two engineered traits gave folate increments of 20 fold compared to control fruit. However, precursors still accumulated, which indicates a new constrain in the pathway. This work showed that biofortification of plant foods with folates is possible, and this is supported by similar outcomes obtained in rice and Arabidopsis from other groups. Therefore, this two-gene strategy can be applied to other crops with high possibilities of obtaining similar increments. However, some fine-tuning of the engineering may be needed, and more research about folate bioavailability and stability is required to characterize their potential and safety as enhanced foods for human nutrition.
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