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Structural and functional characterization of three key enzymes in the biosynthesis of vitamin C from *Myrciaria dubia*

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Myrciaria dubia (Kunth) McVaugh, known as camu-camu or aracá, is a fruit native to the Amazon, characterized by its high content of L-Ascorbic Acid (Vitamin C or AsA). (1) For this reason, the enzymes that participate in AsA biosynthesis (2-3) may have potential biotechnological applications for the artificial production of AsA. (1) However, up until now the structural biology of these enzymes has been poorly explored. Here, we describe the biophysical, functional, and structural properties for three enzymes of the Smirnoff/Wheeler pathway (MdGME, MdGDH and MdGalDH). All enzymes were purified by affinity chromatography followed by molecular exclusion. L-GDP-D-mannose 3',5' epimerase (MdGME), which catalyzes a double epimerization of GDP-mannose to produce GDP-L-galactose and GDP-L-gulose, proved to be a dimer in solution and its crystal structure was solved at 1.3 Å resolution. L-galactose dehydrogenase (MdGDH) is monomeric in solution with a K_m of 0.128 mM and an optimal pH of 7. The activity of the enzyme was not affected by high concentrations of AsA as has been reported for its homolog from spinach (SoGDH). This, we show to be due to a change in pH. The crystal structure of SoGDH was solved to 1.4 and 1.75 Å in its apo and holo (NAD⁺ bound) forms respectively. The structure allows us to rationalize the use of NAD⁺ (and not NADP⁺) as the cofactor and to describe differences within the active site when compared to other aldo-keto reductases, which are related to substrate specificity. Finally, L-galacton-1,4-lactone dehydrogenase (MdGalDH), presents a K_m of 0.041 mM and an optimal pH of 8 and its crystal structure was solved at 2.4 Å. The present work contributes to a broader understanding of structure-function relationships of enzymes involved in the synthesis of vitamin C.

Palavras-chave: Crystallographic structure. Vitamin C. *Myrciaria dubia*.

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Referências:

- 1 CASTRO, J. C. *et al.* Camu-camu - *Myrciaria dubia* H. B. K. (McVaugh). In: RODRIGUES, S., SILVA, E.; BRITO, E. (ed.) **Exotic fruits** : reference guide. Amsterdam: Elsevier, 2018. 490 p.
- 2 CASTRO, J. C. *et al.* De novo assembly and functional annotation of *Myrciaria dubia* fruit transcriptome reveals multiple metabolic pathways for L-ascorbic acid biosynthesis. **BMC Genomics**, v. 16, p. 997-1-997-13, 2015. DOI: 10.1186/s12864-015-2225-6.
- 3 WHEELER, G. L.; JONES, M. A.; SMIRNOFF, N. The biosynthetic pathway of vitamin C in higher

plants. **Nature**, v. 393, n. 6683, p. 365-369, 1998.