Investigation of novel enzymes for Apocarotenoid Biosynthesis in “Azafran de bolita” (Ditaxis heterantha Zucc.)

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Abstract *(Between 300 and 400 words)*

Ditaxis heterantha, a plant belonging to the Euphorbiaceae family, thrives in the semi-arid regions of Mexico. It is commonly known as "azafran de bolita" or "azafrancillo" due to its seeds, which feature a vibrant orange endosperm traditionally used by local populations as a dye and food additive, similar to the use of Crocus sativus (saffron).

Previous studies have demonstrated that Ditaxis heterantha seeds are rich in fatty acids, tocopherols, and phytosterols, all of which contribute to their significant antioxidant activity. In addition to these known compounds, in 2005 two novel apocarotenoids were found in the endosperm: Ditaxin (C31) and Heteranthin (C27). These compounds add a new dimension to the already impressive phytochemical profile of the plant.

To further explore the potential of Ditaxis heterantha, we evaluated the bioactivity of its endosperm extracts on various cancer cell lines. Our results revealed a significant reduction in cell viability even at low extract concentrations, indicating potent anti-cancer properties. This finding underscores the potential of Ditaxis heterantha as a source of new therapeutic agents.

In order to fully understand the plant's metabolic profile, both targeted and untargeted metabolomic analyses were conducted on the endosperm, peel, leaves, and roots. Additionally, analyses on DNA content and karyotype through flow cytometry and fluorescence microscopy were conducted to enhance the understanding of this species.

This comprehensive profiling provides a detailed understanding of Ditaxis heterantha's phytochemical landscape and emphasizes its potential as a source of bioactive compounds with applications in pharmaceuticals and nutraceuticals.

Moreover, our analyses are focused on elucidating the biosynthetic pathway of the two aforementioned apocarotenoids: Ditaxin and Heteranthin. We are integrating both metabolomics and transcriptomics approaches in order to pinpoint potential carotenoid precursor/s and identifying the specific genes involved in the biosynthesis of these apocarotenoids; in a first stage, we decided to focus on the carotenoid cleavage dioxygenases (CCDs) responsible for the biosynthesis of these new apocarotenoids, and in silico studies are still in progress. Future research will also consider other enzymes involved in the biosynthesis of carotenoids and apocarotenoids of D. heterantha. Once the list of candidate genes will be compiled, their roles will be verified using enzymatic activity assays in bacteria, yeast, and plants.

Keywords: Ditaxis heterantha, apocarotenoids, endosperm, transcriptomics, metabolic profiling

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