

## Moderate Electric Field Influence on Carotenoids Extraction Time from *Heterochlorella luteoviridis*

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**Abstract :** Carotenoids are high value added pigments that can be alternatively extracted from some microalgae species. However, the application of carotenoids synthesized by microalgae is still limited due to the utilization of organic toxic solvents. In this context, studies involving alternative extraction methods have been conducted with more sustainable solvents to replace and reduce the solvent volume and the extraction time. The aim of the present work was to evaluate the extraction time of carotenoids from the microalgae *Heterochlorella luteoviridis* using moderate electric field (MEF) as a pre-treatment to the extraction. The extraction methodology consisted of a pre-treatment in the presence of MEF (180 V) and ethanol (25 %, v/v) for 10 min, followed by a diffusive step performed for 50 min using a higher ethanol concentration (75 %, v/v). The extraction experiments were conducted at 30 °C and, to keep the temperature at this value, it was used an extraction cell with a water jacket that was connected to a water bath. Also, to enable the evaluation of MEF effect on the extraction, control experiments were performed using the same cell and conditions without voltage application. During the extraction experiments, samples were withdrawn at 1, 5 and 10 min of the pre-treatment and at 1, 5, 30, 40 and 50 min of the diffusive step. Samples were, then, centrifuged and carotenoids analyses were performed in the supernatant. Furthermore, an exhaustive extraction with ethyl acetate and methanol was performed, and the carotenoids content found for this analyses was considered as the total carotenoids content of the microalgae. The results showed that the application of MEF as a pre-treatment to the extraction influenced the extraction yield and the extraction time during the diffusive step; after the MEF pre-treatment and 50 min of the diffusive step, it was possible to extract up to 60 % of the total carotenoids content. Also, results found for carotenoids concentration of the extracts withdrawn at 5 and 30 min of the diffusive step did not presented statistical difference, meaning that carotenoids diffusion occurs mainly in the very beginning of the extraction. On the other hand, the results for control experiments showed that carotenoids diffusion occurs mostly during 30 min of the diffusive step, which evidenced MEF effect on the extraction time. Moreover, carotenoids concentration on samples withdrawn during the pre-treatment (1, 5 and 10 min) were below the quantification limit of the analyses, indicating that the extraction occurred in the diffusive step, when ethanol (75 %, v/v) was added to the medium. It is possible that MEF promoted cell membrane permeabilization and, when ethanol (75 %) was added, carotenoids interacted with the solvent and the diffusion occurred easily. Based on the results, it is possible to infer that MEF promoted the decrease of carotenoids extraction time due to the increasing of the permeability of the cell membrane which facilitates the diffusion from the cell to the medium.

**Keywords :** moderate electric field (MEF), pigments, microalgae, ethanol

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