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INFLUENCE OF THE EXTRACTION METHOD ON THE CHLOROPHYLLS OBTAINED FROM SPINACH LEAVES

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Abstract: Chlorophylls are green pigments present in plants and certain algae that are responsible for photosynthetic processes. Recently, several investigations have shown the high biological potential of these compounds, such as antioxidant and antibacterial activities [1,2]. There are even several herbal medicines, which contain chlorophylls being currently marketed. Thus, due to the high biological and economical importance of the chlorophylls, the present investigation aimed to evaluate the influence of the extraction method on the efficiency of chlorophylls extraction from spinach leaves. Aiming to compare five different types of extraction methods (infusion, maceration, decoction, extraction by Soxhlet equipment, and turbolization), they were carried out, in triplicate, using the same weight/volume rate (3 g/ 200 mL) as well as the same extractor solvent (ethanol). After the extraction processes, the solvent was removed by rotaevaporation and the obtained weights were measured. Ethanolic extracts were analysed by TLC, using silica as the stationary phase and hexane-acetone, 8:2, as the mobile phase. Spectra on UV-VIS (200-1100 nm) were recorded as well. Such analyses of ethanolic solutions (1 g/ 5 mL) were carried out in order to obtain chlorophyll a/chlorophyll b rate. The results (Table 1) have shown that infusion was the best process to extract chlorophylls, allowing an extraction of 7.51 %, while other methods have presented percentages of extractions in the range of 1.13-1.61 %. According to extractions percentage, the order of efficiency of extraction processes was the following: infusion>>decoction>turbolization>maceration>Soxhlet. TLC analyses showed that all methods allowed the extraction of the same kind of pigments, while UV-Vis analyses indicated that chlorophyll a/chlorophyll b rates were the same for decoction, Soxhlet and turbolization methods. Therefore, the present investigation has shown that extraction processes influence significantly the efficiency of chlorophylls extraction and the type of such pigments that is extracted.

Extraction method	% EXTRACTED	Chlorophyll a/Chlorophyll b relationship (A _{647 nm} /A _{664 nm})
Infusion	7.51 ± 0.16	0.693 ± 0.060
Maceration	1.21 ± 0.12	0.426 ± 0.003
Decoction	1.61 ± 0.05	0.987 ± 0.005
Soxhlet	1.13 ± 0.03	0.985 ± 0.004
Turbolization	1.39 ± 0.26	0.985 ± 0.006

Table 1.	Efficiency of	chlorophylls	extractions by	different methods.
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References:

[1] Gyawali, R., Ibrahim, S.A. 2014. Natural products as antimicrobial agents. Food Control, 46: 412-429.

[2] Ragonese, C., Tedone, L., Beccaria, M., Torre, G., Cichello, F., Cacciola, F., Dugo, P., Mondello, L. 2014. Characterization of lipid fraction of marine macroalgae by means of chromatography techniques coupled to mass spectrometry. Food Chemistry, 145: 932-940.