EXTRACTION OF STINGING NETTLE (*URTICA DIOICA L.)* WITH SUPERCRITICAL CARBON DIOXIDE

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Abstract

The extraction of the stinging nettle leaves was performed with 20 kg/h flow rate of supercritical CO_2. The influence of the working parameters: pressure (140 bar, 210 bar and 350 bar), temperature (from 40 ^oC to 60 ^oC) on the quantity of extract and contents of chlorophyll *a+b* and β-carotene in it is determined. The biggest quantity of extract (4.48% in relation to the dry matter) with 0.88% contents of chlorophyll *a+b* and 0.50% β-carotene is obtained at 350 bar, 60 ^oC, 6h extraction time.

Key words: stinging nettle, supercritical CO_2, chlorophyll, β-carotene, yield of extract

1. Introduction

The stinging nettle designated as a detestable weed plant, spread almost all over the world is a treasure of active components. Due to the fact that stinging nettle leaves are rich in flavonoids, chlorophylls and carotenoids and their degradation products, vitamins [1-3], proteins [4], mineral materials, organic acids, oil [5,6], and other components, the stinging nettle is of high value in the folk medicine as well as in scientific medicine. The large number of produced preparations from the stinging nettle is found to be used in the process of prevention or treatment of various diseases. The sitosterols and agglutinins [7] isolated from the nettle root, which show positive effects on the treatment of benign prostate hyperplasias [8] are lately becoming more and more interesting.

Apart from the use of the organic solvents, supercritical fluids, especially CO_2, are used for extraction of plant materials [9-11].

The lack of literature data for extraction of stinging nettle with supercritical CO_2 set the objective of this work. The influence of the pressure, temperature, density of supercritical CO_2 on the contents of chlorophylls and β-carotene in the obtained stinging nettle extracts was followed.

2. Experimental

Materials

Stinging nettle leaves (*Urtica dioica* L., *Urticaceae*) were picked up in the Republic of Macedonia in May, 1997. The content of moisture, proteins, mineral materials and fats in the stinging nettle leaves determined by standard procedure [12] were 8.3%, 27.2%, 12.5 and 7.0%, respectively.

Purity of the carbon dioxide supplied by Tehnogas-Zrenjanin (SR Jugoslavia) was better than 99.8%.

Extraction

The pilot extraction plant is a product of Uhde GMbH (Germany). It is equipped with an extraction vessel of volume 4 L and inner diameter (i.d.) 115 mm, 60-500 bar working pressure and 20-130 ^{o}C temperature. The separator is with 4 L volume, i.d.=100 mm, 50-120 bar pressure and 20-120 ^{o}C temperature. In all performed experiments, the operational parameters in the separator were 50 bar and 25 ^{o}C. The pump with teflon membranes is product of Lewa Company (Austria), type M611.

The extraction of the stinging nettle by supercitical CO_2 was performed under controlled pressure and temperature at 20 kg/h CO_2 flow rate for 6 h. For each experiment, the extractor tube was filled with 1000 g stinging nettle leaf with definite particle size (0.630-0.125 mm) and moisture content (8.3%). For "step-wise" extraction, an extraction of material (5000 g, 5 batches per 1000 g) was performed in three steps. Each step was run for 6 h at 20 kg/h supercritical CO_2 flow rate. The working pressure and temperature were for step I: 140 bar and 40 ^{o}C, for step II: 210 bar and 50 ^{o}C, for step III: 310 bar and 60 ^{o}C.

By the yield of the extract, expressed in relation to the dry matter in the stinging nettle leaf, as well as by the evaluation of the total contents of chlorophyll *a*+*b* and β-carotene in the obtained extracts is estimated the influence of the conditions under which the extraction is performed. The content of chlorophyll *a*+*b* and β-carotene is determined by measuring the extinction on the acetone and petroleum ether solution [13] of the obtained extract on UV-VIS spectrophotometer type HP UV 8452.

3. Results and discussion

1. "Step-wise" extraction of stinging nettle

In the first step (I) of "step-wise" extraction of stinging nettle is obtained 2.49% quantity of extract expressed expressed in relation to the dry matter in the stinging nettle. The contents of chlorophyll *a*+*b* and β-carotene in this extract is 0.08% and 0.14% adequately.

In the second (II) and third (III) step is extracted 1.69% and 1.0% extract. The bigger concentrations of coloured components chlorophylls and β-carotene are determined in the extracts obtained at higher pressure and temperature. Using CO_2 as an extraction means under its supercritical conditions for "step-wise" extraction of stinging nettle, it is determined that the chlorophylls showed bigger ability for dissolution at high pressure of 210 bar and temperature of 50 ^{o}C, whereas β-carotene showed good dissolution at the lower examined pressure of 140 bar and 40^{o}C temperature (Fig. 1).

In the fourth (IV) of the "step-wise" extraction performed by acetone maceration is obtained 1.9% quantity of extracts and 8.3% chlorophyll and 1.1% β-carotene (Fig. 1).

The chlorophylls and β-carotene showed higher solubility in acetone than in supercritical CO_2, due to the differences in the physical-chemical characteristics of the acetone and supercritical CO_2, expressive polarity of acetone and slightly polarity of supercritical CO_2.

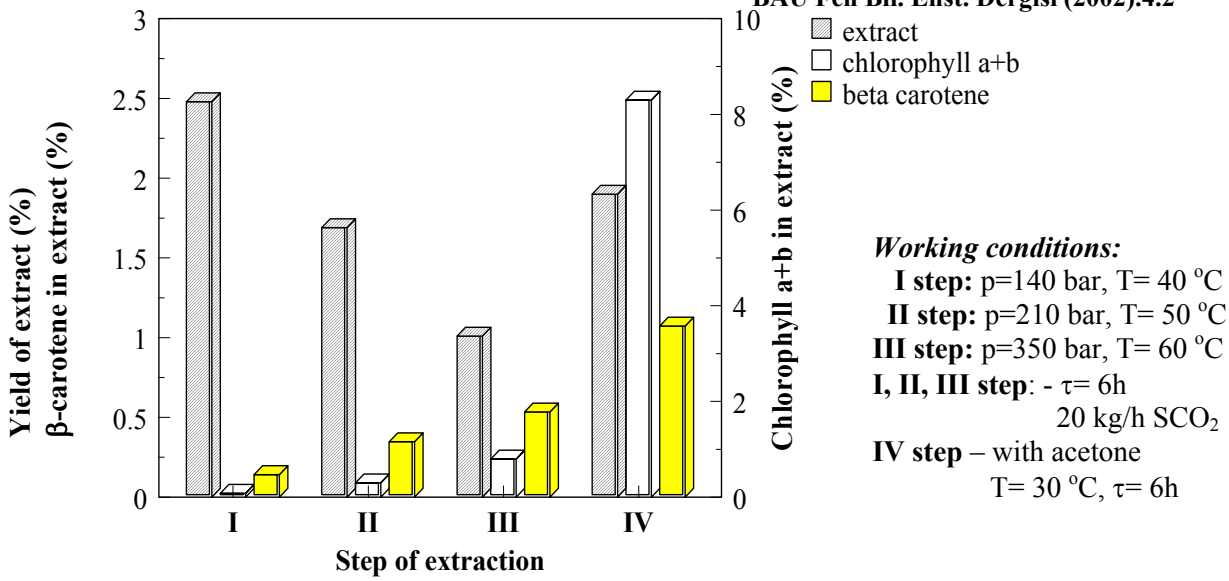

Figure 1. "Step-wise"extraction of stinging nettle

2. Influence of Pressure and Temperature of extraction i.e. density of supercritical CO_2

With increase of the pressure and the temperature at extraction of stinging nettle i.e. with increase of the density of supercritical CO_2 from 0.750 g/cm^3 to 0.855 g/cm^3, the extracted quantity of extract and content of chlorophyll *a+b* and β-carotene in extract increase (Fig. 2).

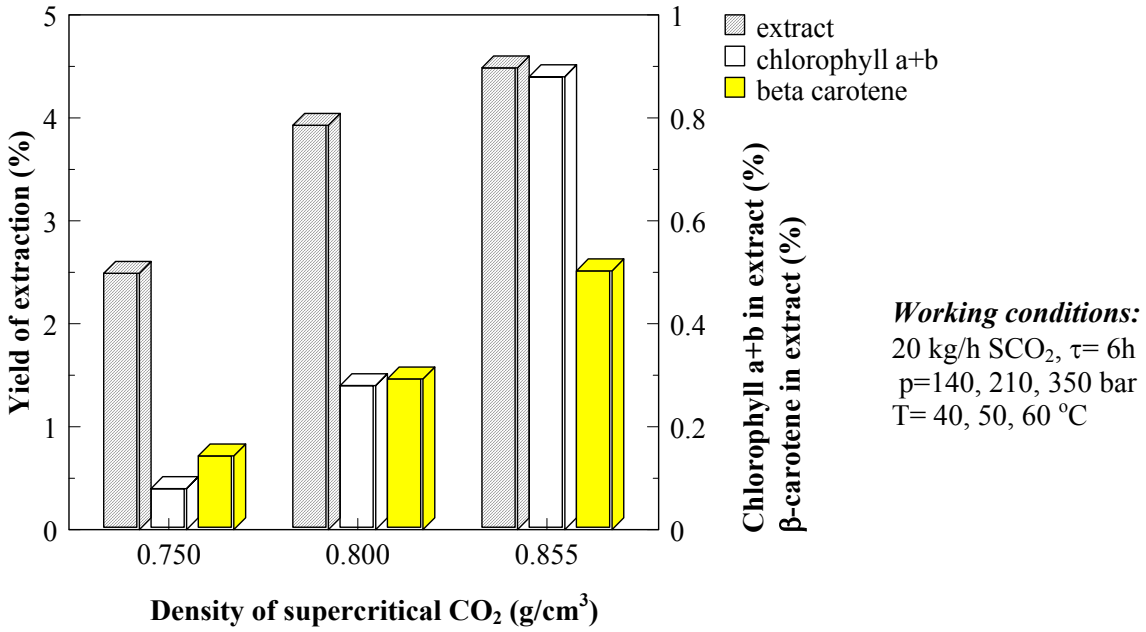

Figure 2. Extraction of stinging nettle at different density of supercritical CO_2

The yield of stinging nettle extract obtained at 0.750 g/cm^3, 0.800 g/cm^3 and 0.855 g/cm^3 density of supercritical CO_2 is adequately 2.49%, 3.93% and 4.48% (Fig. 2). The

biggest quantity of chlorophyll *a*+*b* (0.88%) and 0.50% content of β-carotene is determined in the extract of the stinging nettle obtained at 350 bar pressure and 60 ^{o}C temperature (Fig. 2). Under those conditions, the used CO_2 for extraction had the biggest density (0.855 g/cm^3). The increase of the extracted quantity of extract from the stinging nettle increasing the extraction pressure and temperature is a result of the increased density of the extraction means and its improved transport features. Because of this, for some components from the plant materials the dissolving power is increased.

The lower values for the extracted quantity of extract and the contents of coloured materials in it, obtained at the extraction of stinging nettle with supercritical CO_2 at different pressure and temperature (Fig. 2), compared with the cumulative values for the same responses of the system obtained at the "step-wise" extraction (Fig. 1) are a result of the fact that at the "step-wise" extraction in every subsequent stage is used the same extracted raw materials on which every time is applied new quantity of the extraction means. With this the dissolving power is increased, which enabled obtaining bigger quantity of extract and some components which are of interest for the extraction.

4. Conlusions

The results obtained at the extraction of the stinging nettle leaf with CO_2 at supercritical conditions guide to the following acknowledgements:

- The "step –wise" extraction of the stinging nettle performed at 140 bar and 40 ^{o}C, 210 bar and 50 ^{o}C, 350 bar and 60 ^{o}C, enables obtaining of bigger quantity of β-carotene in the first step of the "step-wise" extraction. In the extract obtained at the third step of the "step-wise" extraction (350 bar, 60 ^{o}C) the biggest quantity of chlorophyll *a*+*b* is determined.
- By increase of the pressure and temperature i.e. density of supercritical CO_2 increases the extracted quantity of extract where in bigger quantity are present the coloured components chlorophyll *a*+*b* and β-carotene.

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