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Pressure effect on ginger essential oil extraction by supercritical carbon dioxide and steam distillation

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ABSTRACT

Ginger is a plant with Asian origin and it is widely used in food and pharmaceutical industries. Its essential oil obtained by supercritical fluid extraction (SFE) or steam distillation (SD) is composed mainly of α-zingiberene, among a series of monoterpenes, terpenoids and sesquiterpenes compounds that present many antioxidant and antibacterial activity. This study aims to find the best conditions for the extraction of *Zingiber officinale* essential oil using the SFE and SD techniques, regarding the maximum oil yield and aromatic potential. Principal Component Analysis (PCA) is used to evaluate the similarity between the composition of the essential oil in different pressures and extraction methods. The SD extractions were performed in a pilot unit at three different pressures (1, 2 and 3 bar) using 2 kg of ginger rhizome. On the other hand, the SFE method was evaluated screening the process pressure (80, 90, 100 and 110 bar), maintaining the temperature fixed at 40 °C, using CO_2 as solvent and 0.2 kg of rizhome. Firstly, the plant was milled and submitted to the SFE with known humidity and all the experiments were performed in triplicate. A curve of accumulated mass *versus* extraction time was plotted and three different mathematical models (first order kinetics, Crank and Reverchon) were fitted for both methods, obtaining the relevant mass transfer parameters. The essential oil compounds were identified by gas chromatography coupled with mass spectrometry (GC-MS), with α-zingiberene as the main component with different contents (from 11.9 to 28.9%). The obtained data indicate that the best condition for the SFE is 100 bar, 40° C (0.0697 g_{oil}/g_{plant}) with 19.34% of α-zingiberene and for the SD extraction the one performed at 3 bar (133° C) (0.007 g_{oil}/g_{plant}) with 28.9% of α-zingiberene, measured on a dry basis.

Keywords: *Zingiber officinale*, SFE, PCA, modeling.

INTRODUCTION

Ginger (*Zingiber officinale)* is a plant of Zingiberaceae family, which is characterized by an herbaceous and perennial plant; its rizhome is widely used in food and pharmaceutical industry [1]. The plant has Asian origin, with India being the biggest producer in 2017 [2]. Many ginger proprieties have been already proven by scientific studies, as anti-inflammatory activity [3], antibacterial activity [4] and hypoglycemic activity [5], among others. Anticancer properties of determined compounds of ginger as 6-gingerol and 6-shogaol were studied and demonstrated effective against lung [6], ovarian [7], liver [8] and skin [9] cancers.

Rhizomes of ginger have a volatile oil composed of many monoterpenes (5%), sesquiterpenes (65%) and oxygen compounds (30%). This last one found in the oil-resinn

[10]. These compounds are responsible for its characteristic flavor, which the main compound is the sesquiterpene α-zingiberene.

The steam distillation is a common method for natural products extraction; however, it has many disadvantages such as the use of high temperatures, which may degrade thermolabile compounds, solvent residue in the product and solvent wastes. In contrast, supercritical fluid extraction (SFE) can be used as an alternative, which uses non-toxic solvents as carbon dioxide and promotes a complete separation from the product; however, the implementation costs are still a barrier for this type of process [11-13]. Different extraction methods lead to products with different compositions, mostly because of the solvent-extract interactions. The Principal Component Analysis (PCA) performs a statistical comparison between different sets of data, and is widely used in the case of essential oils extracted in different conditions [14].

On the other hand, the mathematical modeling of the extraction process is an important step for its scale up. A great variety of models that can describe solid-liquid extractions: power law models have an extensive use in adsorption processes [15]. The extraction process can also be described using a mass transfer model based on diffusive mechanisms with association to equilibrium-phase, as the one described by Crank (1979), which can be modified according to process restrictions. Another option is the use of a differential mass balance for each phase solved through different numerical methods [16].

In this work, the pressure effect is evaluated for two different process, steam distillation and SFE. The composition of the essential oil was determined by gas chromatography coupled to mass spectrometry (GC-MS) analysis and the essential oil composition at different pressures and extraction methods were compared from a statistical analysis. In addition, three different models are used to fit the experimental data: a kinetic model based in a power law, a model based in the diffusion mechanism related to phase-equilibrium and a model build by a differential balance in the extraction bed. A comparison between the models was made and the relevant parameters were obtained.

MATERIALS AND METHODS

Sample preparation

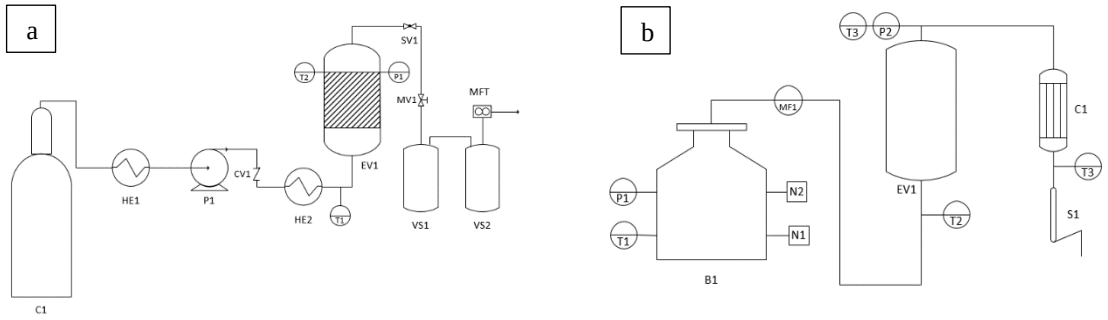
The rhizomes of *Zingiber officinale* were cut into irregular pieces, without removing its bark. The moist plant was milled using a knife mill and a sample was collected in order to quantify its humidity (thermogravimetric balance - BEL Engineering) as well as its thickness.

Extraction Methods

The supercritical fluid extractions (SFE) were carried out on pilot-scale equipment represented in a schematic diagram (Figure 1a). The pilot unit has a high-pressure pump (Maximator-G35) for carbon-dioxide (P1), a storage cylinder of CO_2 (C1), two pre-heaters (HE1, HE2), a system to measure the CO_2 flow and two vessels for separation (VS1, VS2), which are made of glass (Ilmabol TGI Boro 3.3) [17]. The supercritical fluid extractions were performed in an extraction vessel (Waters) with 500 mL capacity, 6.3 cm diameter and 19 cm height. The investigated conditions were determined according to previous works range in order to obtain the volatile extracts using the SFE methodology: four different pressures (80, 90, 100 and 110 bar) at 40° C, with a 1000 $g.h^{-1}$ CO_2 flow rate [18-20] .

For the highest essential oil yield, the experimental mass *versus* time curve was built in triplicate for further mathematical modeling. Samples were collected with time

interval of 10 min in order to evaluate the extract mass until the plant exhaustion (extract mass constant after three consecutive measurements).



The steam distillation extractions were performed on pilot-scale equipment which is represented in a schematic diagram as shown in Figure 1b. The equipment has a boiler (B1) with a capacity of 20 L of solvent (water) and a power source of 2 kW, with level sensors (upper and lower), measuring pressure and temperature [21].

Figure 1. (a) - Supercritical extraction experimental apparatus: C – CO_2 cylinder, HE – heat exchanger, CV – check valve, P1 – CO_2 high pressure pump, EV – extraction vessel, T – temperature transmitter, P – pressure transmitter, VS – separation vessel, MFT – mass flow transmitter, SV – Shut-off valve. (b) – Steam distillation apparatus: B – boiler, EV – extraction vessel, C – condenser, S – separator, T – temperature transmitter, P – pressure transmitter, MF – flow measure, N – level switch.

The steam distillation process was performed with the same plant material submitted to the procedure described in sample preparation. In this case, the ginger mass was 2000 g and the extraction vessel used (EV1) has 9.4 L capacity, 31.3 cm height and 19.3 cm diameter. The extractions were conducted in three different absolute pressures (1, 2, and 3 bar). The procedure was done in triplicate, and for the highest extraction yield, the experimental curve yield *versus* extraction time was built by measuring the oil volume for each 5 minutes interval.

The essential oil specific mass was determined through the mass measure of 1 mL of the oil using an analytical balance (Marte AW220 e=± 0.0001g). This procedure was done in triplicate. The plant specific mass was determined by pycnometer (Quantachrome MVP-6DL) analysis.

Chromatographic analysis

Zingiber officinale essential oils were dehydrated using anhydrous sodium sulfate (Na_2SO_4 - Synth), and diluted in cyclohexane (1:2) (Merck). The chemical composition was determined using a gas chromatograph equipped with a mass spectrometer (Hewlett Packard e Agilent system GC/MS model 7890A and mass detector 5975C). The carrier gas was helium (0.8 $mL.min^{-1}$), injector temperature was 250 °C, volume injected was 0.2 mL, split mode with split ratio of 1:55. The capillary column was HP-5MS (Hewlett Packard e Agilient, 5% fenil metil silox, 30 m 250 mm 0.25 mm). The temperature programming was 60 °C (8 min), 60C -180 °C, 3C/ min, 180 °C (1 min), 180 C- 250 °C, 20C/min, 250 °C (10 min).

The components of the essential oils were identified by comparison of their Retention Index (RIs) on the column, determined in relation to a homologous series of n-alkanes, with those from pure standards or reported in literature. Comparison of fragmentation patterns in the mass spectra with those stored on the GC-MS databases was also performed.

Mathematical Modeling

Three mathematical models were used to simulate the supercritical fluid extraction and steam distillation of *Zingiber officinale*, based on different approaches. The first model is based in a power law equation [15]. In order to describe the extraction kinects the equation becomes as showed in equation 1.

$$\frac{dM}{dt} = k(\Delta M)^n = k(M_\infty - M)^n \quad (1)$$

Where M_∞ ($g_{extract}$) is the maximum oil mass (the essential oil mass in an infinite time), M is the oil mass in a determined time, n is the model order and k is the extraction rate constant (s^{-1}). Assuming the beginning of the extraction time being equal to zero, the solute concentration present in the solvent is zero. Then the first order model ($n=1$) is obtained through Equation 2, resulting in which is showed in equation 2.

$$M = M_\infty(1 - exp^{-kt}) \quad (2)$$

The second mass transfer model used came from the Fick`s law of diffusion for an infinite plate sheet, considering uniform initial concentration, symmetry of concentration and a constant concentration present in the surface. The diffusion coefficient is assumed constant during the extraction process. External mass transfer resistance is negligible. The sample geometry during the extraction is considered a slab with half of thickness equal to L (mm). The diffusion is assumed to be only in thickness direction. The proposed model was solved, and the solution presented by Crank [22] is expressed in the equation 3.

$$\frac{M}{M\infty} = 1 - \frac{8}{\pi^2}\sum_{n=0}^{\infty}\frac{1}{(2n+1)^2}\exp\left\{-\frac{(2n+1^2)\pi^2 Dt}{4L^2}\right\} \quad (3)$$

Where M and $M\infty$ are the mass in a determined time and an infinite time (maximum mass obtained in the extraction), respectively, D is the diffusivity of the solute inside the particle ($m^2.s^{-1}$), t is the extraction time (s).

The third model used in this work was based in the model developed by Reverchon (1996) [23]. The model consists of one-dimensional mass balance for the extract, assuming the hypothesis of a linear behavior for the solid-fluid phase equilibrium. Two independent variables, time (t) and the fixed bed height (z) was considered only, and the radial dispersion along the column is assumed negligible, by these assumptions the model was developed. The mass balance is given below (equations 4 and 5) [24].

Fluid phase mass balance:

$$\frac{\partial C(z,t)}{\partial t} = -\upsilon\frac{\partial C(z,t)}{\partial z} - \frac{1-\varepsilon}{\varepsilon}\rho_s\frac{\partial q(z,t)}{\partial t} \quad (4)$$

Mass balance in the solid phase:

$$\frac{\partial q(z,t)}{\partial t} = -k_{TM}[q(z,t) - K \cdot C(z,t)] \quad (5)$$

The concentration of the essential oil in the vapor phase is given by the function $C(z,t)$ and the concentration in the aromatic plant is described by the $q(z,t)$ function. Where υ is the interstitial vapor velocity; ε is the porosity of the bed; k_{TM} is the internal mass transfer coefficient; ρ_s is the specific mass of the aromatic plant and K is the equilibrium constant between the phases. The model also considers some initial and boundary conditions: $q(z,0) = q_0$ and $C(z,0) = 0$, q_0 is defined by the total amount of extract contained in the solid phase and the $C(z,0) = 0$ as a boundary condition.

RESULTS AND DISCUSSION

The average yield (dry basis) obtained through the experimental data acquired by the extractions is presented in Table 01. The average specific experimental mass of the essential oil obtained was ρ_{oil} = 0.8736 g.cm^{-3} and the plant specific mass determined by the use of the pycnometer was ρ_{plant} = 1.24 g.cm^{-3}.

Table 01. SD and SFE yield results.

Extraction method	Pressure (bar)	Global yield[a] ($g_{EO}/100g_{plant}$)
SD[b]	1	0.252
	2	0.252
	3	0.616
SFE[c]	80	3.30
	90	4.06
	100	5.08
	110	4.48

a = measured per 100 g of dried plant
b = saturated water vapor
c = fluid temperature 40 °C

According to the proposed methodology, the mathematical modeling was performed, and the results are presented in the Figure 2. The modeling was done for the highest yield extraction conditions (3 bar for SD and 100 bar for SFE). The estimated parameters values along with the determination coefficient (R^2) for each method are presented in Table 2.

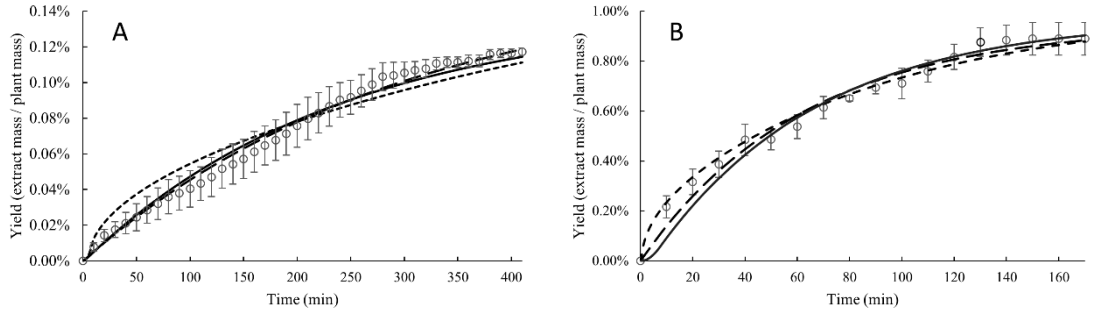


Figure 2. Steam distillation (A) and CO_2 supercritical fluid extraction (B) yield curves *vs.* time at 3 bar and 406.15 K (A) and 100 bar and 313.15 K (B) : (○) experimental data; (— —) first order model; (– –) Crank model; (▬) Reverchon model.

Table 2. Parameters obtained through the modeling of supercritical fluid extraction (SFE) and steam distillation (SD) data.

	First order parameters		**Crank parameters**		**Reverchon parameters**		
	$k \cdot 10^4$ s^{-1}	R^2	$D \cdot 10^{13}$ m^2.s^{-1}	R^2	$K \cdot 10^4$ m^3.kg^{-1}	$k_{TM} \cdot 10^4$. s^{-1}	R^2
SD	0.568	0.9904	1.04	0.9217	2.560	0.702	0.9863
SFE	2.670	0.9809	5.73	0.9838	0.011	2.780	0.9768

The coefficient of determination corresponds to the adhesion of the model to the experimental data. For the three models the coefficient indicates that the models are good and representative of the experimental data. However, each model has a level of complexity, and thus, different applications. For a scale up the best model would be Reverchon, which allows modifications to the input variables and a series of additions like vessel geometry, bed porosity and solvent flowrate. In this way, the Reverchon model could be used for further simulations with different equipment and scales.

Analysis of the *Zingiber officinale* essential oil

The main compounds identified by chromatographic analysis of the *Zingiber officinale* essential oil extracted by steam distillation at 1 , 2, and 3 bar were respectively: α-zingiberene (19.1%, 20.39% and 28.9%), β-sesquiphellandrene (6.820%, 7.920% and 10.610%), camphene (8.41, 5.59 and 3.86), and geranial (8.09%, 5.42% and 1.33%). The total identification of the compounds was 76.880%, 75.860% and 72.530% to 1, 2, and 3 bar, respectively. These major compounds found for the *Zingiber officinale* essential oil of are in accordance with those found by Alhassane and Zhang [25].

The main compounds identified in the extract of *Zingiber officinale* obtained by supercritical fluid extraction unit at 80, 90 100, and 110 bar and 40 °C were, respectively: α-zingiberene (18.123%; 15.471%; 19.384% and 19.355%), geranial (13.909%; 17.617%; 21.182% and 25.054%), Z-α-bisabolene (8.476%; 6.768%; 8.543% and 8.586%) and neral (8.278%; 6.952%; 7.081% and 7.750%). The total identification of the compounds was 90.073%, 90.149%, 92.299% and 93.219% to 80, 90, 100, and 110 bar at 40 °C.

Principal Component Analysis (PCA)

From the Principal Component Analysis, it was observed that six compounds stood out from the others, geraniol, neral, ar-curcumene, β-sesquiphellandrene, geranial and α-zingiberene (Figure 3). Since the constituents geranial and α-zingiberene present the main composition areas their covariance in relation to the others is also the greatest.

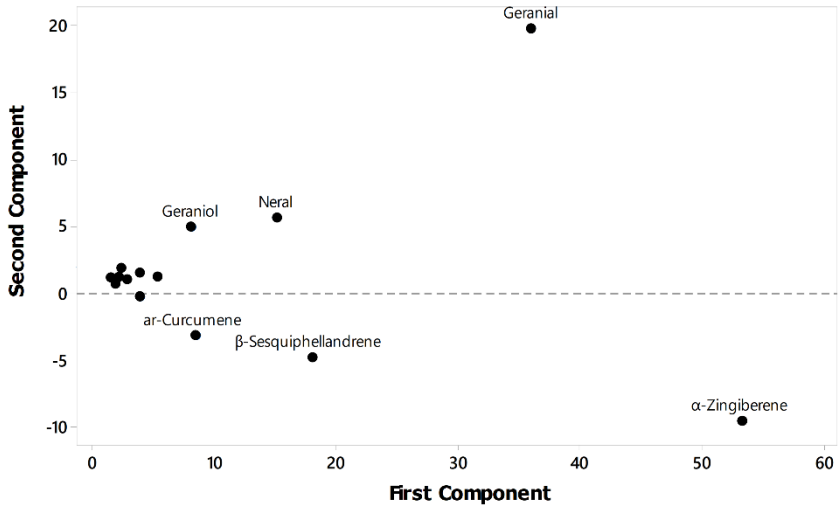


Figure 3. Variation behavior of the essential oil compounds of *Zingiber officinale*.

In Figure 4, which represents the behavior of the *Zingiber officinale* essential oil composition related to the variation of the extraction pressure and the extraction method it is possible to observe the formation of four distinct groups. This clear division of the groups is attributed to the similarity of the components percentage area obtained by GC/MS.

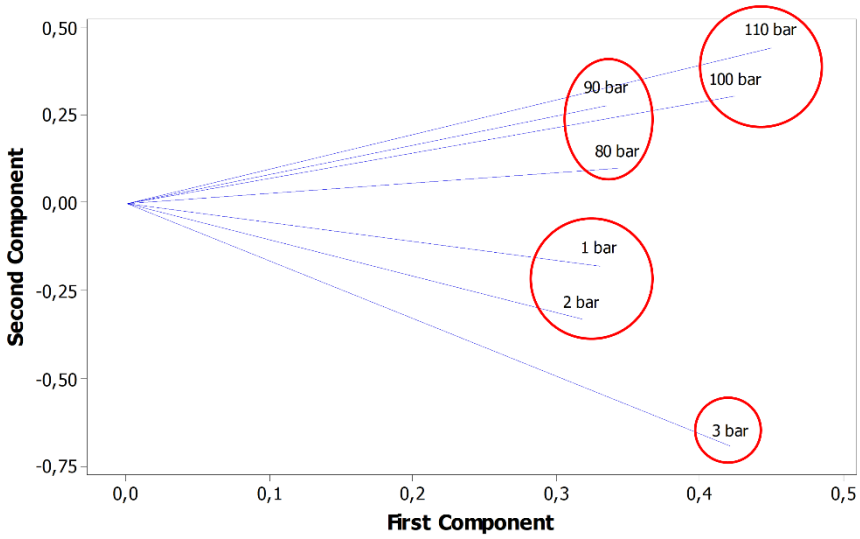


Figure 4. Variation behavior of the *Zingiber officinale* essential oil in relation to the different pressures and extraction methods.

CONCLUSION

The *Zingiber officinale* essential oil was obtained through two different extraction methods. For each method, the experimental mass *versus* time curve was built for the highest yield condition. Three mathematical models, with different complexities, were used in order to simulate the extraction process and significant mass transfer parameters were fitted from the experimental data. All the samples were analyzed by GC/MS and statistically compared through a PCA. For the SD essential oil there are similarities between the 1 and 2 bar compositions. The SFE essential oil presented two similar groups, between 80 and 90 bar, and between 100 and 110 bar.

The steam distillation extractions were performed in a pilot unit at three different pressures (1, 2 and 3 bar). The SFE method was evaluated by screening the process pressure (80, 90, 100 and 110 bar), maintaining the temperature fixed at 40 °C. The best results, in terms of yield, were 3 bar and 100 bar.

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