Fundamentals and experimental studies in essential oil extraction by instant autovaporisation using the instant controlled pressure drop DIC technology

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Essential Oils and Aromas Green Extractions and Applications

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Farid CHEMAT is Professor of Chemistry and Director of the laboratory for green extraction techniques of natural products (GREEN) at the Université d'Avignon et des Pays de Vaucluse, France. Born in Blida - Algeria (1968), he received his engineer diploma (1990) in organic process engineering from the University of Blida (Algeria) and his doctoral (1994) degree in process engineering from the Institut National Polytechnique de Toulouse where he worked with Professor Jacques Berlan on microwave and ultrasound reactors suitable for organic synthesis. After periods of postdoctoral research work with industrial companies Rhône Poulenc (1995), Unilever (1996) and Prolabo-Merck (1997) on laboratory and pilot scale continuous microwave and ultrasound reactors, he spent two years (1997-1999) as senior researcher in the group of innovative technologies of Dr Paul Bartels of the University of Wageningen (The Netherlands). In 1999, he moved to the University of La Réunion (France) to start his independent academic career. He obtained his "Habilitation à Diriger les Recherches" in 2004 in food and natural products chemistry and since 2006 holds the position of Professor of Chemistry at the University of Avignon (France).

His research activity is documented by more than 80 scientific peer-reviewed papers, and about the same number of communications to scientific meetings, 10 book chapters and 4 patents. His main research interests have focused on innovative and sustainable extraction techniques for food, pharmaceutical and cosmetic applications. His research group GREEN (Groupe de Recherche en Eco-Extraction des produits Naturels) at the University of Avignon has been involved in the area of microwave, ultrasound and new solvents for sustainable extraction of natural products at laboratory and industrial scale since 2006 and today his laboratory features a variety of state-of-the-art dedicated microwave and ultrasound instruments from all major equipment manufacturers. During the last few years his lab has authored over 50 original research articles, reviews and book chapters on microwave and ultrasound-assisted extraction.

In addition to his regular consulting and lecturing on green extractions, he is a short course instructor on microwave and ultrasound extraction for PEIFL (Pôle Européen d'Innovation en Fruits et Légumes) and PASS (Pôle Arômes Senteurs et Saveurs). He initiates a new Master degree in food technology focussed on sustainable extractions of food ingredients. He has also organized symposia, conferences, short courses and exhibitions on microwave and ultrasound as sustainable extraction techniques for education, research laboratories, and industries.

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Fundamentals and Experimental Studies in Essential Oil Extraction by Instant Autovaporisation Using the Instant Controlled Pressure Drop DIC Technology

Colette Besombes, Magdalena Kristiawan, Vaclav Sobolik, Karim Allaf *

Summary

Fundamental studies of transfer processes in steam extraction (steam-distillation) lead to identify a paradoxical situation where temperature gradient and the gradient of the partial pressure of essential oil vapour are both directed toward the centre; implying a motion completely opposite to that required for the sought extraction operation; the steam distillation is then a front progression operation. In order to intensify the operation of essential oil extraction, improve the quality of both extract and residue, the instant controlled pressure drop (DIC) treatment was used as a direct extraction – separation treatment. DIC treatment allows volatile molecules to be removed by both evaporation (during the high temperature -high steam pressure stage), and autovaporization by instantaneously dropping the pressure. This last is controlled through a total pressure gradient (Darcy-type law) allowing its kinetics to greatly increase. In addition, such great kinetics (processing time is about 4 minutes instead of some hours in hydro or steam extraction) are linked to low heat energy consumption. The final extracted essential oil could answer the various quality requirements in terms of absence of solvent and thermal degradation. Finally, this DIC treatment also allows the compact vegetal pieces to expand, and its technological aptitude to a second operation (drying, solvent extraction ...) to increase with higher kinetics avoiding the local high temperature generally due to the crushing.

Introduction

For the last 3000 years essential oils have been known, used, and studied. These natural products were originally exploited in religious rituals and for medicinal purposes in Egypt, Iraq, China, Iran, and India. Historically, the first essential oil produced for perfumery was rose oil, and there are references to the production of essential oils going back to the late 13th century. Nowadays, natural molecules are getting growing interest in various industrial

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*Abdul Karim Salim ALLAF, University of La Rochelle (France) fields (food, cosmetics and pharmaceutics) [1- 6]. Essential oils well known for their aromatic uses, antimicrobial properties, and even aromatherapy field, are usually extracted using the hydro distillation or the steam extraction; such techniques have had very little change until recently. The both ones allow heat to be provided from the boiling water or the steam by convection and/or condensation. At operational level, these techniques which are evaporation, condensation, and separation of essential oils and water, have in common a particularly slow kinetics (several hours to several tens of hours), which implies a large consumption of energy and some degradation of sensitive molecules [7-27].

The essential oils are generally a volatile mixture of organic compounds derived from a single botanical source. These oils, sometimes composed of hundreds of chemical aromas and compounds, are primarily responsible for the characteristic smell of the natural plant source. A main part of essential oils exist as they are extracted in the natural source material, but certain oils are formed only as a result of an enzymatic reaction once the plant tissues have been crushed or specially treated. Essential oils are located throughout the plant tissue or in special sacs, cells, glands or ducts found in several parts of plants such as flowers, leaves, bark, stems, seeds and roots.

Many of the developing countries have vast potential in cultivating the herbs due to their climate, rainfall and geographical conditions. It is estimated that 65 % of the world production of essential oils is produced by developing countries such as India, China, Brazil, Indonesia, Mexico, Egypt and Morocco. However the USA is also a major producer of essential oils such as peppermint and other mints. The demand for essential oil by fragrance industry is 60 %, flavour industry 20 % and pharmaceutical industry 20 %. Globally, the essential oil industry - valued at around $10-billion is enjoying huge expansion as consumers increasingly gravitate towards naturals in flavour and fragrance. This vibrant essential oils industry would bring substantial and sustained benefit to local communities through job creation (essential oil sector is a labour-intensive crop), strengthening regional economies and creating new development modes.

Various different methods have been used for the extraction of essential oils and fragrances from aromatic plants for commercial purposes as well as for research. The main operations are the steam or water distillation. Some other methods include Soxhlet extraction, organic solvent extraction as subsequent or concurrent steam or water distillation. Losses of some volatile compounds, long processing time, low extraction efficiency, compounds degradation and toxic solvent residue in the extract may be encountered using these conventional essential oil extraction methods [21,22]. The solvents residue levels in the extracts are becoming unacceptable to many legislators and regulators in the food and pharmaceutical industry. This is likewise the case with many consumers, who are becoming acutely aware of the toxic nature of the residuals. And waste disposal of solvents is an additional problem, adding extra charge for the environment.

The modern process such as supercritical fluid extraction, mainly using carbon dioxide, allows us to obtain a high quality extract, but the high fixed cost as well as the low kinetics due to the weak diffusivity within the plant as porous medium, reduce its application to the only high added value products [28,29]. Moreover, this process is less convenient for polar compounds and it extracts also culticular waxes and lipids [30,31]. The green technologies for extraction of essential oil from aromatic plants, such as vacuum microwave hydrodistillation (VMHD) and solvent free microwave extraction (SFME), have been successfully developed [25,32-37]. They consists in combining microwave heating and dry distillation, performed at atmospheric pressure (SFME), and followed by slow pressure drop rate into vacuum (VMHD), without added any solvent or water. These processes yielded better quality extracts (more oxygenated compounds) with faster extraction kinetics than the conventional ones. However, Klima [38] reported the difficulty to reach uniformity of material temperature due to non-homogeneity of electromagnetic field during microwave heating.

Allaf *et al.* [39,40] developed a new process, the instant controlled pressure drop (DIC), which isolate high quality essential oils in a short time, with no organic solvent required. Rezzoug *et al.* [41] reported that in the case of isolation of rosemary oil, one DIC cycle during 10 minutes at a low pressure (0.1 MPa) had an isolation efficiency of 95.8 %. On the other hand, more severe DIC conditions were needed for the isolation of oil from juniper berries and cananga flowers. Mellouk *et al.* [42] needed two DIC cycles at a pressure of 0.6 MPa to isolate within 150 s 95 % of the juniper oil as against 12 h for steam distillation. Kristiawan *et al.* [43] found that a cananga oil yield of 2.77 % of dry matter which is comparable with the yield of 24 h of steam distillation was obtained with eight DIC cycles at 0.6 MPa in 4 min.

Fundamentals

The plants can often be considered as porous medium. In this medium, processes of heat and mass transfer intervene. Special zones implied in the secretion of essential oils are located within plant tissues (deep structures) or on the surface of the plant (peltate glandular trichomes). Process of the essential oil extraction should be dependent on the oil storage and the type of the zones of secretion. A certain amount of heat is needed to allow the product to get higher temperature; however, the main part of the heat is used for a phase change (liquid - vapour) with a partial pressure, which is a temperature function.

Case of steam extraction

In steam extraction, the external transfer processes are the heat transfer from the outside towards the external surface of the grain, and the mass transport (vapour of water and other volatile molecules) from the surface of grain to the surrounding environment to reach the condenser. In this operation, the driving force in the surrounding atmosphere, which is the gradient of partial pressure, can be intensified by increasing the temperature of exchange surface of the material, by relatively decreasing the condenser temperature, or by setting them as close as possible. The heat is absorbed at the exchange surface with the steam by convection as well by partially condensation of steam.

In steam extraction, the surface temperature rises very rapidly the steam temperature, and gradually spreads heat within the solid. The internal heat transfer is mainly processed by conduction; the global phenomenon may be traduced by a like conduction transfer where the gradient of temperature as a driving force with a higher value of effective conductivity. Such an effective conductivity of porous structure, closely depending of the presence of air and water, is as higher as the evaporating / condensation phenomenon intervenes inside the holes. The main part of such heat flow is used for evaporating liquid water and essential oils.

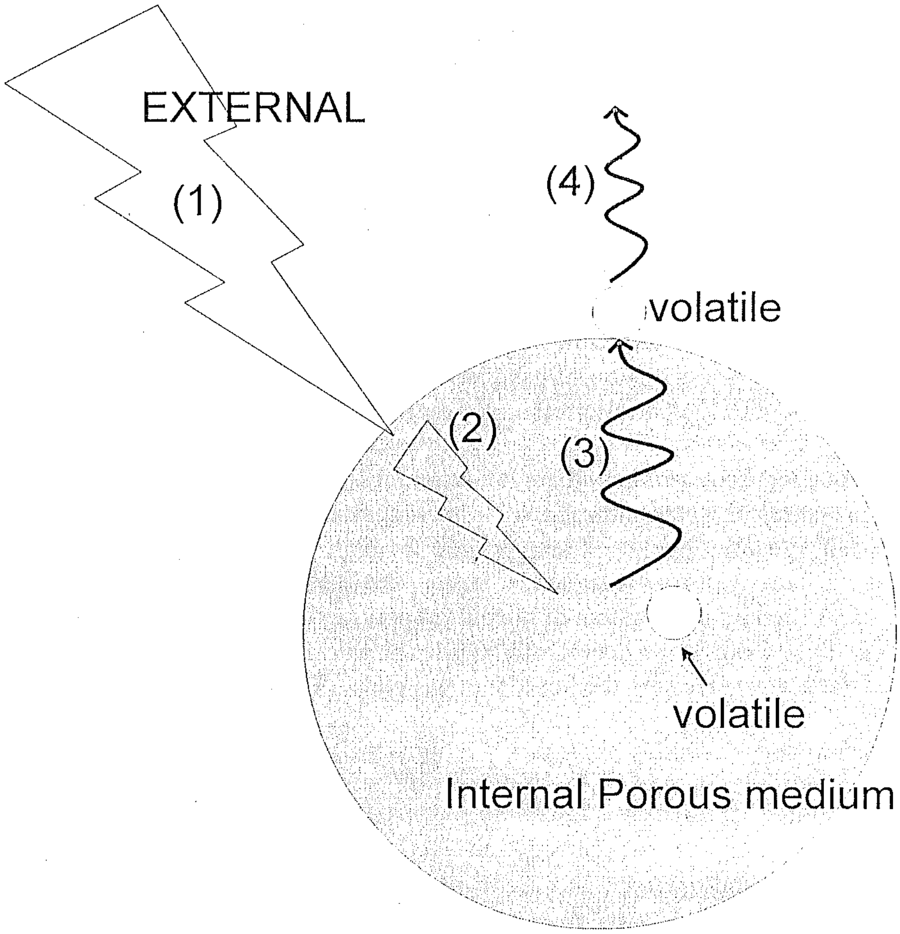


Figure 1. Diagram illustrating the transfer processes in the steam extraction of volatile compounds. (1) Heating the material surface occurring by convection and by condensation of steam. (2) Internal heat transfer through effective conduction. (3) Internal evaporation and migration of various volatile molecule compounds. (4) Transport of volatile molecules in the atmosphere with, as a driving force the partial pressure gradient of each of the compounds.

The internal evaporation inside the holes closely depends of temperature through the partially pressure of water and each volatile compounds; to study the operation, one would define the thermodynamic behaviour of each molecule normally leading to determine the partially vapour pressure at different temperatures.

A mass transfer process may be done by complex phenomena, depending on the nature and the localization of the molecules and the phase in which they appear and migrate. This operation may be conducted by capillary force when it is a liquid / solid interaction or by diffusion of the gas phase (interaction gas / solid). It also depends on several factors, including temperature, but also the structure and the morphological state of matter (degree of porosity, specific surface area, the permeability of secretion element walls...). In steam extraction, it can be assumed negligible the transfer of essential oils under the liquid phase. The main transfer phenomenon which can be considered is a diffusion of gas phase, which can be traduced by a Fick-type law related by an effective diffusivity D_{eff}, and the partial pressure gradient of each volatile compound. However, the intervention of the structure, the presence of secretion element walls as barriers and more generally the porosity of the material contribute to the macroscopic mass transfer inside of the material.

Whatever the type of the mass transfer within the plant as a porous medium, it is assumed to be the limiting process because it would be much slighter than the heat transfer. The transfer of essential oils is assumed to be negligible under a liquid phase; the main mass transfer phenomenon is a diffusion of gas phase, which can be traduced by a Fick-type's law with the gradient of the partial pressure of each volatile compound as driving force.

Paradox of coupled heat and gas diffusion transfers

The partial pressures of water and each volatile compound inside the holes of the plant as porous medium closely depend of temperature through the thermodynamic liquid/gas equilibrium of the mixture implying each molecule. The diffusion of any gas within a porous medium is controlled by the gradient of the partial pressure as driving force. In the present case, the essential oil extraction necessarily implies an amount of heat capable to change, in any point of the porous medium, the liquid into gas phase. The heat transfer is traduced by:

$$div\vec{\varphi} + \left(\rho_s c_{p_s} + \rho_e c_{p_e} + \rho_w c_{p_w}\right)\frac{\partial T}{\partial t} + \frac{\partial\left(p_e \varepsilon \frac{M_e L_e}{RT} + p_w \varepsilon \frac{M_w L_w}{RT}\right)}{\partial t} = 0 \qquad (1)$$

Where:

T: the temperature (K),
t: the time (s),
φ: the heat flow within the porous material ($W.m^{-2}$),
ρ_e: the apparent density of liquid essential oils in the material ($kg.m^{-3}$),
ρ_w: the apparent density of liquid water in the material ($kg.m^{-3}$),
ρ_s: the apparent density of dry material ($kg.m^{-3}$),
c_e: the specific heat of liquid essential oils in the material ($J.kg^{-1}.K^{-1}$),
c_w: the specific heat of liquid water in the material ($J.kg^{-1}.K^{-1}$),
c_s: the specific heat of dry material ($J.kg^{-1}.K^{-1}$),
ρ_e: the partial pressure of essential oils in the material (Pa),
ρ_w: the partial pressure of vapour in the material (Pa),
M_e: the Molar mass of essential oils ($kg.mol^{-1}$).
M_w: the Molar mass of water ($kg.mol^{-1}$).
L_e: the essential oil vaporisation latent heat ($J.kg^{-1}$).
L_w: the water vaporisation latent heat ($J.kg^{-1}$).

As the interne transfer is carried out by conduction:

$$div\left(-\lambda.\overrightarrow{gradT}\right) + \left(\rho_s c_{p_s} + \rho_e c_{p_e} + \rho_w c_{p_w}\right)\frac{\partial T}{\partial t} + \frac{\partial}{\partial t}\left[\frac{\varepsilon}{RT}\left(p_e M_e L_e + p_w M_w L_w\right)\right] = 0 \qquad (2)$$

Where

λ: the global conductivity of the porous wet material ($J.m^{-1}.K^{-1}$),

A main part of heat transfer is "only" used for evaporating essential oils and water; the repartition of temperature may be assumed to be stationary during the main part of the operation:

$$div\left(-\lambda.\,gr\vec{a}dT\right)+\frac{\partial}{\partial t}\left[\frac{\varepsilon}{RT}(p_eM_eL_e+p_wM_wL_w)\right]=0 \tag{3}$$

The ALLAF's [44] formulation of Fick-type law may be traduced by

$$\frac{p_e}{\rho_s}(\vec{v}_e-\vec{v}_s)=-D_{eff_e}\;g\vec{r}ad\left(\frac{p_e}{\rho_s}\right) \tag{4}$$

$$\frac{p_w}{\rho_s}(\vec{v}_w-\vec{v}_s)=-D_{eff_w}\;g\vec{r}ad\left(\frac{p_w}{\rho_s}\right) \tag{5}$$

By neglecting the possible shrinkage phenomena, one can assume that ρ_s = constant and v_s = 0, the equations (4 and 5) may be transformed into:

$$p_e\vec{v}_e=-D_{eff_e}\;\overrightarrow{grad}\,p_e \text{ and } p_w\vec{v}_w=-D_{eff_w}\;\overrightarrow{grad}\,p_w \tag{6}$$

As the external water vapour partial pressure is saturated, one can assume the internal vapour partial pressure p_w within the porous material as approximately constant, it can be postulated:

$$-\lambda.div\left(gr\vec{a}dT\right)+\varepsilon M_eL_e\frac{\partial}{\partial t}\left[\frac{p_e}{RT}\right]=0 \tag{7}$$

and

$$p_e\vec{v}_e=-D_{eff_e}\;\overrightarrow{grad}\,p_e \tag{8}$$

By assuming that each "particle" of plant is spherical, homogeneous, and isotropic, Equations 7 and 8 are transformed respectively into:

$$-\lambda.\frac{\partial^2T}{\partial r^2}+\varepsilon M_eL_e\frac{\partial}{\partial t}\left[\frac{p_e}{RT}\right]=0 \tag{9}$$

$$p_ev_e=-D_{eff_e}\frac{\partial p_e}{\partial r} \tag{10}$$

On the other hand, as p_e is as higher as the temperature is higher:

$$\frac{\partial p_e}{\partial T}>0 \tag{11}$$

The values of essential oil partial pressure p_e are then higher at the exchange surface than in the granule heart. As the essential oil flow obviously follows the temperature one, the result is a paradox [45-46] implying a motion completely opposite to that required for the sought extraction operation.

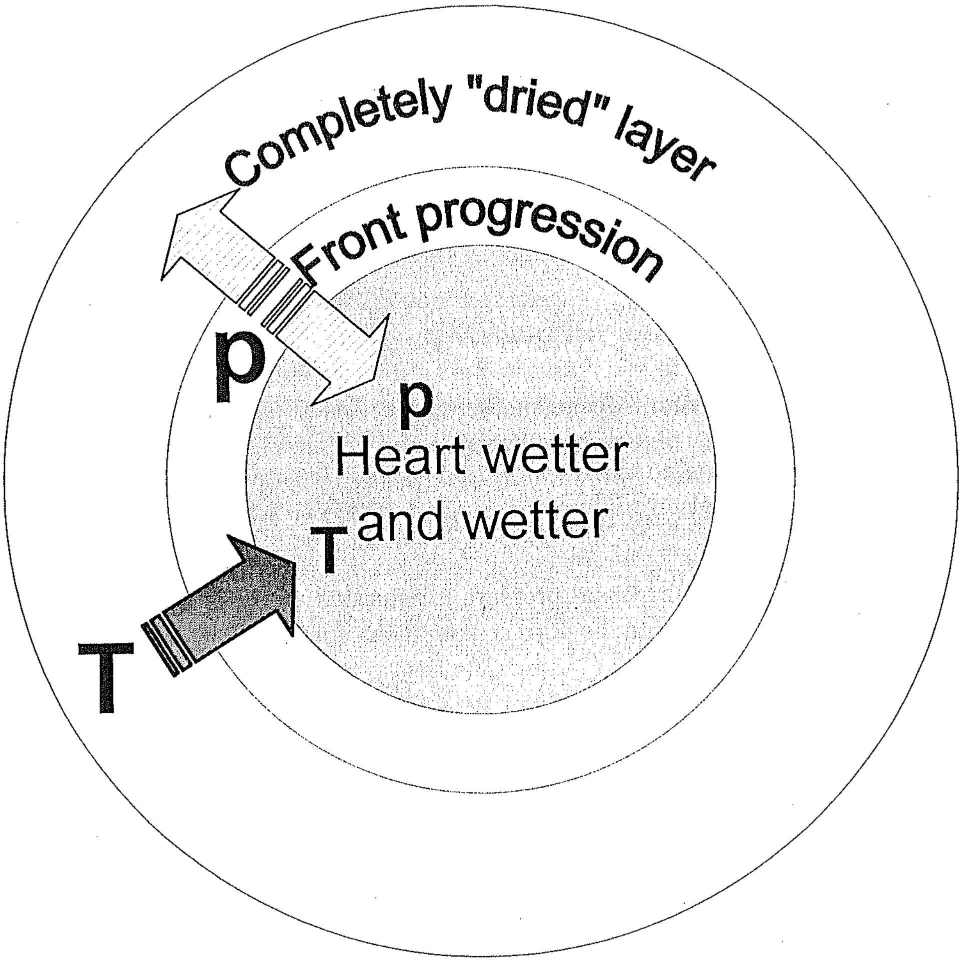
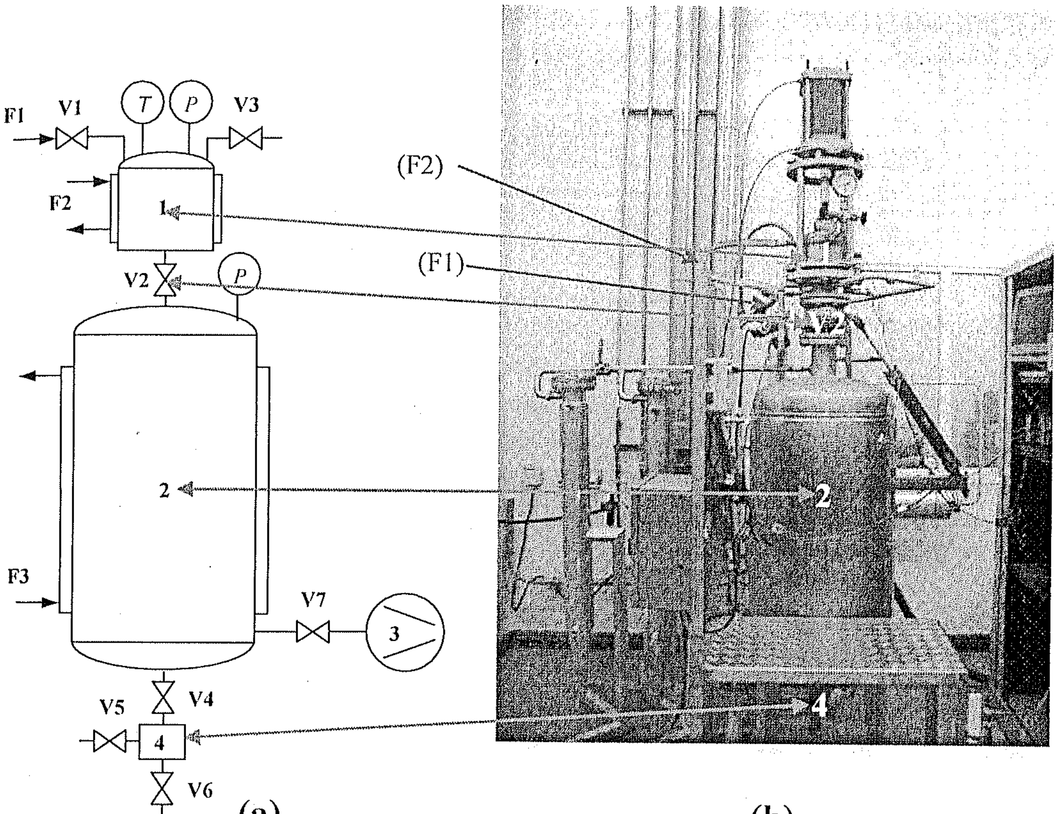


Figure 2. Paradox of heat and vapour transfers of essential oil extraction implying "front progression" kinetics.

In the usual steam extraction of essential oils, the operation is achieved by successive layers and the kinetics is a "front progression" one. This may further explain why in the steam extraction a grinding step is always required to reduce the granule shape. However, it would be possible to remedy this situation and make a notable improvement in the kinetics by adopting other manners for heating (microwaves) and / or moving such molecules (DIC process with a mass transfer by Total Pressure Gradient TPG process).

Essential oil DIC extraction

As essential oil extraction process, DIC is based on the thermal effects induced by subjecting the raw material for a short period of time to high pressure saturated steam with about 0.1 to 0.6 MPa according to the product (Figure 4-c and d); various essential oil compounds get then higher partial pressures than the ones gotten by usual steam distillation.



(a) (b)

Figure 3. Instant controlled pressure drop DIC apparatus.

(a): Schematic diagram and (b) Photo
(1): autoclave with heating jacket
(V2): rapid valve
(2): vacuum tank with cooling water jacket
(3): vacuum pump
(4): extract container
F1 and F2: steam flow
F3: cooling water flow

This high temperature – short time stage is followed by an abrupt pressure drop towards a vacuum at about 5 kPa (Figure 4-e); such an abrupt pressure drop, whose rate $\Delta P/\Delta t$ is higher than 0.5 MPa/s, provokes simultaneously autovaporization of volatile compounds, instantaneous cooling of the products which stops thermal degradation, and swelling even broking the cell and likely secretion element walls. The created porous structure then enhances mass transfer. Therefore, the mass transfer is not limited by internal diffusion as in the case of conventional extraction method. However, the main impact of DIC process is to remedy the paradox of the mass transfer of the essential oils and to get it as a gradient of total pressure (Darcy-type law with a Mass Transfer by Total Pressure Gradient), flowing from the heart of material to its surface [47].

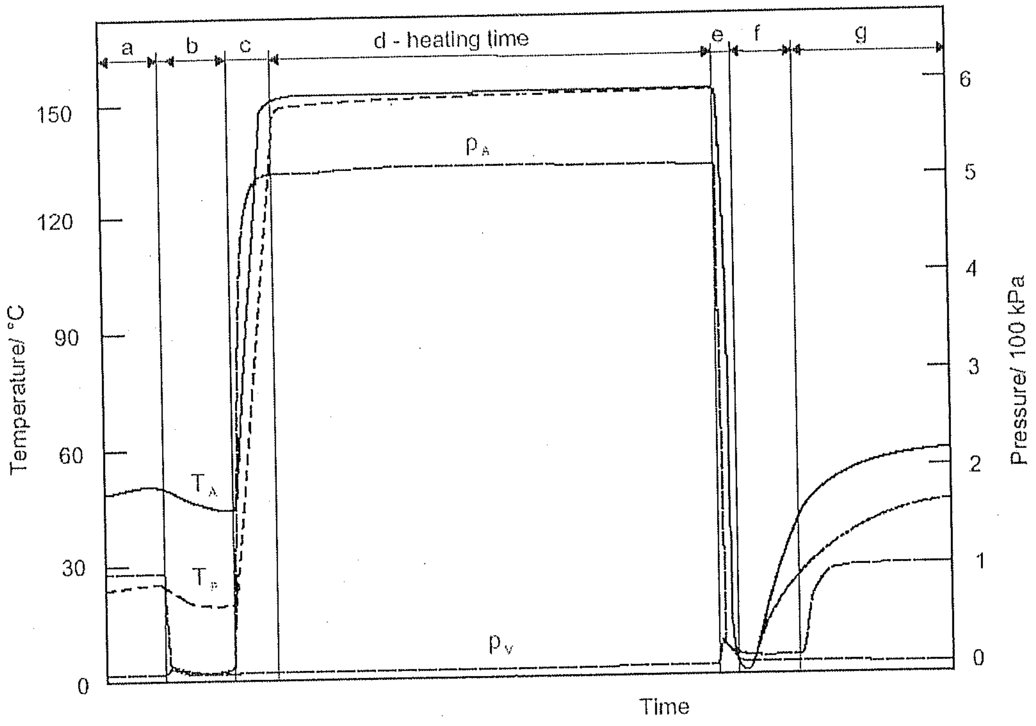


Figure 4. Temperature and pressure history of a DIC processing cycle

P_A: steam pressure in autoclave
P_V: pressure in vacuum tank
T_A: temperature in autoclave
T_P: temperature of product
(a): sample at atmospheric pressure
(b): initial vacuum
(c): saturated steam injection to reach the selected pressure
(d): constant temperature corresponding to saturated steam pressure
(e): abrupt pressure drop towards vacuum
(f): vacuum
(g): releasing to the atmospheric pressure

Usual analysis of various DIC stages

During the heating stage (d), the heat flux from the steam to the product is:

$$\varphi = hS(T_v - T) \tag{12}$$

And the quantity of heat during the thermal treatment time of a cycle is then:

$$Q_c = hS\langle T_v - T\rangle_{LN}.t_c \tag{13}$$

Where

Q_C: is the quantity of heat furnish by the steam to the surface of the product
h: coefficient of heat transfer by condensation and convection ($W.m^{-2}.K^{-1}$)
S: exchange surface (m^2)
$\{T_V - T\}_{IN}$: is the logarithmic mean of the difference of temperature between the steam (T_V) and the product (T),
t_C: is the duration time of the heating stage (d) of a DIC cycle.

During this heating stage (d), convection and condensation of saturated steam assure the establishment of the high temperature level at exchange surface with a very great value of h; the operation is very quick and may be considered as instant process. The initial vacuum state permits a closer contact between external product surface and steam (higher value of the surface S), intensifying thus such a process. During this same stage, two heat and mass transfers intervene within the product. The first is carried out by conduction with a Fourier-type law.

$$\varphi = -\lambda_{eff}.\overrightarrow{gradT} \tag{14}$$

The second is a normal Fick-type water diffusion mass transfer:

$$\frac{\rho_1}{\rho_2}(\vec{v}_1 - \vec{v}_2) = -D_{eff}\ \overrightarrow{grad}(\frac{\rho_1}{\rho_2})\ [44] \tag{15}$$

Where:

λ_{eff}: the effective conductivity of material ($W.m^{-1}.K^{-1}$)
$\rho_1 = \rho_w$: apparent density of liquid water in the material ($kg.m^{-3}$)
$\rho_2 = \rho_s$: apparent density of dry material ($kg.m^{-3}$)
$v_1 = v_w$: absolute velocity of liquid water flow within the porous medium ($m.s^{-1}$)
$v_2 = v_s$: absolute velocity of solid medium ($m.s^{-1}$)
$D_{eff} = D_{ws}$: effective diffusivity of liquid water within the solid medium (en $m^2.s^{-1}$)

By neglecting the possible expansion phenomena, one can assume that ρ_s=constant and $v_s = 0$, the equation (15) may be transformed into:

$$\rho_w \vec{v}_w = -D_{ws}\ \overrightarrow{grad}\rho_w \tag{16}$$

And by assuming a radial transfer, one can write:

$$\rho_w v_w = -D_{ws} \frac{d\rho_w}{dr} \tag{17}$$

By using mass conservation and the continuity, the second Fick law becomes:

$$\frac{\partial \rho_w}{\partial t} = div\left[D_{ws} \overrightarrow{grad} \rho_w\right] \tag{18}$$

where t is the time.
The diffusivity D_{ws} is assumed to be constant only through the hypothesis of both structural and thermal homogeneities. Indeed, D_{ws} considerably varies versus the porosity and temperature of the granule. A general quantification of physical processes and adequate experiments can normally confirm this hypothesis allowing the equation (4) to become the second Fick's law:

$$\frac{\partial \rho_w}{\partial t} = D_{ws} * div\left[\overrightarrow{grad} \rho_w\right] \tag{19}$$

And, by assuming a unidirectional flow, Eq. 19 becomes:

$$\frac{\partial \rho_w}{\partial t} = D_{ws} \frac{d^2 \rho_w}{dr^2} \tag{20}$$

The time t_c of such a stage (d) must generally be defined for allowing the both temperature T and moisture content W to be uniform in the product; usually, the first parameter is much more quickly established; the quantity of added moisture absorbed by the product from the surrounded saturated steam during this (d) stage is:

$$m_v = Q / L = m_s \left(c_{ps} + \left\langle W c_{pw} \right\rangle\right)\left(T_f - T_i\right) \tag{21}$$

Where:
m_v: is the mass of condensate vapor from the saturated surrounded steam (kg)
L: is the mean of the specific latent heat of condensation in water ($J.kg^{-1}$)
c_{ps}: is the specific heat at constant pressure of dry material ($J.kg^{-1}.K^{-1}$)
c_{pw}: is the specific heat at constant pressure of water ($J.kg^{-1}.K^{-1}$)
W: is the moisture content dry basis ($kg.kg^{-1}$)
$<Wc_{pw}>$: is the mean value of Wc_{pw} during the stage (d)
T_f and T_i: are respectively the final and initial temperatures of the product; they usually are $T_f = T_v$: the temperature of saturated steam, and $T_i = T_e$ the "equilibrium temperature"

After the pressure drop stage (e) and during the vacuum stage (f), the deep total pressure is mainly due to the vapour of water and essential oils in the porous medium. The transfer of essential oils within the porous plant as expanded granule is assumed to be carried out as gas phase transfer in a homogeneous and isotropic medium. The main part of the mass transfer is then carried out from the heart towards the surrounding medium through the gradient of the total pressures. A Darcy-type law traduces this transfer:

$$\rho_m\left(\vec{V}_m - \vec{V}_s\right) = -\frac{K}{\nu_m}\,\overrightarrow{grad}\,dP \tag{22}$$

Where:

ρ_m: is the apparent density of the essential oil and water vapour mixture ($kg.m^{-3}$)

V_m: is the velocity of the essential oil and water vapour mixture within the porous medium ($m.s^{-1}$)

V_s: is the absolute porous medium velocity ($m.s^{-1}$)

K: is the permeability of the essential oil and water vapour mixture within the porous medium (m^2)

V_m: is the kinematic viscosity of the mixture of essential oils and water ($m^2.s^{-1}$)

P: is the total pressure of the vapour of the mixture of water and essential oil (Pa)

By neglecting the expansion phenomena possibly due to the autovaporisation by instant pressure drop, one can assume that $V_s = 0$. On the other hand, as the transfer is achieved with gaseous phase of essential oils, and by assuming a radial transfer within a spherical shape material, Eq. 22 may be written as:

$$\rho_m\vec{V}_m = -\frac{K}{\nu_m}\frac{dP}{dr} \tag{23}$$

By using mass conservation and the continuity, and by integrating between the hole (whose radius R_o) and the external radius R_s of the spherical shape Eq. 23 becomes:

$$\dot{m}_m = 4\pi\frac{K}{\nu}\frac{\left(P_o - P_{ext}\right)}{\left(\dfrac{1}{R_o} - \dfrac{1}{R_s}\right)} \tag{24}$$

The value of the total pressure P_o in the hole decreases versus the time t, depending of the flux $\dot{m}_m$. The time t_v of such a stage (f) must generally be defined for allowing the vapour mixture to be transferred towards the surrounding medium.

One can assume the quantity of essential oil and water vapour mixture issued from the autovaporisation to be:

$$m_m = Q / L_m = m_p c_{pp}.\left(T_f - T_i\right) \tag{25}$$

Where:

m_m: is the mass of evaporated essential oil and water mixture of per DIC cycle (kg)

L_m: is the mean of the specific latent heat of the essential oil and water mixture evaporation ($J.kg^{-1}$)

c_{pp}: is the specific heat at constant pressure of the product ($J.kg^{-1}.K^{-1}$)

T_f & T_i: are respectively the final and initial temperatures of the product; they usually are $T_f = T_v$ the temperature of saturated steam, and $T_i = T_e$ the "equilibrium temperature"

The DIC process seems to be a highly relevant operation for remedying the paradoxical phenomenon and greatly intensifying the essential oil extraction process by using the Total Pressure Gradient TPG instead the normal diffusion phenomenon.

Experimental work

In the present work, the experimental work was carried out for analysing, optimising and modelling at the macro-scale. Various raw materials have been used, as well as different extraction processes and assessment methods. The treatments by hydro-distillation and steam distillation have been done as references to compare with the instant autovaporisation by instant controlled pressure drop DIC.

The main part of our investigation aimed to establish the effect of the DIC steam pressure, total heating time and number of cycles in terms of total yields, and the possibilities of fractional extraction of different compounds; it was achieved according to the protocol shown in Figure 5.

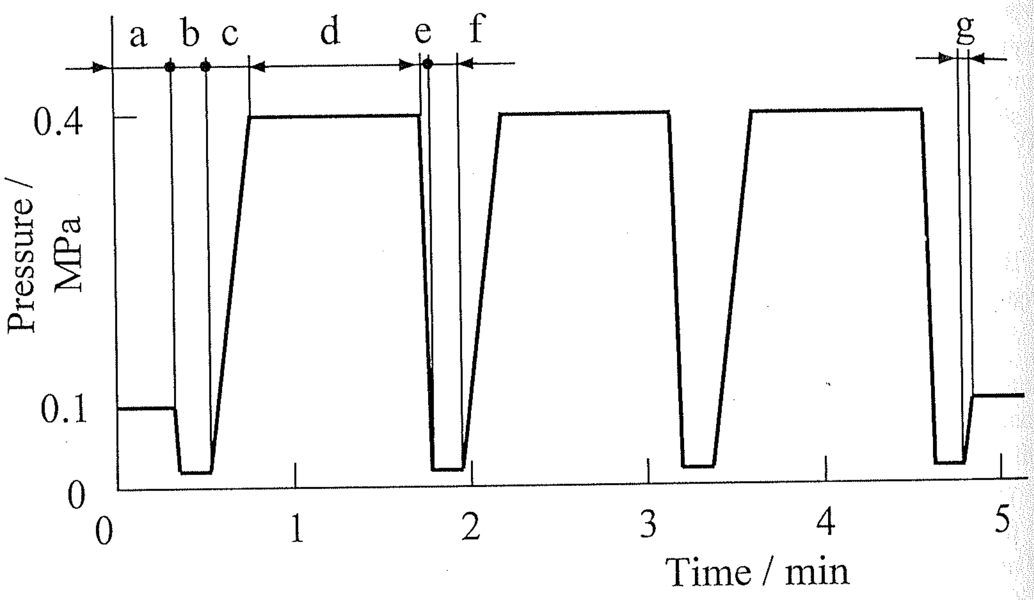


Figure 5. Pressure history in multi-cycle DIC process

The main part of our investigation aimed to establish the effect of the DIC steam pressure, total heating time and number of cycles in terms of total yields, and the possibilities of fractional extraction of different compounds; it was achieved according to the protocol shown in Figure 5.

The raw material

Grosso variety lavender used for the study [4, 48-50], was purchased from the Distillery Bleu Provence co. (Nyon; France). The main part of experiments was carried out at 20 % db as moisture content.

All other dry aromatic plants used in this research work were purchased from Cailleau Herboristerie (France). Their specifications can be found in Table 1. They were stored at room temperature prior to use. For angelica seed, it was ground into coarse powder prior to use (Grinder, 5000 rpm, 5 s).

Table 1. Raw material of dried aromatic plants

No.	Raw material and exploited part	Moisture content (% dm)	Essential oil secretor
1	lavender	20 %	Secretory cells and glandular trichomes
2	Angelica seeds	7.35 %	Secretory ducts
3	Cinnamon bark	13.70 %	Secretory cells
4	Roman chamomile flowers (petals)	15.97 %	Secretory ducts
5	Lemon grass leaves	13.47 %	Secretory cells
6	Patchouli leaves	12.03 %	Secretory cells and glandular trichomes
7	Thyme leaves	12.80 %	Glandular trichomes
8	Valerian roots	15.69 %	Secretory cells

Separation methods

The instant controlled pressure drop (DIC) process

The schematic diagram and the photo of the DIC apparatus are shown in Figure 3. Dry aromatic plants (40 g dm) were treated in batch mode in the 6 L autoclave (1) equipped with a double jacket, thermocouples and a pressure gauge. Steam used for plants heating was injected through the valve (V1). The autoclave was connected via 80 mm spherical valve (V2) to a vacuum tank (2) with a volume of 285 L. The valve was controlled by a rapid pneumatic actuator. The vacuum tank was cooled through a double jacket for condensation of the vapour of water and volatile molecules, evolved by vaporisation during the heating stage and by autovaporisation when the high pressure (abruptly) dropped. Condensate was recuperated through a trap (4). A water ring pump (3) maintained the tank pressure at about 4.5 kPa. The atmospheric pressure in the autoclave could be installed by opening a vent (V3).

Temperature and pressure histories of one DIC cycle are shown in Figure 4. After an initial atmospheric pressure stage (a), a vacuum of about 4.5 kPa is established in the autoclave (b), and, just after, a saturated steam is injected into the autoclave (c) and maintained at a fixed pressure level for a predetermined time (d); the initial (a) vacuum state allows closer contact of the steam as heating fluid which enhances the heat transfer towards the plants allowing the vessel to immediately reach the saturated steam temperature. After this thermal treatment the steam is cut off and the spherical valve is opened rapidly (in less than 0.2 s) which results in an abrupt pressure drop in the vessel (e). The vacuum period (f) is followed by a final releasing in order to establish the atmospheric pressure (g), or by the stage (c) of a new DIC cycle for a n-DIC cycle treatment (Figure 6). The last means that n cycles contain n repetitions of the stages (c), (d), (e) and (f), with a total heating time which is the heating time of all these cycles (n x d).

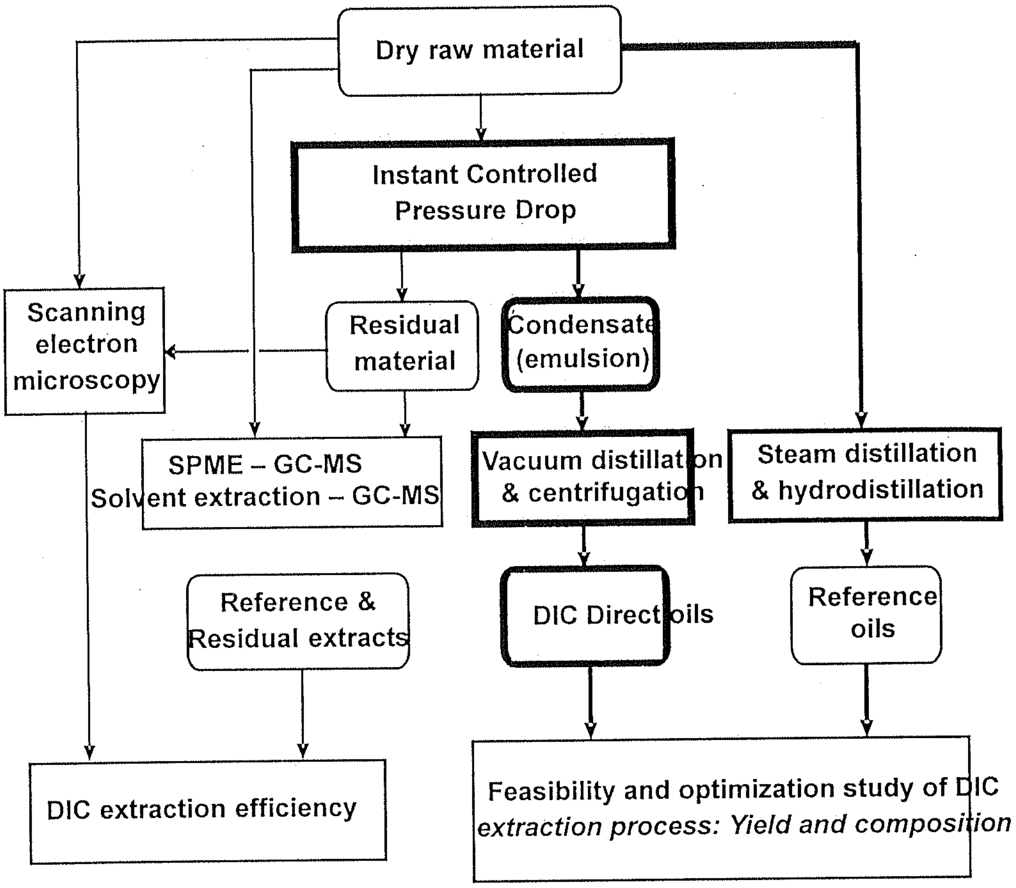


Figure 6. Experimental protocol

By abruptly dropping the pressure, an adiabatic (auto) vaporisation of the overheated water and volatile compounds takes place inducing an instant cooling of the residual material (Figure 4). The vapour engenders mechanical stresses within the plant puffing cells and structure depending on the plant viscoelastic behaviour, and even breaking cell walls; the values of initial water content and plant temperature just before dropping the pressure are very important parameters. The condensates we recovered were usually very stable oil-in water emulsions with droplet diameter less than 0.5 μm. On the other hand, the residual solid plants recuperated after treatment were subjected to structural and chromatography analysis as described in Figure 5.

Experimental design

Response surface methodology was used for the investigation of the effect of various DIC operative parameters on the DIC extraction efficiency of essential oils and their own compounds [51]. The investigation was done in two stages. Firstly, the range of DIC parameters which gave a reasonable DIC extraction efficiency of essential oil was identified for each aromatic plant. These value ranges of DIC operative parameters were defined from our previewed know-how, from some preliminary experiments and measurements, and from the identification of the structure of raw material, as well as of the type of secretor elements. In the case of Lavender, the operative parameters chosen as independent variables were the steam pressure ranged from 0.14 to 0.56 MPa and the thermal treatment time (t_d) ranging from 1 to 8 min (Table 2) were varied to build a 2-independent parameter five-level central composite design (CCD) [52] as a relevant RSM statistic method. The number of cycles was freely varied in order to get more precise answer concerning the total yields and the one of each compound: In the cases of the other dried aromatic plants, the level of DIC steam pressure was fixed for each raw material as it is shown in Table 3:

Table 2. Experiment design of DIC treatment of lavender with $W_t = 20$ % moisture dry basis

RSM operative variable parameters		-a	-1	0	+1	+a
$X_1 = P$	Steam Pressure (MPa)	0.14	0.2	0.35	0.5	0.56
$X_2 = t_d$	Thermal treatment time t_d (s)	60	122	270	418	480

a (axial distance) = $\sqrt[4]{2^N}$, N is the number of independent variables
In the present case: N = 2 and a = 1.4142.

Table 3. Level of steam pressure of DIC treatment

No.	Raw material	Pressure (MPa)
1	Angelica seeds	0.6
2	Cinnamon bark (10 mm x 5 mm x 1 mm)	0.5
3	Chamomile flowers	0.4
4	Lemon grass leaves	0.5
5	Patchouli leaves	0.5
6	Thyme leaves	0.5
7	Valerian roots	0.6

In these last cases, the number of cycles (1 - 9) and the heating time (80 - 240 s) were varied to build a 2-independent parameter five-level central composite design (CCD) [52] as a relevant RSM statistic method. A second order polynomial model was used to express the responses as a function of independent variables:

$$\eta = \beta_o + \sum_{i=1}^{2} \beta_i X_i + \sum_{i=1}^{2} \beta_{ii} X_i^2 + \beta_{12} X_1 X_2 \qquad (26)$$

where η is the response, β_0, β_1, β_2, β_{11}, β_{22} and β_{12} are the regression coefficients and X_1 and X_2 are the independent variables with actual values in original units.

Based on predicted model equations, surface plots were generated by Statgraphics Plus 5.1 software. The contour plots for dependent variables were superimposed to find out the optimum combinations of independent variables. Analyses of variance (ANOVA) were performed to determine significant differences between independent variables (P d" 0.05). For two variables, the central composite design yielded 12 experiments with four factorials points, four extra points (star points) and four central points for replications. The experiments were run in random in order to minimize the effects of unexpected variability in the observed responses due to extraneous factors. The coded and uncoded independent variables are shown in Tables 2 and 4.

Table 4. Independent variables used in response surface methodology at a fixed steam pressure value in the cases of dried aromatic plants

	Coded level	$-\alpha$	-1	0	1	$+\alpha$
$X_1 = C$	Number of cycles	1	2	5	8	9
PE 1 $X_2 = t_d$	Heating time (s)	80	104	160	216	240

α (axial distance) = $\sqrt[4]{2^N}$

N is the number of independent variables

In the present case: N = 2 and α = 1.4142

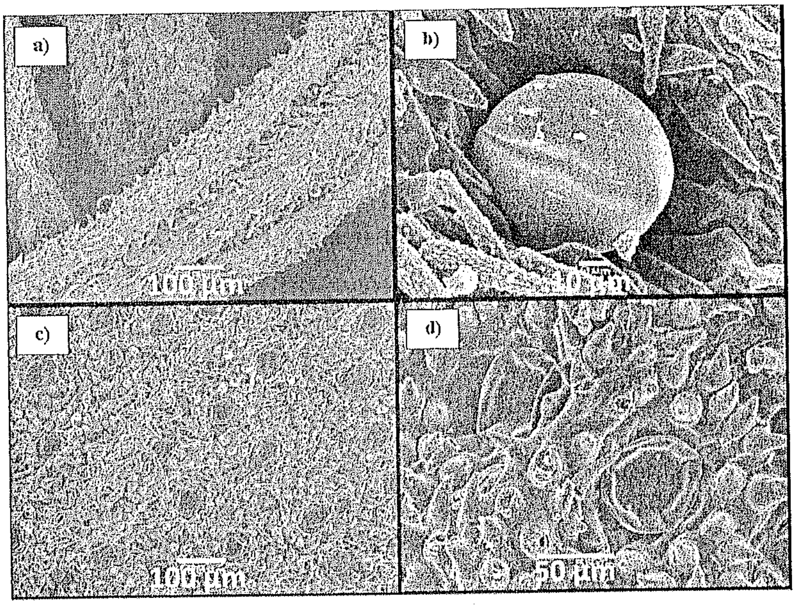


Figure 7. Scanning electron micrographs of peltate glandular trichomes on the adaxial surface of thyme leaf: intact glands before DIC treatment (x 150) (a), single intact gland (x 1000) (b), deflated glands after DIC treatment at 0.5 MPa, 9 cycles, 160 s (c) and deflated glands (x 500) (d)

Steam distillation

Steam distillation was carried out as a reference for comparing the essential oil DIC extraction. A Pignat P3734 laboratory apparatus was used. A packed bed of 50 g of dry aromatic plants was placed on a stainless steel grill fixed above a distillation vessel containing 2 L of water. Electric heating of the water produced 2.5 kg/h steam which in turn passed through the plant bed where it evaporated and carried away the essential oil. The vapours were condensed and then collected in a receiver vessel. At the beginning the receiver was filled with 1 L of water. A cooling tap water system around the receiver vessel cooled the distillate from 100° - 25°C in order to avoid the loss of volatile compounds. During the isolation process, the excess of water in the receiver vessel was recycled into the distillation vessel in order to restore the evaporated water. When the distillation time (processing time) had elapsed, the condensate was subjected to liquid-liquid extraction step using chloroform as solvent as described in previous paragraph in order to isolate the essential oil. After the preliminary kinetics study, 7 h was found as the optimum distillation time for each plant.

Chromatography analysis

HS-SPME-GC-MS

In this study, a 50/30 ìm PDMS/DVB/CAR fiber was used because it is know that its medium polarity reduces discrimination toward polar compounds, thus allowing the characterization of compounds involved on the overall aroma within a wide range of polarity [53,54]. The fibre was conditioned prior to use according to supplier's prescriptions. Before sampling each fibre was reconditioned for 5 min in the GC injector port at 250°C to eliminate the possible remains on the coated fibre. HS-SPME parameters including sample mass, extraction temperature, equilibrium time (10 min at a fixed extraction temperature), extraction time and, desorption time were optimized. The final conditions were given in Table 5. A fixed quantity of sample (see Table 5) was placed in a 10 ml vial and the vial was tightly capped with a PTFE-face silicone septum (Supelco, USA) and located into the sample tray of the CombiPaL (Varian, France).

SPME desorption

The SPME fibres were fixed in the CombiPal (CTC Analytics AG, Zwingen, Switzerland) autosampler for analysis. After the equilibration time, the fibre was exposed to the headspace of which the extraction parameters can be seen in Table 5. Once sampling was finished, the fibre was withdrawn into the needle and inserted to the GC injection port for desorption time of 10 min at 250°C. Once the analysis was finished, the fibre was automatically cleaned by heating according to supplier's instructions.

Gas chromatography - mass spectrometry (GC-MS equipment)

The volatiles molecules dissolved in chloroform extracts and the DIC, hydrodistilled and steam distilled oils were analyzed by gas chromatography coupled to mass spectrometry (GC/MS). A Varian computerized system comprising a 3900 gas chromatograph equipped with a fused-silica-capillary column factor four (30 m × 0.25 mm i.d.; 0.25 µm film thickness VF-5MS; Varian, France) was connected to a 2100T mass spectrometer. The column temperature program can be found in Table 4. The other chromatographic conditions were as follows: injector temperature at 250°C and He carrier gas flow at 1.0 ml/min. The measurements were performed under the following conditions: carrier gas He; flow rate 1 mL/min; split 1:100; injection volume 0.1 µL; injection temperature 250°C; the column temperature program for the extracts/essential oils of each aromatic plant can be found in Table 5. The ionization mode was an electronic impact at 70 eV. Mass spectra and reconstructed chromatograms were obtained by automatic scanning in the mass range m/z 30-400 a.m.u at 2.2 scan/s. Identification of the components was achieved by a comparison of their retention indices relative to C10-C40 n-alkanes (RI) and mass spectral fragmentation patterns with those found in the literature [25,55-58], and stored in the data bank (Varian NIST MS Database 1998 and Saturn libraries).

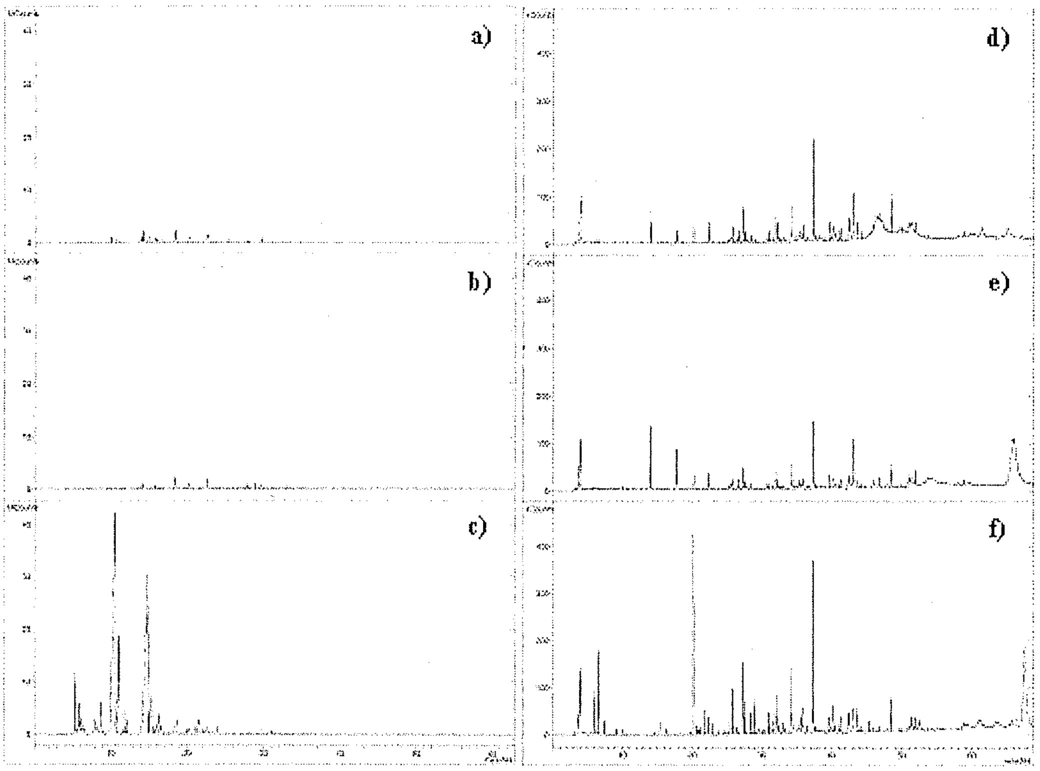


Figure 8. Gas chromatogram: - SPME extracts of DIC treated chamomile flowers at a) 0,4 MPa, 9 cycles, 160 s and b) 0,4 MPa, 9 cycles, 6 min and c) untreated flowers. - Chloroform extracts of DIC treated valerian roots at d) 0,6 MPa, 9 cycles, 160 s and e) 0,6 MPa, 9 cycles, 6 min and f) untreated roots.

A quantitative analysis was performed by the peak area normalization method without appliying response factor corrections. Because of non-availability of authentic standards, all relative response factors were taken as one. Individual calibration of each constituent with its authentic compounds as external standards can be easily done with simple oils, but it is impossible with more complex oils containing many interesting constituents. Thus, in the present work, the quantity of essential oil and their principal components was determined by the external standard method using only one authentic compound, i.e. methyl nonadecanoate. The standard curves were generated by analysis of known concentrations of methyl nonadecanoate dissolved in chloroform. Then the quantification was made with linear calibration curve of the external standard and dilution factor.

Table 5. GC-MS and SPME-GC-MS parameters

Extracts	GC-MS	SPME-GC-MS		
	Oven temperature	Oven temperature	Ext. temp.	Ext. time
Angelica seeds	60°C: hold for 2 min 60-250°C: 3°C/min 250°C: hold for 1 min	60°C: hold for 3 min 60-170°C: 2°C/min 170°C: hold for 3 min	60°C	50 min
Chamomile flowers	60°C: hold for 2 min 60-250°C: 2°C/min 250°C: hold for 10min	60°C: hold for 3 min 60-230°C: 1°C/min 230°C: hold for 3 min	50°C	60 min
Cinnamon bark	60°C: hold for 2 min 60-250°C: 4°C/min 250°C: hold for 1 min	60°C: hold for 3 min 60-160°C: 2°C/min 160°C: hold for 3 min	70°C	40 min
Lemon grass leaves	60°C: hold for 2 min 60-250°C: 3°C/min 250°C: hold for 1 min	60°C: hold for 3 min 60-230°C: 3°C/min 230°C: hold for 3 min	60°C	50 min
Patchouli leaves	60°C: hold for 2 min 60-250°C: 2°C/min 250°C: hold for 1 min	xxx	50°C	xxx
Thyme leaves	60°C: hold for 2 min 60-250°C: 2°C/min 250°C: hold for 1 min	60°C: hold for 3 min 60-160°C: 1,5°C/min 160°C: hold for 3 min	30°C	50 min
Valerian root	60°C: hold for 2 min 60-250°C: 3°C/min 250°C: hold for 1 min	60°C: hold for 3 min 60-230°C: 3°C/min 230°C: hold for 3 min	50°C	40 min

The last stage was the quantification of volatiles in DIC condensate at the optimum parameters resulted from the above described RSM analysis. The sum of the volatiles in DIC condensate and DIC treated plants (defined as total availability) was compared with the amount in the non-treated plants. The plant structure before and after DIC treatment was investigated by scanning electron microscopy (Jeol 5410 LV SEM).

In the first stage of work concerning the estimation of DIC extraction efficiency, the headspace-solid phase micro extraction-gas chromatography-mass spectrometry (HS-SPME-GS-MS) method was selected for quantifying rapidly the difference in volatiles availability in the non-treated and DIC treated plants.

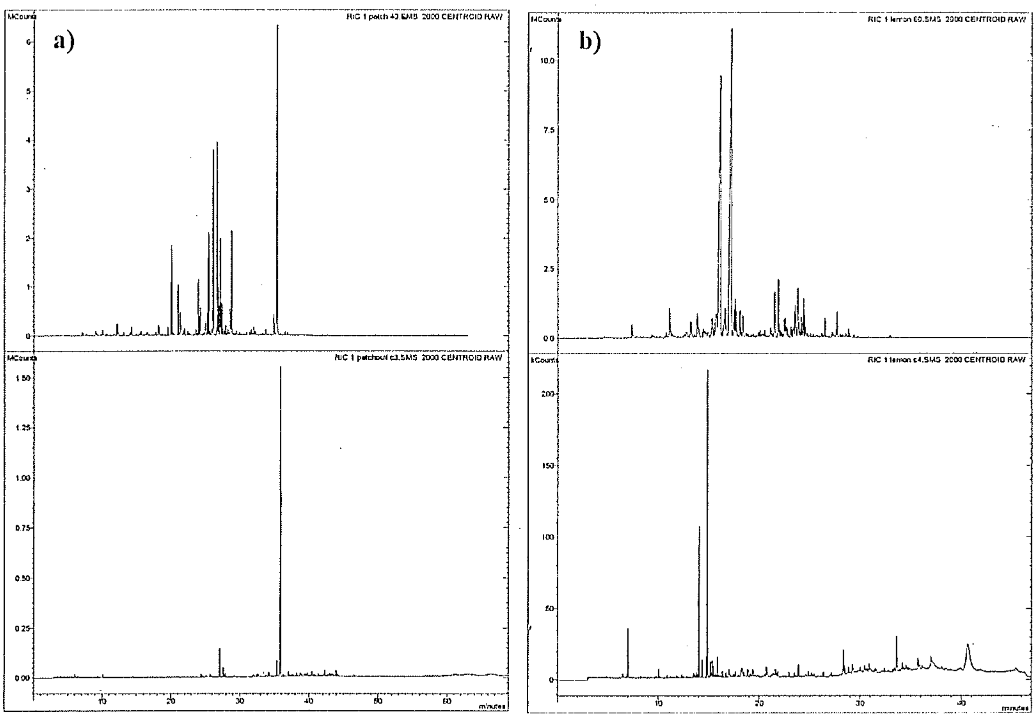


Figure 9. Comparison of aromatics profile between HS-SPME (above) and chloroform extraction (below). a) Patchouli extracts; b) lemongrass extracts

HS-SPME integrates sampling, extraction, concentration and sample introduction into one step and single device without the need for any organic solvent [50,60]. This technique which typically followed by gas chromatography (GC) analysis was successfully applied for analysis of volatile compounds in environmental and food samples [61-65]. This technique is simple, rapid, reproductive and efficient, involving immersing a phase-coated fused silica fibre into the liquid sample or the headspace above the liquid or solid sample, to adsorb/absorb the analytes of interest. The adsorbed/absorbed analytes are then thermally desorbed in the injection port of the gas chromatograph and transferred to the analytical capillary column. Due to its higher confirmatory potential mass spectrometry (MS) detection is commonly applied, working in full scan or SIM modes typically using ion trap or quadrupole detectors. The quantity of a given compound adsorbed onto the fibre depends on both the partitioning of the compound into the headspace from the plant matrix and the partitioning of the compound into the fibre coating from the headspace. SPME method also avoids the chemical modification and artefact formation that can occur in conventional methods.

The headspace mode of SPME appeared to be particularly advantageous for "volatile" and "semivolatile" substances because of the very low chromatographic background. In order to study the DIC effect on the improvement of total availability of essential oil, it is necessary to isolate and quantify totally the volatiles in the non-treated and DIC treated plants. It is impossible to realize this measurement with HS-SPME-GC-MS method because the latter is an equilibrium extraction technique (non-quantitative extraction technique) and not an exhaustive one [66]. Then, the solvent organic extraction using Randall method was applied for this purpose. The risk of thermal degradation can be reduced using solvent extraction rather than steam distillation.

External standard solutions at 0.2, 0.6, 1.2, 2, 3, 5 mg/mL of methyl nonadecanoate (puriss. grade, Sigma-Aldrich Corp., Germany) in chloroform (GC grade, Carlo Erba Reagenti, France) were prepared for GC-MS analysis. NaCl (purum grade, Sigma-Aldrich Corp.), anhydrous Na_2SO_4 (for analysis, purity > 99.5 %, Fischer Chemicals AG), and chloroform (GC grade) were used as demulsifier, drying agent and solvent, respectively, in the liquid-liquid extraction step of DIC, hydro and steam distillation condensates; the latter was also used in the solvent extraction of untreated and residual flowers. The n-alkane standard mixture (C10-C40, 50 mg/L in n-heptane each) was purchased from Sigma-Aldrich Corp. (Germany). The SPME holder and the fibre of 50/30 μm PDMS/DVB/CAR were purchased from Supelco (USA).

Yield and extraction efficiency

The main response of the RSM analysis was chosen as the essential oil availability in the DIC treated plant. This residual availability denotes the ratio of oil yields in the DIC treated (residual oil) and non-treated plants (reference oil). By assuming that the sum of the volatiles in residual oil and DIC condensate was equal to the quantity in reference oil, the amount of oil which was removed by DIC treatment could be estimated. The yield of reference oil (y_{of}) and DIC direct and residual oils (y_{od} and y_{os}) is based on dry matter of plants:

$$y_{of} = \frac{m_{of}}{m_{dm}} \tag{27}$$

$$y_{od} = \frac{m_{od}}{m_{dm}} \tag{28}$$

$$y_{os} = \frac{m_{os}}{m_{dm}} \tag{29}$$

where the oil mass was computed from GC peak area of all volatiles molecules using the external standard method with methyl nonadecanoate. DIC direct oil is composed of the volatiles molecules isolated from the DIC condensate. Reference oil contains the volatile molecules extracted from non-treated dry plants by chloroform during a fixed optimum time (Table 6). DIC residual oil means the volatile molecules isolated from DIC treated plants using the same solvent and time. By assuming that the DIC treatment only isolates a part of a compound and does not increase its availability (A) in residual solid plant, the DIC extraction efficiency could be estimated as:

$$E = 1 - A = 1 - \frac{y_{os}}{y_{of}} \qquad (30)$$

It should be noted that the availability in residual plants was often increased by the DIC treatment which led to underestimation of E.

Table 6. Optimum parameters for solvent extraction using chloroform

No.	Extracts	Sample mass / g (wet basis)	Extraction time (h)
1	Angelica seeds	4	2
2	Cinnamon bark	7.5	3
3	Chamomile flowers	4.5	4
4	Lemon grass leaves	6	4
5	Patchouli leaves	3.5	3
6	Thyme leaves	6	2
7	Valerian roots	9	4

Results

Case of lavender, oregano, and rosemary

In the case of lavender, oregano, and rosemary, the total yields of total extracted essential oils were measured in DIC extraction in comparison with conventional steam distillation and hydro-distillation processes. Systematically, optimised DIC treatment allowed the extracted essential oils to get much higher yield compared to hydro-distillation or steam distillation (Table 7).

Table 7. Yield of essential oils obtained by DIC extraction in comparison with conventional Steam distillation and Hydro-distillation processes

Essential oil	Steam distillation (% db)	Hydro distillation (% db)	DIC-Extraction (% db)
lavender	2.3 %	(3 hours): 4.3 %	DIC (0.6.MPa, 2 cycles, 4 min) 4.37 %
oregano		(6 hours): 0.67 %	DIC (0.6.MPa, 10 cycles, 5 min): 1.97 %
rosemary		(6 hours): 0.13 %	DIC (0.6.MPa, 10 cycles, 5 min): 0.68 %

In this case of lavender, 22 main compounds were firstly identified and then quantified in the plant (after or without DIC treatment) and in the extracts (by solvent, steam distillation or directly by different DIC treatments). Some of their most important thermodynamic characteristics were not available in the bibliography (partial pressure values at various temperatures). (Table 8).

Table 8. Identification of 22 lavender compounds

No.	Name of compound [68]	Ebullition temperature [69]
1	limonene ($C_{10}H_{16}$)	178°C at 0.1 MPa
2	eucalyptol ($C_{10}H_{18}O$)	176,4°C at 0.1 MPa
3	linalool ($C_{10}H_{18}O$)	198°C at 0.1 MPa
4	camphor ($C_{10}H_{16}O$)	204°C at 0.1 MPa
5	lavendulol ($C_{10}H_{18}O$)	
6	borneol ($C_{10}H_{18}O$)	208°C at 0.1 MPa
7	1-terpinen-4-ol ($C_{10}H_{18}O$)	212°C at 0.1 MPa
8	n-hexyl butyrate ($C_{10}H_{20}O_2$)	
9	α-terpineol ($C_{10}H_{18}O$)	217°C at 0.1 MPa
10	hexyl isovalerate ($C_{11}H_{22}O_2$)	
11	linalool acetate ($C_{12}H_{20}O_2$)	220°C at 0.1 MPa
12	lavandulyl acetate ($C_{12}H_{20}O_2$)	196, 29°C at 0.1 MPa
13	ethyl linalool ($C_{12}H_{22}O$)	
14	acetic acid ($C_{13}H_{22}O_2$)	
15	caryophyllene ($C_{15}H_{24}$)	
16	(Z)-α-farnesene ($C_{15}H_{24}$)	
17	α-humulene ($C_{15}H_{24}$)	123°C at 0.1 MPa
18	geranyl propionate ($C_{13}H_{22}O_2$)	
19	t-cadinene ($C_{15}H_{24}$)	
20	caryophyllene oxide ($C_{15}H_{24}O$)	
21	t-cadinol ($C_{15}H_{26}O$)	
22	α-bisabolol ($C_{15}H_{26}O$)	

Whatever the compound, the higher the thermal treatment time and the steam pressure, the higher the yields. However, a steam pressure of 0.6 MPa has been used for studying the effect of the number of cycles and the total thermal treatment time for each compound. The results are the following (Figure 10).

It was clear that some compounds (Limonene ($C_{10}H_{16}$), Eucalyptol ($C_{10}H_{18}O$), Ethyl linalool ($C_{12}H_{22}O$), and α-humulene ($C_{15}H_{24}$)) were completely extracted after the first cycle. After four to six cycles, the main part of compounds [1*] were extracted. Only some ones (α-Terpineol ($C_{10}H_{18}O$), t-Cadinol ($C_{15}H_{26}O$), and α-Bisabolol ($C_{15}H_{26}O$)) seemed to need much more pressure drops. This observation would be very important in a possible strategy implying the possibility of a fractional extraction by DIC; such an operation must then be defined in each case depending of raw material and the needed compounds.

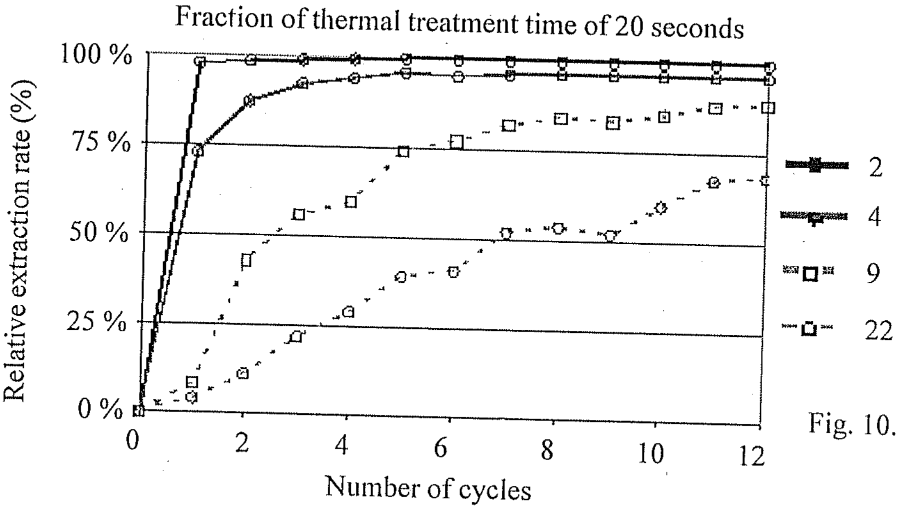

Fraction of thermal treatment time of 20 seconds

Fig. 10.a

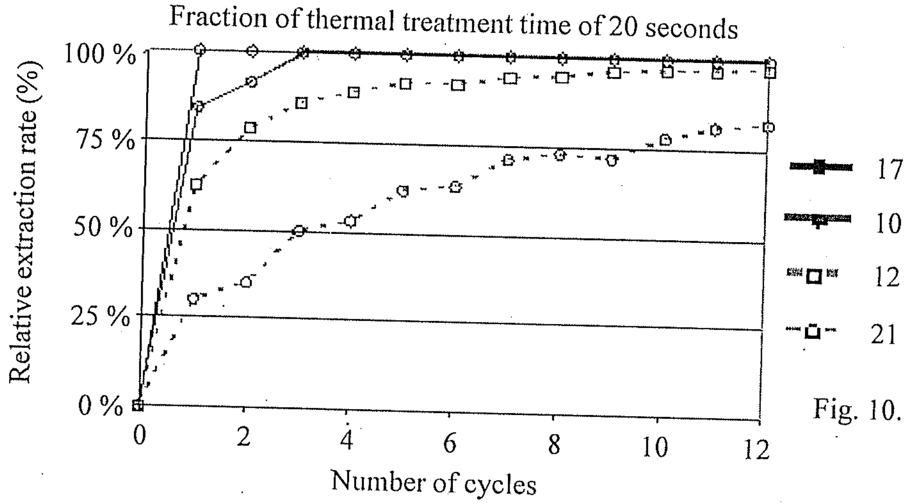

Fraction of thermal treatment time of 20 seconds

Fig. 10.b

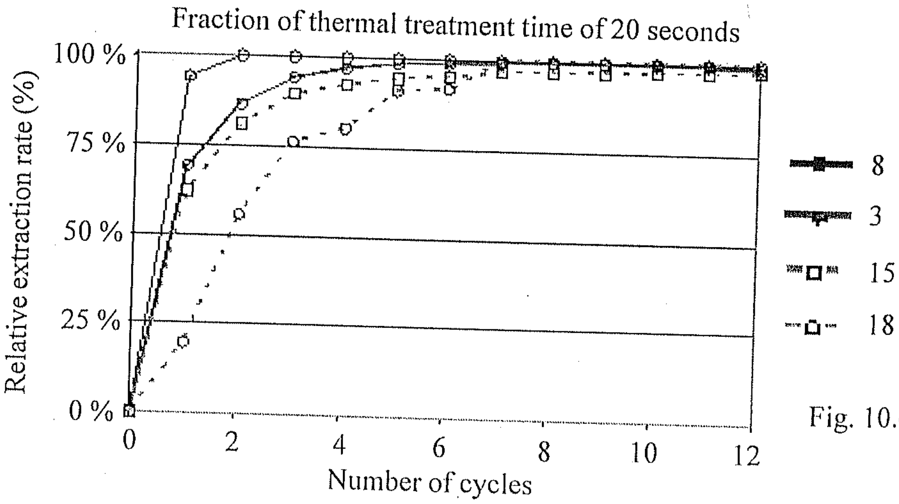

Fraction of thermal treatment time of 20 seconds

Fig. 10.c

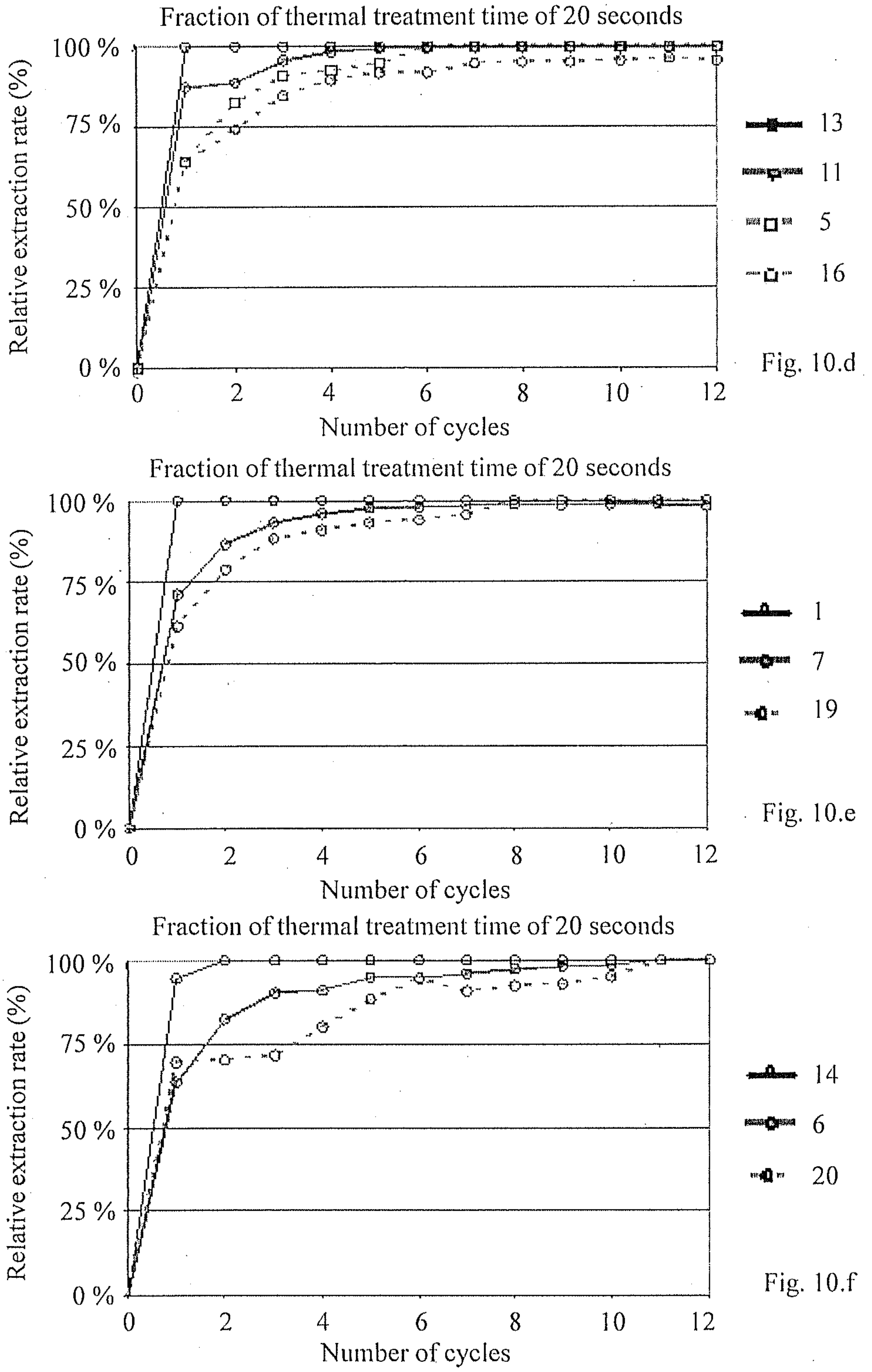


Figure 10: The relative extraction rate of various compounds of lavender essential oils

1* (Linalool ($C_{10}H_{18}O$)
Lavendulol ($C_{10}H_{18}O$)
1-Terpinen-4-ol ($C_{10}H_{18}O$)
Hexyl isovalerate ($C_{11}H_{22}O_2$)
Lavandulyl acetate ($C_{12}H_{20}O_2$)
Caryophyllene ($C_{15}H_{24}$)
Geranyl propionate ($C_{13}H_{22}O_2$)
Caryophyllene oxide ($C_{15}H_{24}O$)
Camphor ($C_{10}H_{16}O$)
Borneol ($C_{10}H_{18}O$)
n-Hexyl butyrate ($C_{10}H_{20}O_2$)
Linalool acetate ($C_{12}H_{20}O_2$)
Acetic acid ($C_{13}H_{22}O_2$)
(Z)-α-farnesene ($C_{15}H_{24}$)
t-cadinene ($C_{15}H_{24}$)

Case of other aromatic plants (Angelica seeds, Cinnamon bark, Chamomile flowers, Lemon grass leaves, Patchouli leaves, Thyme leaves, Valerian roots)
In the case of various aromatic plants, the yield values of total DIC extracted essential oils were systematically, much higher than the ones of hydro-distillation and steam distillation (Table 8).

Table 9. Yield of essential oils obtained by DIC extraction in comparison with conventional Steam distillation and Hydro-distillation processes

Essential oil	Steam distillation (% db)	Hydro-distillation (% db)	DIC-Extraction (% db)
Angelica seeds	2.10 %	1.91 %	-
Cinnamon bark	1.37 %	2.62 %	-
Chamomile flowers	2.53 %	1.89 %	-
Lemon grass leaves	1.51 %	1.00 %	-
Patchouli leaves	2.63 %	2.39 %	-
Thyme leaves	2.71 %	3.68 %	4.34 %X
Valerian roots	1.05 %	0.95 %	-

The experiment design (table 4) had, as variable operative parameters the total thermal treatment time and the number of cycles; the response parameter was the efficiency defined by Eq 30. The empirical models were given in the cases of chamomile, lemon grass, and valerian, in the Table 9:

Table 10. Regression coefficients and optimal values for DIC extraction efficiency

	Chamomile oil	Lemon grass oil	Valerian oil
β_0	1.12764	-2.60213	-0.909677
β_1	-0.00408177	0,.68308	0.140097
β_2	-0.00355253	0.52666	0.335746
β_{11}	-0.000936851	-0.0202954	-0.0104333
β_{21}	0.000178523	-0.023387	0.00501368
β_{22}	7.79614E-06	-0.0315889	-0.0282619
R^2 (%)	77.1	95.6	91.7

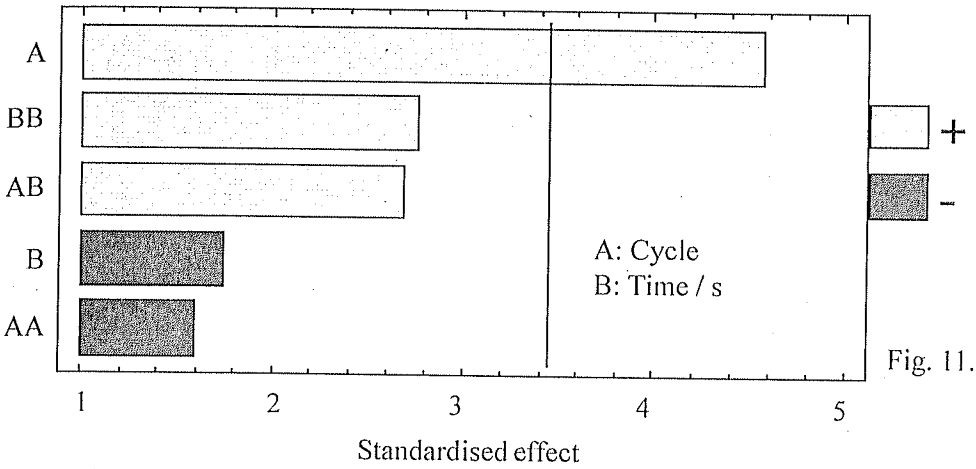


Fig. 11.a

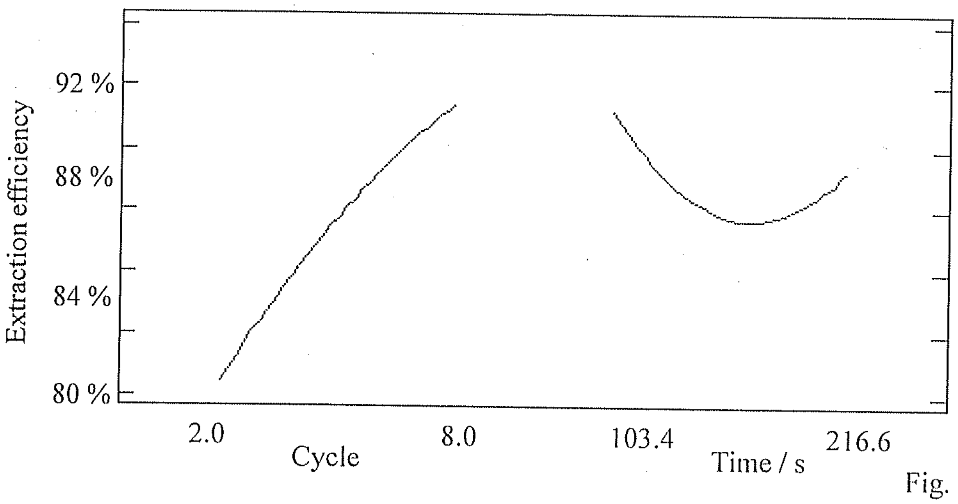


Fig. 11.b

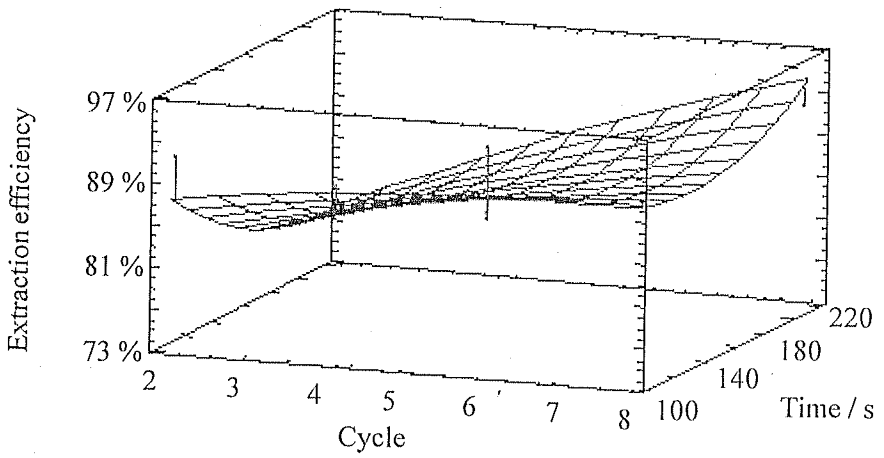


Fig. 11.c

Figure 11. Effect of cycle number and heating time on DIC extraction efficiency of chamomile oil/ a) Pareto charts; b) main effects; c) response surfaces.

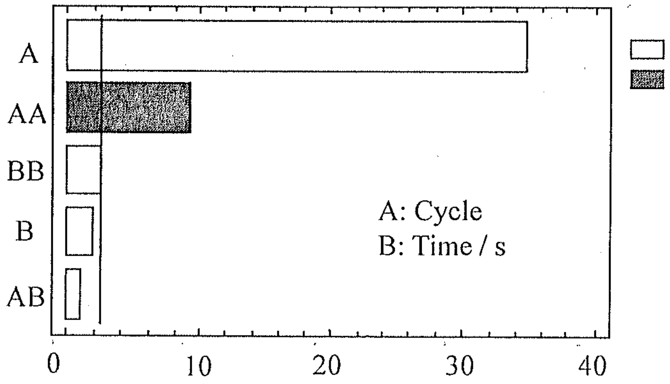
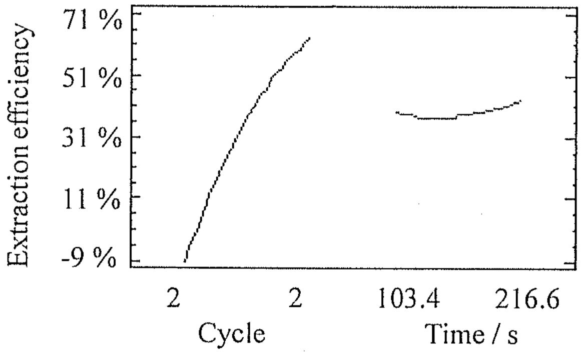


Fig. 12.a



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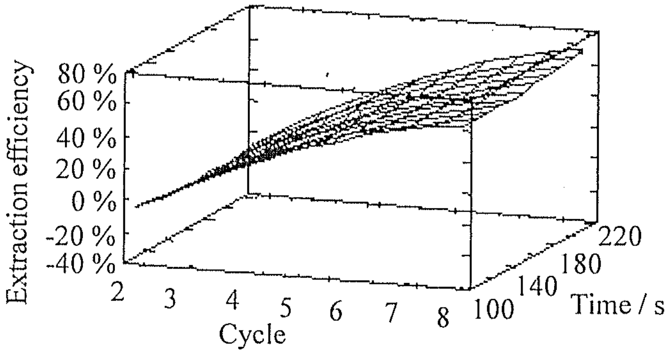

Fig. 12.c

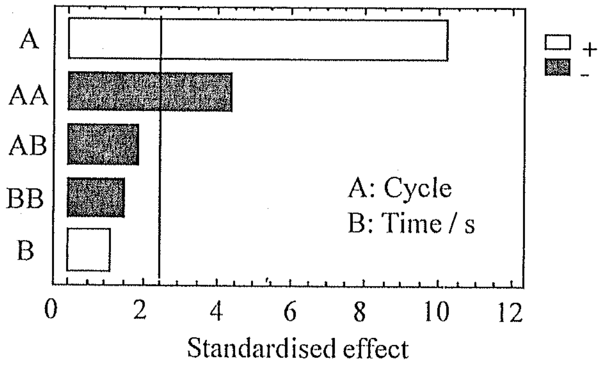

Fig. 12.d

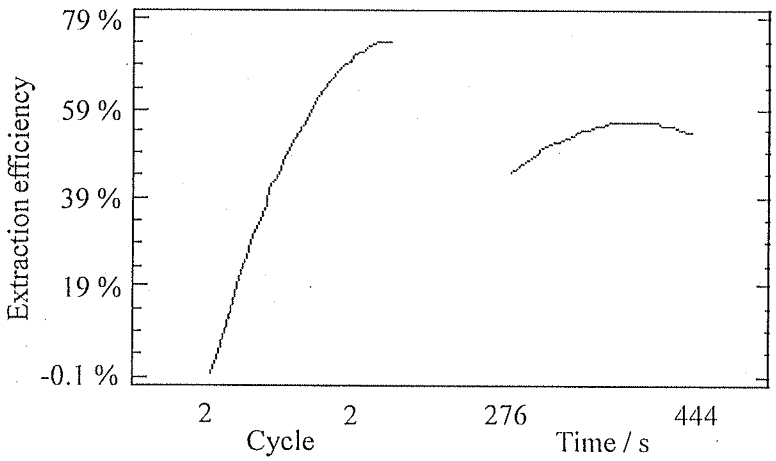

Fig. 12.e

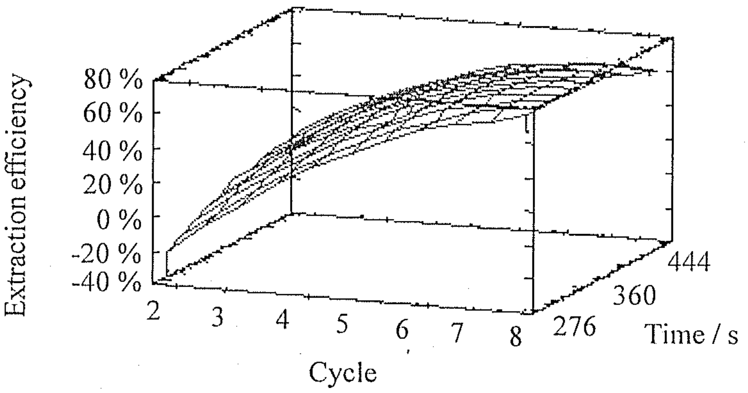

Fig. 12.f

Figure 12. Effect of cycle number and heating time on DIC extraction efficiency of lemon grass oil using shorter (left) and longer treatment time (right). a), d) Pareto charts; b), e) main effects; c), f) response surfaces.

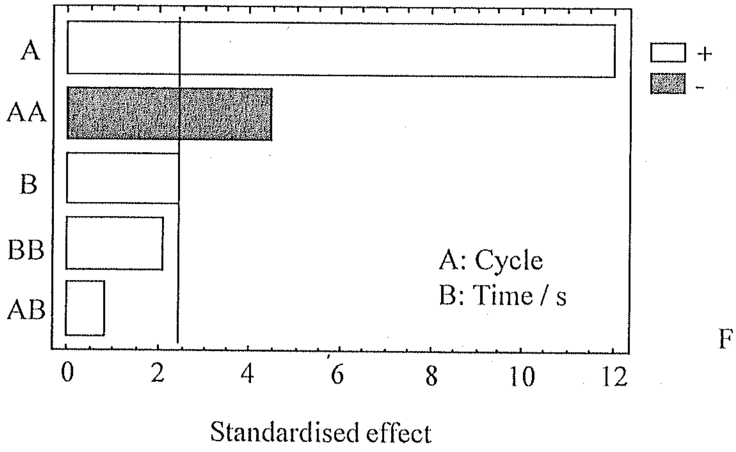

Fig. 13.a

12.a

12.b

12.c

. 12.d

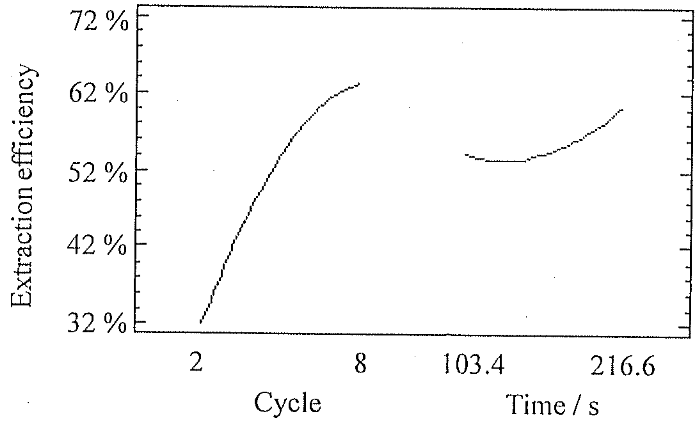

Fig. 13.b

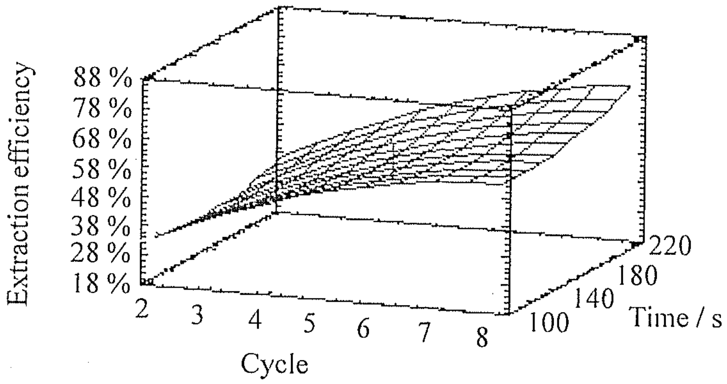

Fig. 13.c

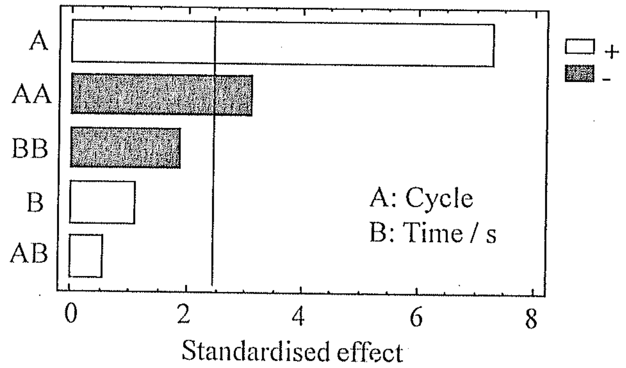

Fig. 13.d

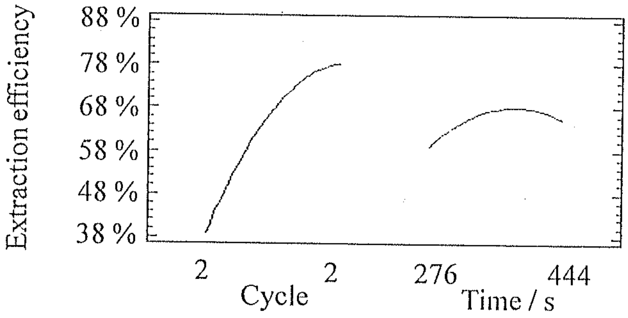

Fig. 13.e

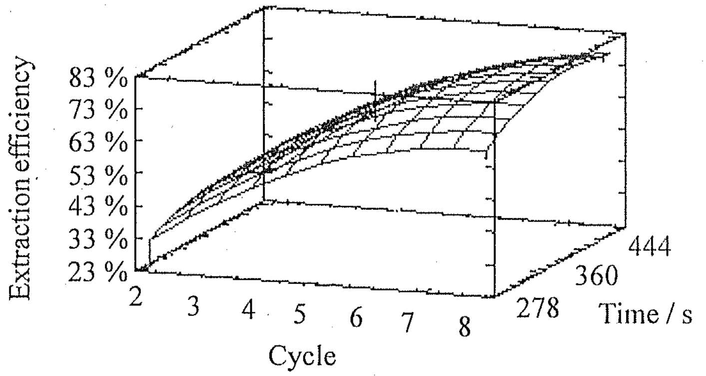

Fig. 13.f

Figure 13. Effect of cycle number and heating time on DIC extraction efficiency of valerian oil using shorter (left) and longer treatment time (right). a), d) Pareto charts; b), e) main effects; c), f) response surfaces

Conclusion and discussions

The DIC treatment is confirmed as a very relevant essential oil extraction technology. There were many preliminary works for various raw materials as follows: kinetics study and optimization of DIC as essential oil extraction in comparison with the conventional extraction processes as solvent extraction, hydro and steam distillation which are tedious and time consumption.

At 0.6 MPa as steam pressure, the DIC extraction of lavender is achieved in 1 cycle and 20 s for limonene, eucalyptol, α-humulene, ethyl linalool. It is 2 cycles and 40 s for n-hexyl butyrate, acetic acid; 4 cycles and 80 s for hexyl isovalerate, 6 cycles and 120 s for camphor, lavendulol, linalool, caryophyllene, linalool acetate, geranyl propionate, borneol, caryophyllene oxide, 1-terpinen-4-ol, and 10 cycles and 200 s for lavendulyl acetate. Some 70 % of α-bisabolol and 80 % of α-terpineol and t-cadinol are extracted by 12 cycles and 240 s. The essential oil extraction by DIC autovaporisation has the advantages of very relevant kinetics, short time, and fractioning process.

The effect of DIC cycle number on the peltate glands of thyme leaf is shown in Figure 5. Micrographs of the glands after the instantaneous pressure drop treatment (DIC treatment) showed evidence of disruption due to a bursting action from within the interior of the glands because of pressure gradient developing across the grand walls during pressure drop. These deflated glands facilitate the liberation and vaporization of volatiles molecules. The same tendency is noted with others aromatic plants, when the number of pressure drop (i.e. number of DIC cycle) increases, the alveolation structure becomes more prominent and results in a higher breaking rate of cell walls. Such complex micro-alveolation structure coupled to auto-vaporization with a total pressure gradient from heart to superficial zones allow essential oil compounds within endogenous site such as secretory ducts and cells, to be removed and liberated.

The gas chromatograms as well as HS-SPME-GC-MS analysis technique are feasible for quantitative comparative study of the residual and reference extracts that obtained from

DIC treated and non-treated plants, respectively. HS-SPME-GC-MS is very rapid, reproducible and no solvent required for replacing the usual chloroform extraction method. For all aromatic plants, we found that the DIC extraction efficiency increases with cycle number and heating time. For all plants, the most significant parameter is cycle number. However, contrary to peltate glands in thyme leaves and secretory ducts in chamomile flowers, the more complex secretory structure and physiology plant, such as secretory cells in lemongrass leaves and valerian roots, the longer heating time was required to obtain a satisfactory extraction efficiency.

For this case, we also observed a low value of extraction efficiency at first cycles and then it increases up to asymptotic value at the last cycles: the DIC treatment has a double effect. First, the vapour generated in the flowers during the expansion breaks and alveolates their structure and carries out the oil. Just after "external" pressure drops, the mass transfer is not controlled by the diffusion of volatile components, but by the total pressure gradient. These facts prove the hypothesis that the DIC essential oil extraction mechanism is mainly based on the auto-vaporization of complex liquid (water + volatile oil).

DIC extraction is a rapid process in comparison with hydro and steam-distillation. In the case of thyme oil, a direct oil yield of 4.3 % dm was isolated using DIC at 0.5 MPa and 9 cycles in 160 s as against 3.5 % dm in 7 h for hydro (HD) and 2.7 % in 6 h for steam distillation (SD).

The DIC direct oil contains more Light Oxygenated Compounds (borneol, eucalyptol, linalool, thymol ....) and Heavy Oxygenated Compounds (caryophyllene oxide, cubenol, eudesmol, ...) than HD and SD oils. A similar result, i.e. an increase of oxygenated compounds, was obtained by Lucchesi *et al.* [39] using solvent-free microwave extraction. On the other hand, the sesquiterpenes hydrorocarbons are present in a higher quantity in SD and HD oils. The DIC process yields more valuable thyme oil than SD. For other aromatic plants, several minutes of DIC process isolates the same or even higher essential oil yield than several hours of HD and SD. All DIC oils have the same tendency in aromatic profiles as thyme oil does. The oxygenated compounds (LOC and HOC) are highly odoriferous, while the sesquiterpenes hydrocarbons contribute only little to fragrance and therefore are less valuable. The quality of essential oils increases with the content of oxygenated compounds which is the case of DIC treatment.

The DIC isolation process is based on auto-vaporization of volatile compounds from modified structure resulted from multi-cycle pressure drop and not on molecular diffusion as the classical separation methods. Thermal degradation risk is avoided by using a short heating time (< 1 min) in each DIC cycle.

The DIC process is very efficient for essential oils extraction. In comparison with steam and hydro-distillation, the DIC process is more efficient in term of rapidity (several minutes versus several hours), essential oil yield (comparable even higher) and higher content of oxygenated compounds. The DIC treatment also increases the total availability of volatiles molecules in the aromatic plants.

The optimum parameters and macro-scale model of DIC extraction were established for each aromatic plant. The model was presented as DIC extraction efficiency in function of cycle number and heating time. The most significant parameter is cycle number, on the other hand, the heating time is not significant. The structure of the plants is more alveolated with number of DIC cycle.

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