THE APPLICATION OF EXTRACTIVE PROCESSES UNDER CRITICAL CONDITIONS IN NUTRITIVE SCIENCES

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Abstract

Nutritive Sciences represent a very inciting and challenging study sphere of a really interdisciplinary nature. They conciliate together chemical, fisical, biological, medical, agricultural, social and engineering disciplines, evaluating the influence of nutrition on health from the basic molecular and cellular levels to the higher structures of organs, organisms and even populations.
It is believed that there are at least 43 chemical components, called essential nutrients which must be present in our food. Furthermore, the present popular belief that everything "natural" is good, provides a positive incentive towards growth of the natural products industry, particularly in the food, flavoring, perfumery and pharmaceutical sectors.
One of the most important study directions of Nutritive Sciences is the application of advanced extraction methods in natural products. This paper aims to give an information about the main characteristics of these methods and their application in nutritive sciences, with the final purpose the effective gain of qualitative nutritive components.
One of such major technologies that has emerged over the last two decades as the alternative to the traditional solvent extraction of natural products is the supercritical fluids extraction technique. The most desirable SCF solvent for extraction of natural products for foods today is carbon dioxide (CO_2). It is an inert, inexpensive, easily available, odorless, tasteless, environment-friendly and GRAS (generally regarded as safe) solvent. In this paper there are also included the recent news, the proven effectivity of the technique through a variety of applications such as extraction and separation of oils and fats, the separation of tocopherols and other antioxidants, the extraction of classical components such as essential oils and their derivates. It is intended to bring this contemporary scientific reality even in our country in the future, offering thus a better opportunity to experimental research with positive technologic, economic and nutritional development.

Keywords: *nutritive sciences, extraction, critical conditions*

Introduction

Nutritive Sciences represent an inciting and challenging study sphere of a really interdisciplinary nature. Their object of study is the composition of food and its nutrition value.
It is actually believed that there are at least 43 chemical components called essential nutrients which must be present in our food. Consequently, it is preferable to obtain them from natural sources, in the form of natural extracts.
Based on this absolute priority, one of the most important study directions of Nutritive Sciences is the application of advanced and optimal extraction methods in natural products, in order to effectively gain qualitative nutritive components. Some of the recent extraction methods include Microwave Assisted Extraction (MAE), Supercritical Fluid Extraction (SCFE) and Pressurized Solvent Extraction (PSE).
These techniques offer high rendiments of the extracted materials; the extraction of a considerable number of components; high results in a very short time; the replacement of older methodics which negatively effect the environment, apparatus, material for extraction or that has to be recuperated by the products of the reaction.
One of the most advanced technologies that has emerged over the last two decades as the alternative to the traditional solvent extraction of natural products is the supercritical fluids extraction technique. This paper gives information about the main characteristics of the technique and its utilization in nutritive sciences, suggesting carbon dioxide (CO_2) as the most desirable SCF solvent for the extraction in natural products.

The extraction with gases under pressure

Some of the most known and classical extraction techniques in the course of the years include distillation, organic solvent extraction in liquid-solid, liquid-liquid systems, absorbation, sublimation, ionic exchange, filtration, different chromatographic applications and so on.
Each of these techniques has its advantages and disadvantages but final study objective remains their optimization or substitution in favor of the most advanced and modern methodics. The latter include gases under pressure followed by numerous scientific applications. The research work is related to the natural products extraction in supercritical conditions, which means temperature and pressure values higher than the critical ones. The extraction with gases under pressure has the priority of extracting natural products. It has various applications in food, cosmetic and pharmaceutical industry.

Definition of Supercritical Fluid

A supercritical fluid (SF) is a state where the matter is compressible and behaves as a gas (it fills and takes the shape of its container). A supercritcal fluid has the typical density of a liquid (between 0,1 – 1 g / ml) and a characteristic dissolving power. SF can also be defined as a heavy

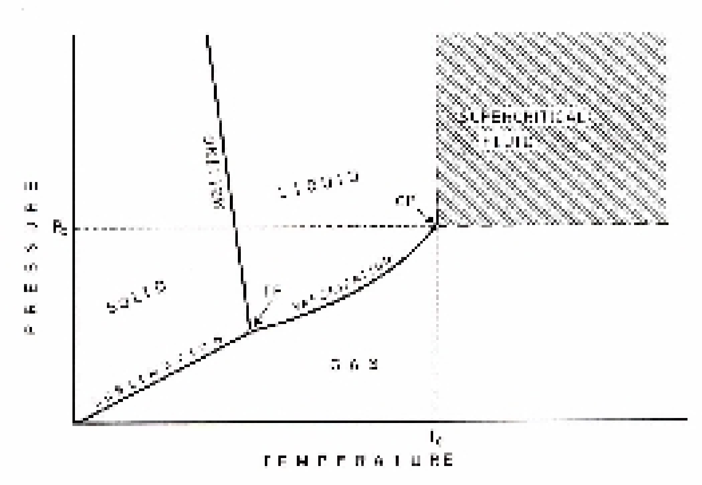
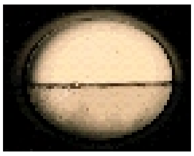


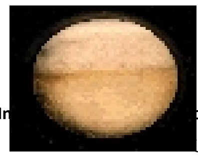
Fig 1. Supercritical Fluid Phase Diagram

gas with a controllable dissolving power or as a form of matter in which the liquid and gaseous state are indistinguishable. A typical phase diagram **(Fig.1)** shows the temperature and pressure regions where the substance occurs as a single phase [viz. Solid (s), liquid (l) or gaseous (g)]. Such regions are bounded by curves indicating the coexistence of two phases (s - g, s - l and l – g, which are involved in sublimation, melting and vaporization equilibria, respectively). The three curves intersect at the co-called *triple point* (TP), where the solid, liquid, and gaseous phases coexist in equilibrium.

The most important compounds used as supercritical fluids are CO_2, ammonia, water, nitrous oxide, ethane, ethane, xenon…etc

In the imagine below **(Fig.2)** the two distinguished phases of the CO_2 can be seen, separated by the meniscus. With the increase of temperature, the difference between the two phases is less evident. The meniscus is still seen, but it is less defined. When the critical temperature and pressure values are reached, the distinction between the





two phases disappears and the separation meniscus is no more seen. The homogenous phase obtained is defined as “Supercritical Fluid” and manifests intermediate properties between those of liquids and gases.

Fig.2 Visual representation of the different CO_2 phase states, in dependence to pressure

The Supercritical Fluid Extractor - Basic Scheme

All designs of SFE apparatus, regardless of complexity and cost, share the same basic components:

a) A source of high – purity supercritical fluid.
b) A high pressure pump delivering the fluid at constant, readily controllable pressure and flow – rate.
c) A cell or chamber holding the sample, thermostated with the aid of an appropriate device.
d) A restrictor (also known as a Back Pressure Regulator- **BPR**) intended to maintain the required pressure in the extraction chamber.
e) A system for collection of extracted components after the SF is depressurised, connected with the extraction chamber via the restrictor. Extracts are usually collected by bubbling the depressurized fluid through a container holding a few millilitres of appropriated solvent.

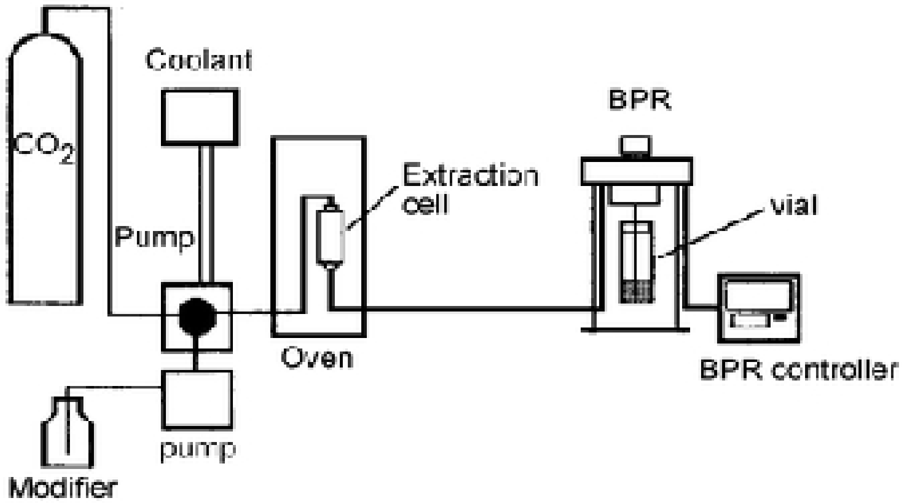


Fig. 3: Basic scheme of a supercritical fluid extractor

The solvent delivery system consists of a pump to deliver liquid carbon dioxide and, optionally, a pump to supply modifier. The oven is used to keep the cell contents above the critical temperature of the extraction fluid. An equilibration coil is also included to help mixing of carbon dioxide and modifier and aid thermal equilibration of the extraction fluid and the insides of the oven. The cell, usually a hollow stainless steel cylinder, is housed in the oven and contains the sample to be extracted. It has a frit at both ends to prevent insoluble material leaving the cell, but allowing soluble substances to pass through unhindered. The BPR serves to keep the pressure in the system above the critical pressure of the extraction fluid. It is, typically, a length of fused silica capillary (50 µm i.d.) or a mechanical or electronic needle valve. The BPR is heated (with a hairdryer or in an oven) to reduce the frequency of blockages by, for example, the formation of ice. Finally, a collection system is required to trap extracted material. It is usually a solid trap or a small glass collector containing a few cubic centimeters of organic solvent.
In comparison with the classical techniques and distillation processes, the extraction with gases under pressure has some distinctive characteristics:

1. Work temperature usually approximates the solvent critical temperature, for this reason, to obtain thermally unstable substances, mild working conditions can be relevant.
2. The selection of gas under pressure can change as parameters vary (pressure and temperature) because its solvent capacity is a function of density.
3. As the pressure in the gas stage diminishes, an almost complete separation of substances, solvent recovery from solutions and extraction of remains can be possible.
4. By changing gas solvent capacity (as the parameters of the extraction change), a fractioned extraction can be attained.
5. Gases under pressure present an appropriate mass transport which is expressed through a fast penetration of the extracting material and good mass exchange. Supercritical fluids have a low dynamic viscosity which can be compared to that of the gas stage, and a high diffusion coefficient, 100 times higher than that in liquids.

6. During extraction under high pressure, oxygen penetration is low, consequently, the oxidizing damage it can cause to vulnerable materials is negligible.

   The most desirable derivative for the extraction of natural products, specifically beneficial for food, cosmetic, and pharmaceutical industries is CO_2. It results effective because of the following reasons:

   - It is the sole, cheapest solvent after water, and quantitatively effective
   - Because of its very good soluble properties, it leaves no remains in the final product, as such, it is ecologic
   - It does no physiologic harm
   - It is non-combustive, bacteriostatic, and nonpolar
   - CO2 realizes the extraction and separation of active components simultaneously.
   - It has a dispersal rate 10 times higher than that of other solvents, and carries out very fast extraction with a higher productivity than the extraction through other methods.

SCFE Applications in Natural Products and Food Industries

- Decaffeination of coffee and tea
- Spice extraction (oil and oleoresin)
- Deodorization of oils and fats
- Extraction of vegetable oils from flaked seeds and grains
- Flavors, fragrances, aromas, and perfumes
- Hops extraction for bitter
- Extraction of herbal medicines
- Stabilization of fruit juices
- Lanolin from wool
- Deoiling of fast foods
- Decholesterolization of egg yolk and animal tissues
- Antioxidants from plant materials.
- Food colors from botanicals
- Natural pesticides
- Denicotinization of tobacco
- Plant and Animal Lipids

Plant and Animal Lipids

Lipids are a large group of fatty organic compounds present in living organisms. Lipids form an important food and energy source in plant and animal cells. Plant lipids comprise a complex mixture of monoglycerides (MG), diglycerides (DG), triglycerides (TG), and free fatty acids (FFA) associated with some minor constituents, such as tocopherols,

sterols, phosphatides (gums), alkaloids, flavonoids, waxy materials, color compounds and volatiles that provide the taste and odor of the oils.
Their extraction is very important for many applications, as in food industry, pharmaceutical, plastic, lubrificant, cosmetic, soap and detergent industry.

The applications of SC CO_2 extraction in plant and animal lipids

In recent years several researchers have established that SC CO_2 can be used as a good alternative solvent for the following lipid processing operations:

- Separation of free fatty acids (FFA) from vegetable oil
- Separation of polyunsaturated fatty acids (PUFA) from animal lipids
- Refining and deodorization of vegetable oil
- Fractionation of glycerides
- Recovery of oil from oil-bearing materials
- Deoiling of lecithin
- Decholesterolization and delipidation of food products

Flavour Extracts and Essences

Essence is the mixture of natural flavour substances produced by certain plant species/sources, with an oily consistence, a lipophilic nature, relatively volatile and that can be carried out by steam distillation.
According to their chemical constitution, essence compounds are classified into three main groups:

- Isoprenoidic Compounds
- Fenilpropanic Compounds
- Linear Unbranched Chain Compounds (Perberes me vargje lineare te padegezuara)

Recovery of essential oils from their plant sources can be carried out by a variety of both old and new processes, such as steam distillation, hydrodiffusion, enfluerage, maceration, mechanical (cold) expression, solvent extraction, turbodistillation and most efficiently, supercritical fluid extraction.

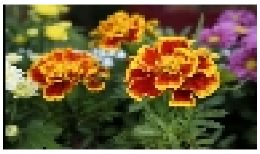
SC CO_2 EXTRACTED FLORAL FRAGRANCES

A large number of flowers have been extracted with subcritical (liquid) or supercritical (fluid) CO_2 to concentrate and isolate components of interest. To be mentioned are:

Rose Fragrance Jasmine Fragrance Lavender Inflorescence Fragrance



Sandalwood Fragrance Calendula Fragrance Orange Flower Fragrance

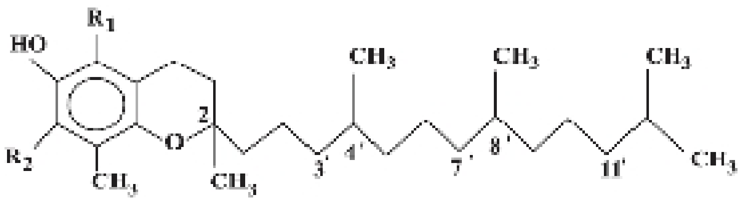

Natural Antioxidants

Natural antioxidants are those phenolic or polyphenolic compounds commonly occurring in plant materials, which interfere with the formation of free radicals and also deter the propagation of oxidation or the free radical chain reactions, thus preventing formation of hydroperoxides.
It is well known that some antioxidants occur naturally in different amounts in all foods and herbal medicines. There is definite scientific evidence that dietary supplementation of natural antioxidant nutrients, such as vitamins A, C, E, and flavonoids to foods may prevent many human diseases caused by oxidative damage including aging, cataract, coronary heart diseases, cancer, etc.
One of the important trends in the food industry today is the demand for natural antioxidants from plant materials.

The Main Representatives of Natural Antioxidants

The Supercritical Fluid Extraction (SC CO_2) is a very successful technic for the recovery of these natural antioxidants:

- **TOCOPHEROLS**



R_1	R_2	[illegible]
CH_3	CH_3	α-Tokoferol
CH_3	H	β-Tokoferol
H	CH_3	γ-Tokoferol
H	H	δ-Tokoferol

Fig. 4 Chemical structure of tocopherols.

- Commonly known as vitamin E
- Strong biological antioxidant activity.
- A very large group of commercial natural antioxidants currently being marketed

- FLAVONOIDS

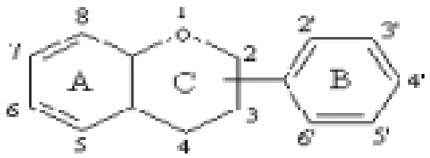


Fig. 5 Chemical structure of flavonoids

- The most common natural antioxidants, occuring widely in nature
- Synergism of flavonoids with other compounds.
- Classification into 5 chemical groups: (genuine flavonoids, calcons, aurons, isoflavans, neoflavons)

- CAROTENOIDS

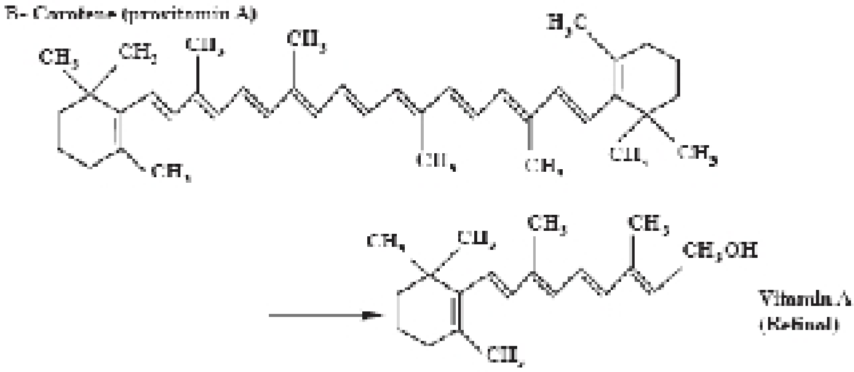


Fig.6 Structure of β-carotene and retinol (vitamin A).

- Natural fat-soluble substances

- Protection against uncontrolled oxidations, which are implicated in a multitude of degenerative diseases (e.g., tumors, cardiovascular diseases, etc.)
- One of the major groups of natural pigments that find widespred utilization in the food industry

Conclusions

- Nowadays, the application of advanced extraction methods under critical conditions in the natural products constitutes one of the most significant study objectives of nutritive sciences. It aims at employing them with the final intention of effectively obtaining qualitative, nutritive components.
- The classical techniques applied entwine a serial of frequently prolonged and complicated operations in the presence of strong chemicals and harsh conditions of work causing as such decomposition, thermal degradation, and toxic remains. Extraction under critical conditions is simpler, with many more advantages and possibilities to be automated.
- The desirable derivative for the extraction of natural products bearing special interest for food, cosmetic and pharmaceutical industry is CO_2 which results effective for the following reasons:
  - Extraction is carried out in low temperature, avoiding in this way degradation
  - Extracts are natural and bear multifarious characteristics
  - High productivity is attained in a short time and energy is saved
  - CO2 is a non-toxic, ecologic and cheap gas
  - The methodology is generally safe, posing no danger for the surrounding environment.

  Extraction by employing CO_2 has resulted effective through a variety of applications in plant and animal lipids, aromatic extractions and essences, food colorants, antioxidants and natural pesticides. The processes that follow present special interest for the group of plant and animal lipids:
  - Separation of free fat acids (FFA) from plant oil
  - Separation of polyunsaturated fatty acids (PUFA) from animal lipids
  - Glyceride fractioning
  - Removal of cholesterol and lipids from food products.
  - Along with lipids, another particularly important group is that of aromas and essences. Some of the main essences ever increasingly on demand are rose, jasmine, orange, and lavender ones.
  - Natural antioxidants bear the same importance for nutritive sciences. Besides, flavonoids, tochopherols, carotenoids can be included in their group. Their contribution to the enrichment of food products, protection of human organism

from oxidative processes and diseases related to them, and positive synergy effects adds to their appreciation and gives a drive to the research in the field of fractioning and studies on obtaining clean components and their inclusion in the elements of the daily diet.

- This contemporary scientific reality is intended to be present in our country in the future. It will offer possibilities for experimental researches and positive, technologic, economic and nutritional developments.

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