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Microwave Methods of Sample Preparation for the Purposes of Environmental Analysis

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Abstract

The application of microwave techniques for environmental sample preparation is described. Special attention has been given to the theoretical foundations and the possibilities of using microwave radiation in mineralization and extraction. These techniques are compared with conventional methods and examples of their application are supplied.

Keywords: mineralization, extraction, microwave techniques

Introduction

Environmental trace analysis is one of the most dynamically developing scientific branches. It may be stated without any doubt that it stimulates the creation or constant improvement of new measurement techniques. As far as the applications of environmental analysis are concerned one can include physico-chemical methods and those which are separate scientific branches, e.g. mass, molecular and atomic spectroscopy, magnetic nuclear resonance, all the kinds of chromatography, activation methods with the analysis of generated radionuclides and others. Contemporary laboratories and analytical workshops are equipped with more and more perfect measurment apparatus, allowing us to obtain results in a very short time [1].

Every analysis always consists of four equivalent stages: sample gathering, its proper preparation, analysis and the elaboration of the obtained results [2]. In the majority of cases the prepared samples are treated to separate the components prior to determination from the matrix, which is always in excess. This activity (Fig. 1) takes up to 55% of time reserved for the realization of this determination.

Various methods are applied in order to prepare the samples which may generally be divided into physical, chemical, chemical and physico-chemical ones [3]. Depending on the matrix in which the analytes (inorganic and organic) are localized in modern laboratory practice, the greatest interest, apart from adsorption and extraction methods [4,5], is aroused by mineralization. This technique is used successfully for the preparation of the whole group of inorganic analytes contained in different biological matrices. Generally, mineralization methods may be divided into classical and microwave techniques [2].

In this paper the theoretical foundations of microwave techniques as well as their application for environmental sample preparation has been presented. Moreover, the applied apparatus and its construction has been described with regard to defects and advantages of the technique. A part of this work has been devoted to discussion of the examples of the experimental application of microwave technique in reference to several various environmental samples, including medicinal plants and vegetables.

Classical Methods of Mineralization

The process of mineralization consists of converting the elements occuring in complicated combinations, prior to determination, into simple and easy (for final determinations) inorganic compounds. Independent of applied apparatus, the aim is to transform the material in such a way as to obtain a solution which can be subjected to further analyses [6]. Mineralization (as with any chemical process) should fulfill the following conditions:

- proceed quantitatively, i.e. the whole organic part of the examined object should undergo oxidation, whereas the inorganic part should stay quantitatively as the residue after the mineralization,
- proceed quickly – the procedure of digestion is often applied to a great number of samples and the velocity of its course influences in a decisive way the duration of analyses and their costs.

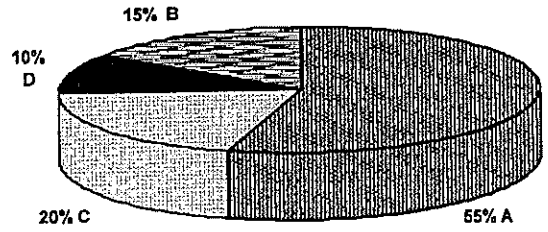


Fig. 1. The extenditure of time in % during the analysis: A – sample preparation, B – sample collection, C – analysis, D – data analysis, [2].

The classical methods of organic compounds mineralization are divided into:

- wet mineralization,
- dry mineralization.

Dry mineralization in its simplest form, consists of heating and roasting the substance in air in evaporating dishes or quartz, platinum or porcelain crucibles with the help of a burner or muffle furnace. Ashes obtained as a result of incineration are dissolved in a suitable acid and the mineral components are determined. This method, despite its analytical simplicity, does not give satisfying results in many cases, due to the difficulty of burning out carbon formed at the first stage of combustion. In order to increase the efficiency of the process various oxidizing agents such as nitric acid, nitrates of ammonium, potassium and magnessium oxide, are added (before or during combustion) to the sample; these agents accelerate mineralization. In order to prevent the volatilization of some elements under determination in the course of dry mineralization, "additions" are introduced which bind these elements into non-volatility and stability at the temperature of combustion combinations [6].

Wet mineralization allows the elimination of many disadvantages which occur during the incineration of samples and to avoid losses of volatile components. The process consists of the digestion of organic material with the help of liquid oxidizing agents such as concentrated nitric or perchloric acids and others used individually or in different combinations [6].

For a better understanding of the process of classical mineralization an example of wet combustion of plant material [7] is presented:

About 10 g of dry sample is placed in a Kieldahl flask with 50 ml of concentrated HNO_3 and left for the night. On the second day the flask is heated carefully, evaporating the acid up to half of its volume. To the cooled flask 50 ml of the mixture of concentrated HNO_3 and $HClO_4$ acids is added. Next, the flask is heated to evaporate the acids and to bleach the residue. The thickened deposit is washed out from the flasks into quartz beakers, evaported to dryness. Afterwards the diluted HCl (1:1) is added, the deposit is heated in water bath and filtered into measuring flasks, giving you a solution for analysis.

The classical methods, however, do not give satisfying effects: they are time-consuming (from several hours to several days), expensive (a great amount of reagents of high purity, great losses of energy during traditional heating) and do not give 100% recovery. The effect of removing the matrix depends, among others, on the parameters of heating: the method and the velocity, the degree of sample break-up, etc. [6]. That is why in literature greater and greater attention has been paid to the application of microwave techniques for heating as a new way of sample preparation [8].

Microwave Methods of Sample Preparation

Microwave

Electromagnetic waves having frequencies from 300,000 MHz to 300 MHz (wavelength from 1 mm to 1 m) are conventionally called microwaves. Within international assignations (Federal Communications Commission – Geneva, 1959) for industrial, scientific and medical applications of microwaves the following ranges of frequencies have been reserved: 915 ± 25 MHz; 2450 ± 50 MHz; 5800 ± 75 MHz; 22125 ± 125 MHz. Especially advantageous seems to be the application of microwave energy to such domains as radio-communication, radio-localization, navigation, medicine, industrial and food calorifics [9].

The interaction of electromagnetic waves with a material body may have different character, depending on the wave frequency and the electric properties of the material. The problem of microwave heating concerns dielectrics and dipole polarization (Fig. 2).

In the dielectric, the electric field polarizes the dipoles of molecules, which through orientation cause friction, the external symptom of which is heating the irradiated center in the whole mass at the same time. The time of this process is very short and depends only on the dielectric properties of the mineralized object. This type of heating is called through. The value of microwave field energy (P) conveyed to the sample is defined by the equation:

$$P = 2\Pi \nu \varepsilon fE^2 \tau \quad (1)$$

The amount of energy required to heat the sample (Q) is:

$$Q = P \operatorname{tg}\delta \quad (2)$$

$$Q = 2\Pi \nu \varepsilon fE^2 \operatorname{tg}\delta \tau \quad (3)$$

where:

- ν – the value of a sample (dielectric);
- ε – dielectric constant;
- E – the intensity of electromagnetic (microwave) field;
- f – field frequency in Hz: from 300 MHz to 300 GHz, corresponding to wavelengths (λ) from 1 m to 1 mm;
- τ – time of exposure (s).

The measure of transformation of energy of great frequency into heat is the value of the tangent of an angle of dielectric losses "tgδ", whereas the dielectric constant "ε" characterizes the properties of the center. The mineralized materials are treated as dielectrics, which diversly absorb the microwave energy and so undergo decomposition either more easily or with more difficulty. For example, for water and wood tgδ is 1500×10^{-4}, for ice 30×10^{-4}, that is during microwave heating of three equal by volume samples there must be at least 50 times greater heating of water and wood as compared with ice (Table 1).

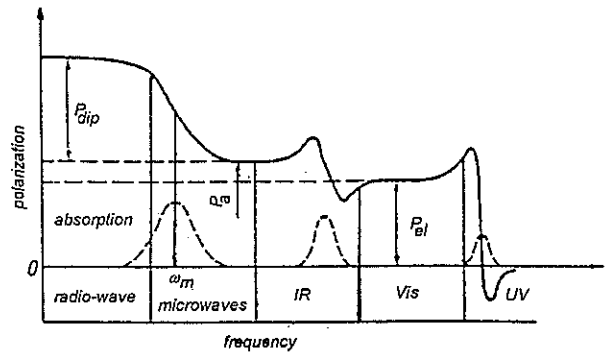


Fig. 2. Types of polarization for different electromagnetic waves, [10].

Microwave Mineralization

The first information concerning the application of microwave energy to mineralization of biological materials is dated 1975 [12]. In 1985, at a conference in Pittsburg, the apparatus for microwave digestion of solid samples of MDS-81 type, constructed by CEM firm was presented [13]. At present, microwave systems are widely applied in modern analytical laboratories, since they enable the decomposition of practically all organic and inorganic materials such as plant and animal tissues, food, wastes, garbage, oils, fuels, lubricating materials, metallurgical samples, soils, rocks, etc. [14].

Microwave systems may be divided into two groups: open and closed, among which low-pressure (to about 110 atm) systems may be distinguished. The characteristic feature of open systems is conducting the process of mineralization on one level of microwaves power, whereas in closed systems different levels of power are used [8, 14]. The system is built from two separate elements: a microwave furnace with the regulated magnetron power with the system measuring the time of heating and appropriate containers made of teflon, polypropylene of another material. Currently, there are many firms producing rotor mineralization (Fig. 3).

Table 1. Relative electric permeability (dielectric constant) and tg δ of materials in the band of microwaves (for the frequency 3 GHz), [11].

Material	Relative electric permeability	tg δ × 10^4
Air	1.0006	–
Tetlon	2.02	1.2
Polystyrene	3.38	11.0
Foamed polystyrene	1.025	064
Textolit	2.43	8.2
Plexiglass	2.6	60.0
Mica	7.0	3.0
Polyethylene	2.15	12.0
Wood	2.80	1500.0
Ice	3.0	30.0
Quartz	4.50	2.0
Water	80.0	1500.0

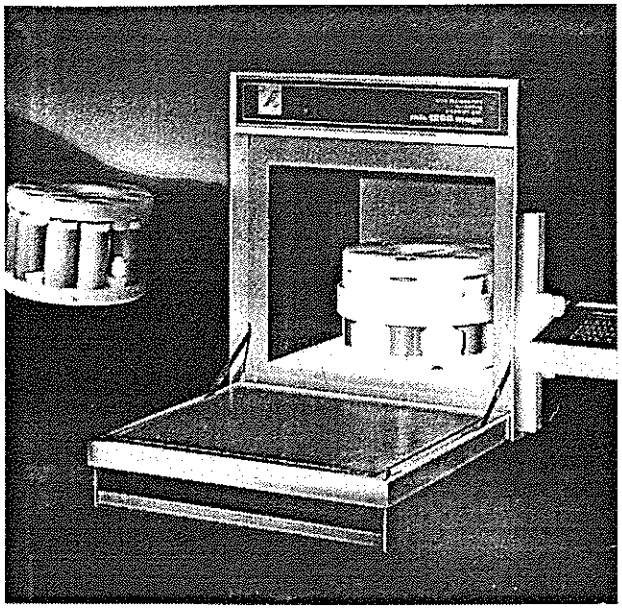


Fig. 3. A set for high pressure mineralization made by Milestone, [17].

The advantages of this method of mineralization are, among others:
- a short time of the digestion process of examined objects,
- great savings in chemical reagents and energy,
- high efficiency,
- small mass of a sample,
- the composition of mineralizing medium (in the case of biological material the nitric acid is most often used by itself or in combination with hydrogen peroxide solution).

The Exemplary Applications of Microwave Mineralization

Many microwave mineralizations have been conducted including solid samples such as pine and spruce needles, grass, vegetables (leek, celery, parsley, garlic, carrot), paper, cardboard, sugar and powdered sugar, blueberry, cherry, strawberry and gooseberry jams, liver or poultry and oxen, wastes after alcohol-distilling and after gas-producing and many other materials. The samples were prepared to meet the requirements of trace environmental analysis (the analysis of the content of heavy metals, sulphur, PAHs). The set for microwave mineralization was used; it consisted of a rotor MDR Milestone 1000/6/100/110 (MDR Technology, Bergamo, Italy) and a Samsung microwave oven of RE 1200 type (Seoul, Korea). 65% HNO_3 (3 ml) and 30% H_2O_2 (1 ml) were applied as an oxidizing medium. The program of mineralization has been presented in Table 2.

Table 2. The procedure of microwave mineralization, [2].

Stage	I	II	III	IV	V
Power (W)	240	0	240	400	560
Time (min)	0.5-1	2-7	5	5	5
% of power	30		30	50	70

The efficiency of the applied procedure was tested on the apparatus for TOC measuring a Model 5000 made by Shimadzu (Kyoto, Japan). Microwave mineralization allows removal of practically 100% of the organic matrix (Fig. 4) [18].

Microwave Extraction

The initial stage of many analyses, often determining its further course, is the isolation of an examined component in the form dissolved in the solution. The process of extraction, submited to the Nernst distribution law, enables isolation of the analyzed component from the examined matrix [19].

In conventional methods of sample isolation (Soxhlett apparatus, shaking with the solvent at room temperature), extraction with the solvent at the boiling point is impossible (the solvent going through the thimble in the Soxhlett apparatus undergoes cooling earlier). It is the cause of a smaller recovery of the examined substance from the given matrix [20]. Further limitations result from the possibility of forming azeotrope [21, 22]. The classical example is the mixture of acetone and hexane, thus the mixture having the volumetric composition 1:1 boils at 52°C, but the composition of the mixture. In such a case the vapour having the composition of 60% acetone and 40% hexane distills and this fact has a great importance in the case of extraction process. That is why the main factor increasing the efficiency of microwave extraction is reaching 100°C higher from the boiling point of the solvent in a very short time; this in turn is connected with the increase of pressure in the extraction vessel (Table 3).

First literature mentions concerning the microwave extraction are dated to 1986-87. At that time organic compounds were extracted from the matrices such as soil, seeds, food. In those investigations the microwave cooker was used to irradiate the suspension: solvent – sample [23].

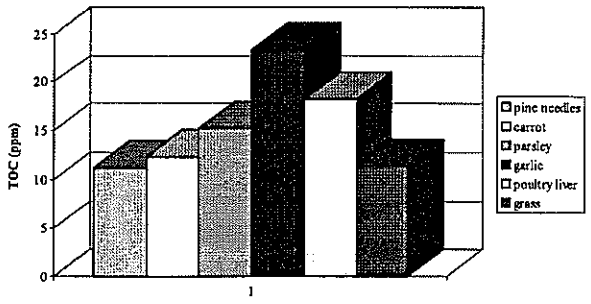


Fig. 4. The content of total organic carbon in various environmental samples (n = 6) after microwave mineralization.

Table 3. The values of solvents during microwave extraction.

Solvent	Boiling point (°C)	Extraction temperature in the vessel under the pressure of 11.9 atm.(°C)
Dichloromethane	40	140
Hexane - Acetone	52	150
Methanol -Water	90	190

At present the Microwave Extraction System (MES) allows isolation of PAHs, PCBs, pesticides from environmental samples, mycotoxins, fatty acids, cholesterol from food, and various organic substances from the products of the chemical and petrochemical industries. There are also procedures describing the extractions of essential oils from plants, as well as natural products from parsley, mint, garlic, etc. [23].

The mixture hexane/acetone (1:1) and dichloromethane are the most often applied solvents in microwave extraction. Hydrocarbon solvents theoretically do not absorb microwave energy, for them the tangent of an angle of dielectric losses is zero, which is why often there is a necessity of adding (> 10%) the polar component [24]. This is illustrated in Fig. 5.

Necessary elements of the set for MES are a generator of microwave energy and an extraction vessel that can be shut, having the possibility of permanent control of temperature and pressure, which allows maintaining the safety of the process as well as the possibility of programming the successive heating stages of the extraction.

MES is an alternative method to conventional methods of isolating the investigated compounds from complicated matrices (distillation, winterizing, lyophilization, extraction in the system: liquid – liquid (LLE), liquid – solid (SPE), solid phase microextraction (SPME), supercritical fluid extraction(SFE). It requires a smaller amount of solvent (Soxhlett 300 ml of solvent, MES – 30 ml), the time of extraction is considerably shorter, there is much greater recovery (Table 3). Moreover, there is a possibility of extraction of several samples at the same time.

The isolation of several PAHs from sea deposits has been conducted and compared with traditional methods. The average recovery of the extraction carried out at room temperature was 52%, whereas for MES the average recovery of 16 PAHs from 6 matrices was 70% at 80°C, 75% at

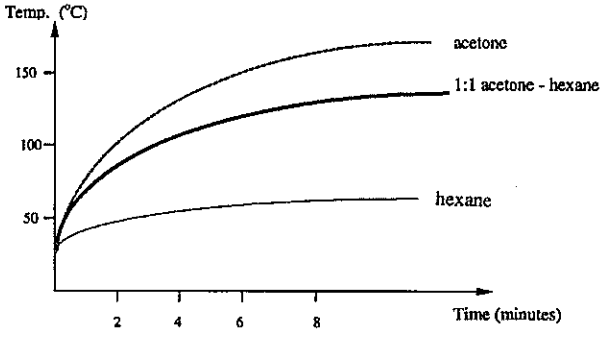


Fig. 5. Typical curves of microwave heating for: a) polar solvent – acetone, b) non-polar – hexane, c) the mixture of acetone – hexane, 1:1, [23].

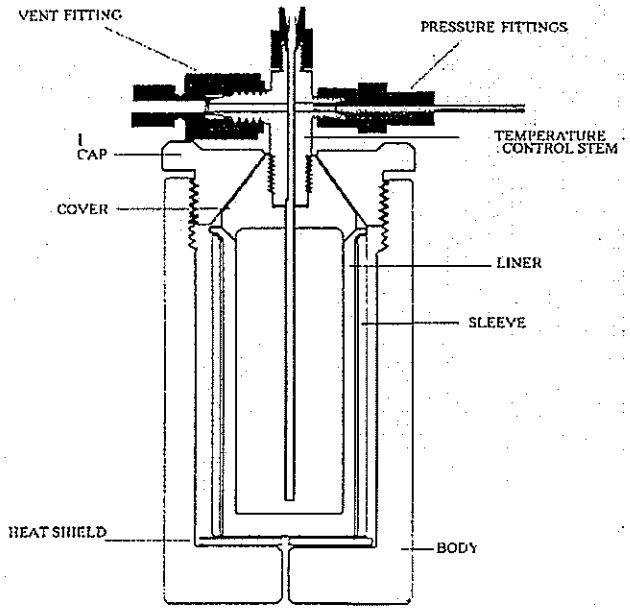


Fig. 6. The scheme of a vessel applied in the process of microwave extraction with the possibility of pressure and temperature control [17].

115°C and 75% at 145°C. The ability of MES system depended on the type of matrix and the isolated component. High recovery was noted for fluoranthene – 105% and benzo(a)anthracene – 85% [12]. The whole process in the Soxhlett aparatus lasted 8 hours; applying microwave heating reduced this time to 15 minutes. Comparing the time of microwave extraction in Soxhlett apparatus is presented in Table 4.

Fig. 7 compares degrees of the recovery after the application of different extraction methods. It follows from this comparison that comparable to SPE, however, the cost of the enterprise (especially the apparatus) is considerably lower.

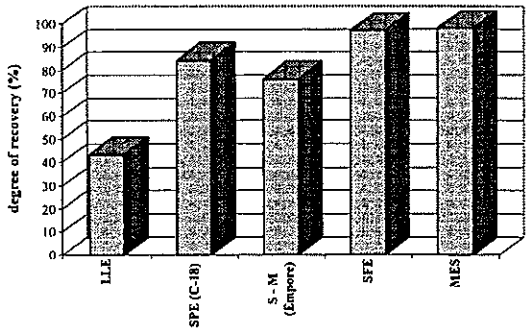


Fig. 7. The comparison of degrees of fluoranthene recovery for different methods of extraction.

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Table 4. Comparison of the time of microwave extraction with Soxhlett apparatus [24].

Sample	Substance determined	Method of determination	Type of solvent	Size of sample		Time of extraction	
				(ml)	(g)	ME (min)	Soxhlett (h)
Soil	initial contaminations	GC-MS	acetone : hexane (1 :1)	30	5	20	16-24
Sea deposits	PAHs	HPLC	dichloromethane	30	1	15	16
Soil	oils	gravimetry	acetone : hexane (1 : 1)	30	5	30	7
Soil	TPH	GC	acetone : hexane (1 : 1)	30	5	20	1
Soil	TPH	gravimetry	acetone : hexane (4 : 1)	30	2	30	4

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