

BS EN 13805:2014



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Foodstuffs — Determination of trace elements — Pressure digestion

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National foreword

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The UK participation in its preparation was entrusted to Technical Committee AW/275, Food analysis - Horizontal methods.

A list of organizations represented on this committee can be obtained on request to its secretary.

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Foreword

This document (EN 13805:2014) has been prepared by Technical Committee CEN/TC 275 "Food analysis - Horizontal methods", the secretariat of which is held by DIN.

This European Standard shall be given the status of a national standard, either by publication of an identical text or by endorsement, at the latest by April 2015 and conflicting national standards shall be withdrawn at the latest by April 2015.

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This document supersedes EN 13805:2002.

The changes between this edition and the previous one are as follows:

- a) restructuring of some clauses;
- b) 5.2.2, "Pressure digestion apparatus with conventional heating" was added;
- c) 5.2.3, "Pressure digestion apparatus with microwave-assisted heating" was added;
- d) 5.2.4, "Pressure digestion autoclave with microwave-assisted heating" was added;
- e) 6.3.2, "Acid addition" was revised"
- f) 6.4, "Example of pressure digestions" was completely revised and completed;
- g) bibliographic referenced were added;
- h) the whole document was editorially revised.

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1 Scope

This European Standard specifies a method for the pressure digestion of foodstuffs intended for the determination of elements. This method has been collaboratively tested in combination with atomic absorption (flame, electrothermal (ET), hydride, cold-vapour) techniques and ICP-MS. Other techniques such as e.g. ICP-OES, voltammetry or atomic fluorescence can be used in combination with this European Standard.

2 Normative references

The following documents, in whole or in part, are normatively referenced in this document and are indispensable for its application. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

EN 13804, *Foodstuffs - Determination of elements and their chemical species - General considerations and specific requirements*

3 Principle

The method is a physicochemical pressure digestion method used to mineralize the sample material and to prepare a measurement solution containing elements to be determined. The method described here is applied when the measurement has been validated in combination with this digestion method and reference is made to this European Standard. This procedure will relate to the total element content depending on reagents and determination procedures used.

The sample is homogenized avoiding contamination. Afterwards it is digested with nitric acid (sometimes with addition of other acids), at high temperatures and pressure in a closed vessel, applying conventional or microwave-assisted heating [1], [2], [3].

4 Reagents

The concentration of the trace elements in reagents and water used shall be low enough to not affect the results of the determination.

In the case of insufficient purity, it is necessary to purify nitric acid and hydrochloric acid in a distillation apparatus (see 5.4 and Figure A.1).

4.1 Nitric acid,

mass fraction w of not less than 65 % and density of approximately $\rho(\text{HNO}_3) = 1,4 \text{ g/ml}$.

4.2 Diluted nitric acid,

mix nitric acid (4.1) and water in a proportion of at least (1 + 9) parts by volume.

4.3 Hydrochloric acid,

w not less than 30 % and approximately $\rho(\text{HCl}) = 1,15 \text{ g/ml}$.

4.4 Hydrofluoric acid, w

not less than 40 % and approximately $\rho(\text{HF}) = 1,14 \text{ g/ml}$.

4.5 Hydrogen peroxide, w

not less than 30 % and approximately $\rho(\text{H}_2\text{O}_2) = 1,11 \text{ g/ml}$.

5 Apparatus and equipment

5.1 General

To minimize contamination, carefully clean all those apparatus coming into contact with the sample by treating it with diluted nitric acid (4.2) and afterwards with water. In addition, a stripping apparatus (Figure A.2) can be used for cleaning vessels or bottles.

5.2 Pressure digestion apparatus

5.2.1 General

Commercially available safety-tested pressure vessels made of acid-resistant materials and comprising inserts of acid-resistant, low-contamination materials are used to accommodate the sample material. The pressure vessels shall be able to withstand a temperature of at least 180 °C as well as be stably operated at this temperature. Ensure safe operation during the whole digestion process by suitable safety devices (e.g. bursting discs, pressure shell, power control).

5.2.2 Pressure digestion apparatus with conventional heating

Apparatus working with or without external autoclave pressure and designed for various working pressures should be used.

Inserts of polytetrafluoroethylene (PTFE), quartz glass, perfluoroethylenepropylene (FEP) [4], [5] or perfluoroalkoxy (PFA) can be used.

If digestion temperatures exceed 230 °C, the use of quartz glass is advantageous. For the determination of mercury and other elements, which are adsorbed easily on rough surfaces (e.g. antimony), quartz glass is recommended.

5.2.3 Pressure digestion apparatus with microwave-assisted heating

Systems with microwave-assisted heating shall be equipped with a temperature-measuring device. The temperature of the digestion solution can be measured internally and/or externally [6]. Reliable temperature measurement values are obtained, e.g. via sensors introduced into the pressure vessel. When using an infrared sensor for measuring the vessel temperature, it shall be ensured through consultation with the manufacturer that the displayed temperature corresponds to the temperature of the solution in the pressure vessel.

Do not use systems with microwave-assisted heating without a temperature-measuring device.

Inserts made of fluorinated plastics such as PTFE, advanced PFA or perfluoropropylvinylether-modified PTFE (e.g. TFM®) as well as quartz glass can be used.

If digestion temperatures exceed 230 °C the use of quartz glass is advantageous. For the determination of mercury and other elements, which are adsorbed easily on rough surfaces (e.g. antimony), quartz glass is recommended.

5.2.4 Pressure digestion autoclave with microwave-assisted heating

Alternatively to the apparatus described under 5.2.2 and 5.2.3, systems with a microwave heated autoclave can be used. The microwave energy is coupled into the absorption liquid and thus the digestion vessels are heated. The digestion vessels are sealed with an autoclave filling pressure of at least 40 bar (= 40×10^5 Pa).

Digestion vessels made of fluorinated plastics (PTFE and derivatives) or of quartz glass are recommended.

If digestion temperatures exceed 230 °C, the use of quartz glass is advantageous. For the determination of mercury and other elements, which are adsorbed easily on rough surfaces (e.g. antimony), quartz glass is recommended.

5.3 Ultrasonic bath with heating device

5.4 Subboiling distillation apparatus,

made of quartz glass or a similar material of high-purity fluoropolymers, according to Figure A.1, for distilling acids at temperatures below their boiling point.

5.5 Stripping apparatus,

according to Figure A.2, for cleaning vessels or bottles with hot acid vapour. Only vessels or bottles made of fluorinated plastics (PTFE and its derivatives), of quartz glass or glass are stable during the stripping procedure.

6 Procedure

6.1 General

WARNING — The application of this standard can involve the use of dangerous substances, operations and equipment. This standard does not purport, however, to address all the safety problems associated with its use. It is the responsibility of the user of this standard to establish the appropriate safety and health measures, which comply with applicable and current regulations.

During each step of the method, contamination shall be as low as possible. It shall be borne in mind that the digestion of carbon-rich materials (e.g. cellulose, carbohydrates, fats, oils) can result in explosions. Alcohol shall be evaporated since its contact with concentrated nitric acid results in delayed violent reactions. Perchloric acid shall not be used for pressure digestion.

Before the pressure digestion apparatus is used, read operating manual and safety instructions carefully. Particular attention shall be paid to the risk to the laboratory staff posed by nitrogen oxides.

6.2 Sample preparation

Prepare the samples using normal household practices. Avoid as far as possible any contamination with the elements to be determined (e.g. when determining chromium and nickel, no stainless steel equipment shall be used for the sample preparation), see EN 13804. The sample preparation shall ensure homogeneous starting material for the test portion.

6.3 Pressure digestion conditions

6.3.1 Sample

Adapt the sample weight to the capacity of the digestion vessel and the maximum permissible pressure increase; in doing so, do not deviate from the manufacturer's information for safety reasons. The maximum sample weight depends on the samples carbon content and water content.

In the case of carbon-rich samples, such as fats and carbohydrate rich foodstuffs, a maximum of 200 mg (to the nearest milligram) of fresh mass can as a rule be digested [5]. If the carbon content is lower, the mass of the sample may be increased up to 3 g of fresh mass or 4 ml of liquid. Depending on the volume of the digestion vessels, other sample weights may be used; but the ratio of sample mass to acid volume (6.3.2), however, shall not be changed.

In the case of samples in powder form with a weight of approximately 200 mg, add 1 ml of water and mix thoroughly in order to obtain a sample that is well suspended in the water. Take care to ensure that mixing does not result in splashing losses or in damage to the digestion vessels.

In the case of materials that cannot be mixed with water, for example fatty materials, do not add water.

With alcoholic samples it is imperative that the alcohol is completely removed after transferring the sample into the digestion vessel, by gentle heating, e.g. in the heatable ultrasonic bath (5.3), at 60 °C, and before the adding the acid. In doing so, it shall be ensured that there are no losses of the elements to be determined.

6.3.2 Acid addition

WARNING — Samples which absorb microwave energy and are not fully covered by the acid, can result in local overheating of the digestion vessel, resulting in local melting, followed by bursting of the vessel.

WARNING — Perchloric acid shall never be used in pressure digestions, not even in small amounts.

The volume of acid required for the digestion depends on the nature of the sample material. Usually 3 ml of nitric acid (4.1) are sufficient to digest the required amounts. Systems with microwave-assisted heating usually require a higher amount of acid. For carbon-rich samples, it may be necessary to increase the amount of acid and to reduce the sample mass. 0,5 ml to 1 ml of hydrogen peroxide (4.5) can be added to prevent adhesion of the samples to the wall of the digestion vessel and to achieve complete mixing with the acid. Hydrogen peroxide causes increased gas formation when the acid reacts with the sample, thereby resulting in thorough mixing. Furthermore, less nitrogen oxides are generated.

Following the addition of the acid, thoroughly mix the sample. Avoid any adhesion of sample clumps to the wall of the digestion vessel. Before closing the vessels, cover the sample with acid. In the case of microwave-assisted digestion, the total volume of the liquid including the sample should correspond with the manufacturer's information. It is recommended to perform a pre-reaction following the acid addition, at room temperature with the vessel open and loosely covered. In the case of reactive samples, this may be carried out overnight. Following the pre-reaction, close the vessel as described by the vessel manufacturer.

For digestions where an addition of hydrochloric acid is necessary, e.g. when analysing the elements tin, antimony and iron, proceed as follows: the addition of at least 0,5 ml of hydrochloric acid (4.3) is required to prevent losses due to adsorption to the vessel wall and to keep the elements fully solved. First add nitric acid and mix with the sample. Add the hydrochloric acid only after the spontaneous reaction caused by the nitric acid is finished. Seal the digestion vessel immediately after addition of hydrochloric acid to make the resulting chlorine gas available for reaction.

The surface finish of the digestion vessels (e.g. roughness) is decisive for the adsorption to the vessel wall. Here, quartz vessels are preferable to digestion vessels made of fluoroplastics. Therefore, following each digestion, check and clean the vessels and remove any deposits on the vessel wall completely.

The digestion of some foodstuffs, e.g. containing silicates or titanium dioxide, may require the addition of hydrofluoric acid (4.4). When using hydrofluoric acid, the appropriate safety guidelines shall be followed. Only inserts of fluoroplastics may be used for digesting with hydrofluoric acid. Quartz glass is damaged irreversibly when it comes into contact with hydrofluoric acid.

6.3.3 Digestion temperature

Digest the samples at a temperature of at least 180 °C. Heat pressure vessels using microwave-assisted heating to the digestion temperature by applying a low heating rate with reduced microwave power, adapted to the reaction behaviour of the samples. Identify the required digestion temperature and in consequence the resulting completeness of the decomposition by the subsequent measurement procedure. Higher temperatures result in lower contents of residual carbon in the digestion solutions. Thereby the background interferences in ET-AAS and ICP-OES measurements are reduced. Interferences in e.g. chromium

determination and influence on signal sensitivity for arsenic and selenium on ICP-MS measurements are reduced [7], [8], [9] and trouble-free voltammetric measurements can be performed.

It has been found that the quality of the digestion improves with an increasing digestion temperature [10], [11]. If organic arsenic compounds are present in the food, a temperature of 320 °C can be necessary if hydride AAS is used for the subsequent determination of arsenic [12]. The temperature of 320 °C can be reached e.g. by pressure digestion apparatus with conventional heating (5.2.2). For the determination of selenium with hydride AAS, the maximum temperature for digestion is 280 °C [13].

For all steps of the digestion process, follow to the manufacturer's safety provisions.

6.3.4 Digestion time

In the case of pressure digestion using conventional heating, the usual digestion time after attaining the final digestion temperature is at least 1,5 h. In the case of discrete pressure vessels, the complete time for digestion is up to 15 h. With microwave systems, the digestion time should be at least 20 min after the digestion temperature has been attained.

6.3.5 Cooling

To reduce the excess pressure, cool the still sealed pressure vessel to a temperature of less than 40 °C.

6.3.6 Treatment of the digestion

Once the digestion vessel has been cooled down, carefully open it whilst suctioning off the liberated gases. Depending on the construction of the pressure vessel, this is preceded by ventilation of the vessel in order to relieve any residual pressure. Afterwards, place the digestion vessel initially under a fume hood until no brown fumes are visible. It is recommended to degas the digestion solution in the ultrasonic bath. The digestion solution shall be clear and its volume shall be roughly the same as before the digestion. An obvious reduction in volume suggests that the pressure vessel was not tight. Repeat the digestion in such cases. Once the digestion solution has been brought to room temperature, make it up to a defined volume with water. Transfer this solution to vessels made of quartz glass (for the determination of mercury use appropriate vessels, e.g. quartz glass or fluorinated ethylene propylene (FEP)) or suitable plastics vessels (e.g. made of FEP or PFA).

NOTE Yellow coloured digestion solutions are caused by incompletely digested organic substances. They can be the result of too high a sample mass and/or too low a digestion temperature. Digestion temperatures exceeding 200 °C usually do not result in yellow coloured digestion solutions. Blue coloured digestion solutions are the result of dissolved nitrogen oxides. Following dilution with water, the blue colour disappears.

6.3.7 Quality control of the analysis

In order to check the results of the analysis, analyse samples together with reference materials having reliably known contents of the elements to be determined. Analyse the reference materials during all the steps of the method, starting from digestion.

Furthermore, perform blank digestions by carrying out the digestion in the same way as the samples described under 6.3.1 to 6.3.6 but without a sample. The blank digestion serves to check for blank values which may have been caused by the vessels and acids or by other contaminations.

6.4 Example of pressure digestions

6.4.1 Example of a high-pressure ashing digestion with conventional heating

When using a 70 ml vessel, weigh in 1 g to 2 g of meat or 3 g of lettuce (fresh mass) or 200 mg of powdered samples (e.g. flour). If necessary, partially evaporate liquid foodstuffs. In cases of foodstuff with a high fat content, reduce the weighed sample. A reduction to 300 mg for chocolate and to 100 mg to 200 mg for fat and oils is recommended. The manufacturer's recommendations are also to be followed.

In the case of powdered samples, add 1 ml of water and mix thoroughly to obtain a sample which is well suspended in the water. Take care to ensure that mixing does not result in splashing losses. Add 3 ml of nitric acid (4.1) and seal the digestion vessel and the pressure vessel as prescribed by the manufacturer.

Afterwards heat from e.g. room temperature to 150 °C within 60 min, then to 300 °C within 40 min. Hold at 300 °C for 90 min before cooling down.

6.4.2 Example of a microwave digestion

When using 70 ml to 100 ml vessels, weigh in 1 g to 2 g of meat or 3 g of lettuce (fresh mass) or 200 mg of powdered samples (e.g. flour). If necessary, partially evaporate liquid foodstuffs. In cases of foodstuff with a high fat content, reduce the weighed sample. A reduction to 300 mg for chocolate and to 100 mg to 200 mg for fat and oils is recommended. The manufacturer's recommendations are also to be followed.

In the case of powdered samples, add 1 ml of water and mix thoroughly to obtain a sample which is well suspended in the water. Take care to ensure that mixing does not result in splashing losses. Add at least 3 ml of nitric acid (4.1) and seal the digestion vessel as prescribed by the manufacturer. It is important to achieve the given minimum filling volume of the manufacturer.

Start the digestion by applying low microwave energy and slowly raise the energy supply to the maximum power in order to achieve the final digestion temperature. For example, start with 100 W, raise to 600 W within 5 min, hold for 5 min, raise to maximum power in order to achieve the final digestion temperature (e.g. 200 °C), hold for 20 min, cool down for at least 20 min to 25 min.

The digestion conditions, i.e. mass of sample, number of vessels and microwave power, shall be chosen such that a digestion temperature of at least 180 °C is achieved.

6.4.3 Example of a digestion in a microwave-heated autoclave

When using 30 ml vessels, depending on the water content, weigh in 0,5 g to 1 g of meat or up to 5 g of liquid foodstuff (beer, juice, wine). If necessary, partially evaporate liquid foodstuffs. In cases of foodstuff with a high fat content, reduce the weighed sample. A reduction to 200 mg for chocolate and to 100 mg for fat and oils is recommended. Weigh in 200 mg in the case of powdered samples (e.g. flour). The manufacturer's recommendations shall also be followed.

In the case of powdered samples, add 1 ml of water and mix thoroughly to obtain a sample which is well suspended in the water. Take care to ensure that mixing does not result in splashing losses. Finally, add 2 ml of nitric acid (4.1) and 1 ml of hydrogen peroxide (4.5) and seal the digestion vessel with a fitted lid.

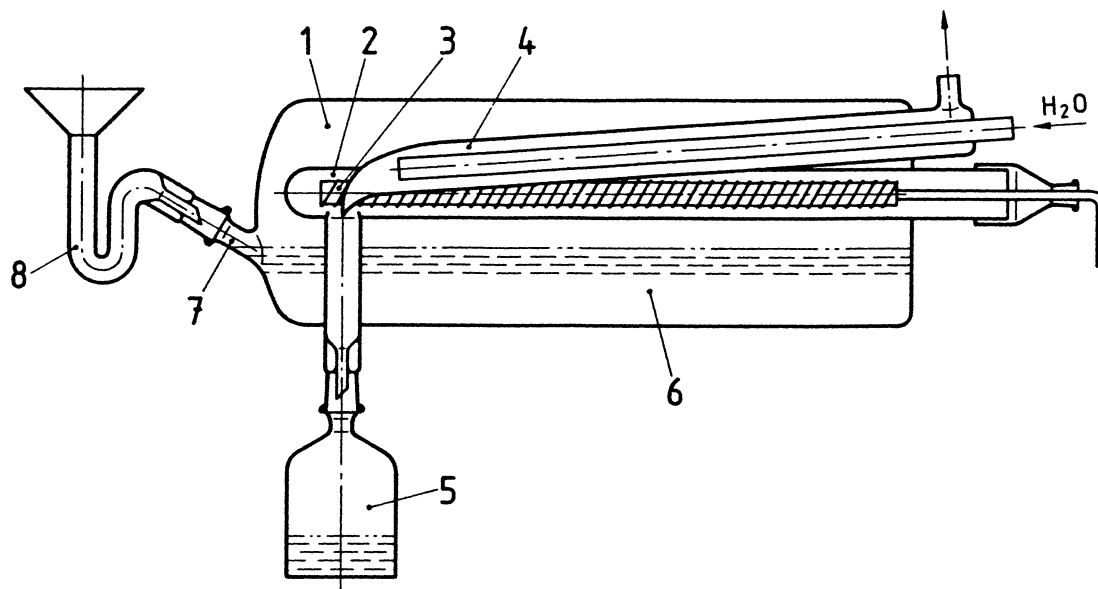
The addition of HCl for digestion is only allowed for specifically constructed autoclave cabinets, referred to the manufacturer's instructions.

For an effective absorption of the microwave power by the absorption liquid, add a defined amount of water, which contains 2 vol% hydrogen peroxide and 0,5 vol% sulfuric acid to the autoclave vessel. The amount of absorption liquid depends on the size of the autoclave vessel. Note the manufacturer's recommendations. After insertion of the digestion vessel and sealing the pressure vessel, fill with inert gas until the boost pressure of 40×10^5 Pa to 80×10^5 Pa.

First conduct the digestion with reduced microwave energy to avoid spontaneous reactions (e.g. 700 W in 12 min to 140 °C). Subsequently apply high microwave power (e.g. 1 000 W) to heat the sample to 260 °C within 15 min. Maintain the end-temperature for at least 20 min to ensure complete digestion.

Annex A
(normative)

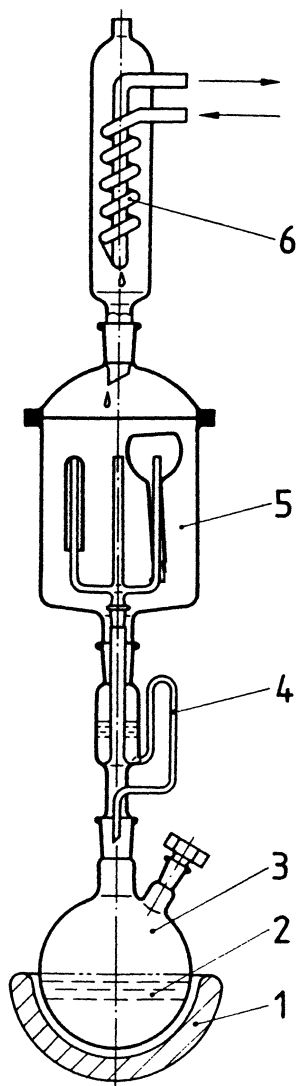
Figures of apparatus



Key

- 1 distillation chamber
- 2 quartz glass tube
- 3 heating filament
- 4 cold finger
- 5 bottle containing purified acid
- 6 acid to be distilled
- 7 connecting piece
- 8 charging funnel

Figure A.1 — Quartz glass subboiling-distillation apparatus



Key

- 1 heater
- 2 nitric acid
- 3 round bottom flask
- 4 siphon
- 5 vapour chamber
- 6 reflux condenser

Figure A.2 — Stripping apparatus

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