
**Soil, treated biowaste, sludge and
waste — Digestion of aqua regia
soluble fractions of elements**

*Sols, biodéchets traités, boues et déchets — Digestion des éléments
solubles dans l'eau régale*

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Foreword

ISO (the International Organization for Standardization) is a worldwide federation of national standards bodies (ISO member bodies). The work of preparing International Standards is normally carried out through ISO technical committees. Each member body interested in a subject for which a technical committee has been established has the right to be represented on that committee. International organizations, governmental and non-governmental, in liaison with ISO, also take part in the work. ISO collaborates closely with the International Electrotechnical Commission (IEC) on all matters of electrotechnical standardization.

The procedures used to develop this document and those intended for its further maintenance are described in the ISO/IEC Directives, Part 1. In particular, the different approval criteria needed for the different types of ISO documents should be noted. This document was drafted in accordance with the editorial rules of the ISO/IEC Directives, Part 2 (see www.iso.org/directives).

Attention is drawn to the possibility that some of the elements of this document may be the subject of patent rights. ISO shall not be held responsible for identifying any or all such patent rights. Details of any patent rights identified during the development of the document will be in the Introduction and/or on the ISO list of patent declarations received (see www.iso.org/patents).

Any trade name used in this document is information given for the convenience of users and does not constitute an endorsement.

For an explanation of the voluntary nature of standards, the meaning of ISO specific terms and expressions related to conformity assessment, as well as information about ISO's adherence to the World Trade Organization (WTO) principles in the Technical Barriers to Trade (TBT), see www.iso.org/iso/foreword.html.

This document was prepared by Technical Committee ISO/TC 190, *Soil quality*, Subcommittee SC 3, *Chemical and physical characterization*, in collaboration with the European Committee for Standardization (CEN) Technical Committee CEN/TC 444, *Environmental characterization of solid matrices*, in accordance with the Agreement on technical cooperation between ISO and CEN (Vienna Agreement).

Any feedback or questions on this document should be directed to the user's national standards body. A complete listing of these bodies can be found at www.iso.org/members.html.

Introduction

Regarding the comparability of the procedure described in this document with those of the other standards mentioned above the next remarks can be made:

- This document describes the digestion of solid samples with aqua regia.
- Differences in the procedures of the different standards are small. An important difference between the reflux procedures as described in ISO 11466 and EN 13657 and EN 16174 concerns the waiting time after addition of the acid to the sample, before the digestion starts. ISO 11466 specifies a waiting time of 16 h, both European standards state that the digestion can start after the first strong reactions have ceased. In validation work it was proven that the difference between 2 h and 16 h of waiting was negligible, therefore this document follows the approach of EN 13657 and EN 16174.
- The heating block procedure was added to the reflux and microwave digestion procedures. The procedure was adopted from the Dutch standard NEN 6961, which specifies a boiling time of 2 h to 4 h. This document specifies a boiling time of 2 h.

The methods specified in this document are providing multi-element aqua regia digestion techniques for soil, treated biowaste, sludge and waste prior to analysis. It is known that the digestion of environmental samples with aqua regia will not necessarily lead to complete element recoveries, and that the extract from a test sample may not reflect the total concentrations of the target analytes. However, for most environmental applications the result obtained based upon digestion methods specified in this document are considered to be fit for the intended purpose.

This document is validated for several types of matrices as indicated in [Table 1](#).

Table 1 — Matrices for which this document is validated

Matrix	Materials used in the validation test
Sludge	Municipal sludge
	Industrial sludge
	Sludge from electronic industry
	Ink waste sludge
	Sewage sludge
Biowaste (Method A)	Compost
	Composted sludge
Soil	Agricultural soil
	Sludge amended soils
Waste	City waste incineration fly ash ("oxidised" matrix)
	City waste incineration bottom ash ("silicate" matrix)
	Ink waste sludge (organic matrix)
	Electronic industry sludge ("metallic" matrix)
	BCR 146R (sewage sludge)
	BCR 176 (city waste incineration ash)

WARNING — Persons using this document should be familiar with usual laboratory practice. Some of the reagents used in this document are highly corrosive and very toxic. Safety precautions are absolutely necessary, not only due to the strong corrosive reagents, but also to the high temperature and high pressure.

The use of laboratory-grade microwave equipment with isolated and corrosion resistant safety devices is required. Domestic (kitchen) type microwave ovens shall not be used, as corrosion by acid vapours may compromise the function of the safety devices and prevent the microwave

magnetron from shutting off when the door is open, which could result in operator exposure to hazardous levels of microwave energy.

All procedures should be performed in a fume hood or in closed force-ventilated equipment. By the use of strong oxidising reagents, the formation of explosive organic intermediates is possible, especially when dealing with samples with a high organic content. Do not open pressurized vessels before they have cooled down. Avoid contact with the chemicals and the gaseous reaction products.

IMPORTANT — It is absolutely essential that tests conducted according to this document be carried out by suitably trained staff.

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Soil, treated biowaste, sludge and waste — Digestion of aqua regia soluble fractions of elements

1 Scope

This document specifies two methods for digestion of soil, treated biowaste, sludge and waste by the use of an aqua regia digestion.

Digestion with aqua regia will not necessarily accomplish total decomposition of the sample. The extracted analyte concentrations may not necessarily reflect the total content in the sample but represent the aqua regia soluble metals under the condition of this test procedure. It is generally agreed that for environmental analysis purposes, the results are fit for the intended purpose to protect the environment.

This document is applicable for the following elements:

Aluminium (Al), antimony (Sb), arsenic (As), barium (Ba), beryllium (Be), boron (B), cadmium (Cd), calcium (Ca), chromium (Cr), cobalt (Co), copper (Cu), iron (Fe), lead (Pb), magnesium (Mg), manganese (Mn), mercury (Hg), molybdenum (Mo), nickel (Ni), phosphorus (P), potassium (K), selenium (Se), silver (Ag), sodium (Na), strontium (Sr), sulfur (S), tellurium (Te), thallium (Tl), tin (Sn), titanium (Ti), vanadium (V), and zinc (Zn).

This document can also be applied for the digestion of other elements, provided the user has verified the applicability.

2 Normative references

There are no normative references in this document.

3 Terms and definitions

For the purposes of this document, the following terms and definitions apply.

ISO and IEC maintain terminological databases for use in standardization at the following addresses:

- ISO Online browsing platform: available at <https://www.iso.org/obp>
- IEC Electropedia: available at <http://www.electropedia.org/>

3.1

aqua regia

digestion (3.2) solution obtained by mixing 1 volume of nitric acid (mass fraction of 65 % to 70 %) and 3 volumes of hydrochloric acid (mass fraction of 35 % to 37 %)

Note 1 to entry: These mass percentages agree with the concentrations of 6.2 and 6.3.

3.2

digestion

mineralization of the organic matter of a sample and dissolution of its mineral part, more or less completely, when reacting with a reagent mixture

3.3

dry residue

dry matter expressed as a percentage by mass after drying at 105 °C ± 5 °C to the constancy of weight

3.4

laboratory sample

sample (3.5) intended for laboratory inspection of testing

[SOURCE: ISO 11074:2015, 4.3.7]

3.5

sample

portion of material selected from a larger quantity of material

[SOURCE: ISO 11074:2015, 4.1.17]

3.6

test portion

analytical portion

quantity of material of proper size for measurement of the concentration or other properties of interest, removed from the *test sample* (3.7)

Note 1 to entry: The test portion may be taken from the laboratory sample directly if no preparation of sample is required (e. g. with liquids), but usually it is taken from the prepared test sample.

Note 2 to entry: A unit or increment of proper homogeneity, size and fineness, needing no further preparation, may be a test portion.

[SOURCE: ISO 11074:2015, 4.3.15]

3.7

test sample

analytical sample

portion of material resulting from the *laboratory sample* (3.4) by means of an appropriate method of sample pre-treatment and having the size (volume/mass) necessary for the desired testing or analysis

[SOURCE: ISO 11074:2015, 4.1.3]

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4 Principle

A test portion is digested with aqua regia according to one of the following heating procedures:

- Method A: procedure under atmospheric conditions
 - A1: reflux for (120 ± 10) min, followed by filtration/centrifugation;
 - A2: heating block at (105 ± 5) °C for (120 ± 10) min, followed by filtration/centrifugation.
- Method B: microwave digestion
 - B1: Temperature controlled procedure: at (175 ± 5) °C for (10 ± 1) min in a closed vessel followed by filtration/centrifugation.

5 Interferences and sources of errors

The container in which the sample is delivered and stored can be a source of errors. Its material shall be chosen according to the elements to be determined (e.g. elemental Hg can penetrate polyethylene walls very fast in both directions. Glass can contaminate samples with its major elements: e.g. B, Na, K, Si and Al).

Grinding or milling samples includes a risk of contamination of the sample by the environment (air, dust, wear of milling equipment). Due to elevated temperature losses of volatile compounds are possible.

For the determination of elements forming volatile compounds (e.g. Hg, As) special care has to be taken during sample pre-treatment.

All glassware and plastics ware shall be adequately cleaned and stored in order to avoid any contamination.

In the case of filtration of the digested solution it is necessary to take care that the filtration procedure does not introduce contaminants.

Ensure that all of the test portion is brought into contact with the acid mixture in the digestion vessel.

Some elements of interest can be lost due to precipitation with ions present in the final digest solution, e.g. low soluble chlorides, fluorides and sulfates.

6 Reagents

Use only acids and reagents of recognized analytical grade to avoid high blank values for subsequent analytical measurements. Use a test blank solution throughout the procedure applying all steps with the same amount of acids, but without a sample.

6.1 Water, e.g. deionized.

6.2 Hydrochloric acid, $c(\text{HCl}) \approx 12 \text{ mol/l}$.

6.3 Nitric acid, $c(\text{HNO}_3) \approx 15 \text{ mol/l}$.

6.4 Nitric acid, $c(\text{HNO}_3) \approx 0,5 \text{ mol/l}$.

Dilute 35 ml nitric acid (6.3) to 1 l with water (6.1).

6.5 Antifoaming agent, e.g. *n*-dodecane ($\text{C}_{12}\text{H}_{26}$) or *n*-octanol ($\text{C}_8\text{H}_{18}\text{O}$) are suitable.

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7 Apparatus

7.1 General

Usual laboratory apparatus. All glassware and plastics ware shall be adequately cleaned and stored in order to avoid any contamination.

Depending upon the concentration of the element of interest, particular care should be exercised with respect to the effective cleaning of the vessels.

7.2 Method A — Apparatus for thermal heating under atmospheric conditions

7.2.1 Method A1 — Thermal heating under reflux conditions

7.2.1.1 Digestion vessel, temperature- and pressure-resistant and capable of containing the mixture of sample and digest solution, for example a quartz vessel. The digestion vessel shall have a volume of at least 5 times of the volume of the aqua regia used. The inner wall of the vessel shall be inert and shall not release substances to the digest in excess of the purity requirements of the subsequent analysis.

NOTE 1 Silica or borosilicate glass vessels can be used instead of quartz vessels.

NOTE 2 It can be necessary to periodically clean the digestion vessels with a suitable surfactant to remove persistent deposits.

7.2.1.2 Reflux condenser, adaptable to the digestion vessel (7.2.1.1).

7.2.1.3 Absorption vessel, volatile species trap, in an open digestion system capable of trapping one or more volatile measurement species, adaptable to the reflux condenser (7.2.1.2).

7.2.1.4 Heating device, for example a heating mantle, thermostatic controlled, or an aluminium block thermostat.

7.2.2 Method A2 — Thermal heating with a heating block with containers

7.2.2.1 Digestion tube, 50 ml propylene tube with a screw cap from polypropylene.

The part of the tube not being heated and the screw cap function as a condenser, but are not really a reflux system. The material of the tube and screw cap need to be tested in order to be sure that release of elements of interest does not take place. Other materials and vessels with other volumes than mentioned above are allowed to be used if suitability has been proven.

7.2.2.2 Temperature controlled heating block, heating block able to heat the tube(s) to a temperature of (105 ± 5) °C.

7.3 Method B — Microwave digestion with temperature control, closed vessels

7.3.1 Digestion vessel, for pressurized microwave digestion, typically 100 ml volume, reagent-, temperature- and pressure-resistant and capable of containing the mixture of sample and digest solution. The vessel shall be suitable for the safe application in the temperature and pressure range applied, capable of withstanding pressures of at least 3 000 kPa.

Digestion vessels made of perfluoroalkoxylalkane (PFA), modified polytetrafluoroethylene (PTFE) or quartz, and equipped with a safety pressure releasing system to avoid explosion of the vessel, shall be used. The inner wall of the vessel shall be inert and shall not release contaminations to the digest solution.

It can be necessary to periodically clean the digestion vessels with a suitable surfactant to remove persistent deposits.

7.3.2 Microwave digestion system, corrosion resistant and well ventilated. All electronics shall be protected against corrosion for safe operation.

Use a laboratory-grade microwave oven with temperature feedback control mechanisms.

The microwave digestion system should be able to control the temperature with an accuracy of ± 5 °C and automatically adjust the microwave field output power within 2 s of sensing. Temperature sensors shall be accurate to ± 2 °C, including the final reaction temperature of (175 ± 5) °C. Temperature feedback control provides the primary performance mechanism for the method. Due to the variability in sample matrix types and microwave digestion equipment (i.e. different vessel types and microwave designs), control of the temperature during digestion is important for reproducible microwave heating and comparable data. Manufacturer specifications of the microwave digestion system must fit these specifications. The accuracy of the temperature measurement system should be periodically tested on blank samples at an elevated temperature according to the manufacturer's instructions. If the temperature deviates by more than 2 °C from the temperature measured by an external, calibrated temperature measurement system, the microwave temperature measurement system should be re-calibrated.

7.4 Sample containers, plastics and glass containers are both suitable.

7.5 Filter paper, usually with a pore size of 0,45 μm and resistant to the diluted aqua regia final digestion solution.

- 7.6 **Volumetric flasks**, usually of nominal capacity of 50 ml or 100 ml.
- 7.7 **Analytical balance**, with an accuracy of 1 mg or better.
- 7.8 **Boiling aids**, anti-bumping granules or glass beads, diameter 2 mm to 3 mm, acid washed.

8 Procedure

8.1 General

Pre-treat, soil, sludge and biowaste samples according to e.g. EN 16179 or ISO 11464 and waste samples according to e.g. EN 15002.

Determine the dry matter content, depending on the matrix of the sample, e.g. according to EN 15934.

For waste samples the next remarks apply:

- Pre-treatment should include drying or grain size reduction below a particle size of 250 µm for solid waste or homogenizing by use of a high speed mixer or sonification for liquid waste samples.
- The mass of test portion for a single digestion has to be selected in a way, that:
 - it is representative for the laboratory sample;
 - it complies with the specifications of manufacturer of the digestion unit.

Referring to the manufacturer's instructions, the upper limits of mass of the test portion shall be taken into account.

- For representativeness reasons a mass above 200 mg is to be preferred for the test portion. Follow, for safety reasons, the manufacturer's instructions regarding the maximum amount of organic carbon in the sample.

8.2 Blank test

Carry out a reagent blank test digestion in parallel with the determination, using the same procedure and the same quantities of all the reagents as in the determination, but omitting the test portion. The laboratory shall define acceptable limits.

NOTE The measurement of a blank is introduced to determine the contribution of the extracting solution, glassware, digestion tube and filter paper used to the measured value.

8.3 Method A — thermal heating under atmospheric conditions

8.3.1 Method A1 — Thermal heating under reflux conditions

Weigh approximately 3 g of the test sample (waste samples 1 g to 10 g), with an accuracy of 0,001 g (or at least three significant figures), and transfer to the digestion vessel (7.2.1.1).

In case of dry samples moisten the test portion with about 0,5 ml to 1,0 ml of water (6.1) and add, dropwise, if necessary, to reduce foaming, with mixing, (21,0 ± 0,1) ml of hydrochloric acid (6.2) followed by (7,0 ± 0,1) ml of nitric acid (6.3). Connect the reflux condenser (7.2.1.2) to the digestion vessel (7.2.1.1). Fill the absorption vessel (7.2.1.3) with approximately 15 ml nitric acid (6.4). Connect the absorption vessel to the reflux condenser, and let stand at room temperature until any effervescence almost ceases to allow for slow oxidation of the organic matter in the sample.

The time of standing at room temperature can have an influence on the digestion rate of aqua regia. For consistency, it is recommended to start heating as soon as possible after the first strong reaction has ceased.

30 ml of aqua regia is only sufficient for the oxidation of about 0,5 g organic carbon. If there is any doubt of the amount of carbon present, estimate the amount of carbon in the sample or carry out a determination of TOC. If there is more than 0,5 g of organic carbon in the test portion, proceed as follows.

Allow first reaction with the aqua regia to subside. Then add an extra 1 ml of nitric acid (6.3) only to every 0,1 g of organic carbon above 0,5 g. Do not add more than 10 ml of nitric acid at any given time, and allow any reaction to subside before proceeding further.

Connect the digestion vessel (7.2.1.1) to the heating device (7.2.1.4) and raise the temperature of the reaction mixture to reflux conditions and maintain for 2 h ensuring that the condensation zone is lower than 1/3 of the height of the reflux condenser, then allow to cool. Add the content of the absorption vessel to the reaction vessel via the reflux condenser, rinsing both the absorption vessel and condenser with further 10 ml of diluted nitric acid (6.4).

Transfer quantitatively the solution content of each vessel into a suitable sized volumetric flask and add water (6.1) to the volume mark.

Alternatively, another procedure can be applied, such that the adjustment to volume with the solid residue still present shall be carried out immediately after digestion.

If the measurement solution contains particles due to precipitation which may clog nebulizers or interfere with an injection of the sample into the instrument, the sample may be centrifuged, allowed to settle, or filtered (7.5).

The measurement solution is now ready for analysis for elements of interest using appropriate elemental analysis techniques.

8.3.2 Method A2 — Thermal heating with a heating block with containers

Weigh an amount of not more than 2 g of the test portion (typically 0,5 g to 1 g of dry sample) containing not more than 0,15 g of organic carbon with an accuracy of 0,001 g (or at least three significant figures) and transfer it into the digestion vessel (7.2.2.1).

The amount of the test sample depends on the amount of organic matter. The maximum amount of organic carbon shall not exceed 0,15 g when 8 ml of aqua regia is used. Per additional 0,1 g organic carbon (more than this 0,15 g), 1 ml of additional concentrated HNO₃ (6.3) shall be added before the digestion process is started.

N.B.: For some elements, e.g. barium and chromium, the additional volume of HNO₃ is essential in order to have a sufficient recovery upon digestion.

In case of dry samples moisten the test portion with a few drops of water (6.1). Add (6,0 ± 0,1) ml hydrochloric acid (6.2) followed by (2,0 ± 0,1) ml nitric acid (6.3). Let stand at room temperature until any effervescence almost ceases to allow for slow oxidation of the organic matter in the sample.

The time of standing at room temperature can have an influence on the digestion rate of aqua regia. For consistency, it is recommended to start heating as soon as possible after the first strong reaction has ceased.

Loosely screw on the tube cap (not very tight!) and place the digestion vessel on the heating block (7.2.2.2) and slowly increase the temperature to the boiling point. Keep the temperature on the boiling point during (120 ± 10) min.

Let the vessel cool down to room temperature and fill up with water (6.1) to the volume mark.

If a non-graduated digestion tube is used, transfer quantitatively the solution into a suitable sized volumetric flask and add water (6.1) to the volume mark. Alternatively, another procedure can be applied, such that the adjustment to volume with the solid residue still present shall be carried out immediately after digestion.

If the measurement solution contains particles due to precipitation which may clog nebulizers or interfere with an injection of the sample into the instrument, the sample may be centrifuged or filtered (7.5).

The measurement solution is now ready for analysis for elements of interest using appropriate elemental analysis techniques.

8.4 Method B — Microwave digestion with temperature control, closed vessels

Weigh an amount of not more than 2 g of the test portion (typically 0,5 g to 1 g of dry sample) containing not more than 0,15 g of organic carbon with an accuracy of 0,001 g (or at least three significant figures) and transfer it into the digestion vessel (7.3.1).

Add for each additional intake of 0,1 g organic carbon 1 ml of concentrated HNO₃ (6.3) before the digestion process is started.

Referring to the manufacturer's instructions, the upper limits of mass of the test portion shall be taken into account.

In case of dry samples moisten the test portion with a few drops of water (6.1). Add separately (6 ± 0,1) ml of hydrochloric acid (6.2) and (2 ± 0,1) ml of nitric acid (6.3) and mix well.

If a vigorous reaction occurs, allow the reaction cease before capping the vessel. If excessive foaming occurs, add a drop of anti-foaming agent (6.5).

This method is an operationally defined method, designed to achieve consistent digestion of samples by specific reaction conditions. The temperature of the digestion mixture in each vessel shall be raised with a heating rate of approximately 10 °C/min to 15 °C/min to (175 ± 5) °C and remain at (175 ± 5) °C for (10 ± 1) min. Cool down to room temperature.

Check this procedure regularly with a blank sample of aqua regia.

WARNING — Too high a temperature increase may cause a vigorous, exothermic reaction in the digestion solution with high pressure increase and blow-off of the security valve. Losses of analytes are possible.

At the end of the microwave programme, allow the vessels to cool according to the manufacturer's instructions before removing them from the microwave system. Cooling of the vessels may be accelerated by internal or external cooling devices.

After reaching room temperature, check if the microwave vessels maintained their seal throughout the digestion. Due to the wide variety of vessel designs, a single procedure is not appropriate. Carefully uncap and vent each vessel in a well-ventilated fume hood according to the manufacturer's instructions. Transfer quantitatively the solution content of each vessel into a suitable sized volumetric flask and add water (6.1) to the volume mark.

Alternatively, another procedure can be applied, such that the adjustment to volume with the solid residue still present shall be carried out immediately after digestion. If the measurement solution contains particles due to precipitation which may clog nebulizers or interfere with an injection of the sample into the instrument, the sample may be centrifuged, allowed to settle, or filtered.

The measurement solution is now ready for analysis for elements of interest using appropriate elemental analysis techniques.

9 Test report

This test report shall contain at least the following information:

- a) the digestion method used, together with a reference to this document, i.e. ISO 54321;
- b) identity of the sample;