Annex 1
Methods for Soil Analysis

Soil Analysis Method 1 (SA01)

Pre-treatment of Samples

Pre-treatment of Samples	
Method sheet	SA01
Reference methods	ISO 11464
Method suitable for	Organic Layer; Mineral Layer

I. Relevance in ICP Forests

All samples (organic and mineral) have to be prepared according to the standard methodology in order to maintain comparability between participating countries.

Priority	**Level I**	**Level II**
Organic Layer	Mandatory	Mandatory
Mineral layer	Mandatory	Mandatory

II. Principle

Organic layer

After removal of living material (such as mosses, roots, etc.) and objects > 2 cm, collected samples (preferably not less than 500 g fresh material) should be transported to the laboratory as soon as possible and should be air dried or dried at a temperature of 40 °C. They can then be stored until analysis. The sample is subsequently crushed or milled to size < 2 mm.

b. Mineral layer

After removal of living material (such as mosses, roots, etc.) and objects > 2 cm, collected samples (preferably not less than 500 g fresh soil) should be transported to the laboratory as soon as possible and should be air dried or dried at a temperature of 40 °C. They can then be stored until analysis.

Living macroscopic roots and all material, mineral and organic, with a diameter larger than 2 mm, should be removed from the samples by dry or wet sieving. The particles not passing the 2-mm sieve (after crushing), may be weighed separately for the determination of the coarse fragments content (SA05). The fraction smaller than 2 mm is used for the soil analysis. The mineral soil samples should not be milled. Only sieving above a 2 mm sieve is allowed. No further grinding will be allowed except for the analysis of Carbonate Content (SA07), Total Organic Carbon (SA08), total Nitrogen (SA09) and Total Elements (SA12).

The sample materials for storage should be kept without preservative under normal room conditions with minimal temperature and humidity fluctuations, shielded from incident light.

III. Apparatus

Drying oven.
Crusher, mill, mortar and pestle.
Plate sieve, mesh sieve

IV. Reagents

No reagents.

V. Procedure

Drying
Spread the material in a layer not thicker than 15 mm. If necessary, the sample is crushed while still damp and friable and again after drying. Dry the complete sample in drying oven at a temperature of 40 ° C, until the loss in mass of the sample is not greater than 5 % (m/m) per 24 h. Break down the size of larger clods (greater than 15 mm) to accelerate the drying process.

Removal of fraction < 2 mm
Remove stones and large objects by hand picking and sieving (< 2 mm). Minimise the amount of fine material adhered. Weigh separately the fraction not passing the 2 mm sieve for determination of coarse fragment content. Crush (not ground) the clods greater than 2 mm taking care that crushing of original particles is minimised. Homogenise the < 2 mm fraction.

Sieving and Milling
The organic sample is crushed or milled to size < 2 mm.

The mineral soil samples should not be milled. Only sieving above a 2 mm sieve is allowed. No further grinding will be allowed except for the analysis of Total Organic Carbon (SA08), Total Nitrogen (SA09) and Total Elements (SA12).

Subsampling
For the preparation of an analysis subsample, split up (by hand, using a sample divider or by mechanical subsampling) the sample into representative portions until the required sample number and sample size is obtained.

VI. Calculation

No calculations.

VII. Report

The mineral fractions (> 2 mm) obtained after sieving with a 2 mm sieve may be used for determination of coarse fragments (SA05).

VIII. Reference

ISO 11464. 1994. Soil Quality – Pretreatment of samples for physico-chemical analysis. International Organization for Standardization. Geneva, Switzerland. 9 p. (available at www.iso.ch)

Soil Analysis Method 2 (SA02)

Determination of Soil Moisture Content

Soil Moisture Content	
Method sheet	SA02
Reference methods	ISO 11465
Method suitable for	Organic Layer; Mineral Layer

I. Relevance in ICP Forests

Recalculation of results obtained by lab analysis to “oven-dry weight”.

Priority	**Level I**	**Level II**
Organic Layer	Mandatory	Mandatory
Mineral layer	Mandatory	Mandatory

II. Principle

Calculation and reporting of the results of soil analysis is done on basis of "oven-dry" soil. The moisture content of air-dry soil is determined prior to soil analysis. To recalculate the analysis results on dry weight basis, the moisture content of the sample has to be determined by oven-drying a sample to constant mass. The difference in mass is used to calculate water content on a mass basis.

III. Apparatus

Moisture tins or flasks (25 – 100 ml) with closely fitting lid
Drying oven
Analytical balance (accuracy 0.001 g)

Note: The use of an automated apparatus for measuring soil moisture content is allowed as long as it is based on the same principle.

IV. Reagents

No reagents.

V. Procedure

Mineral Layer: Transfer 5-15 g air-dried fine earth (fraction < 2 mm) to a dried, tared moisture tin and weigh. Dry at 105±5 °C (lid removed) until constant mass is reached.

Organic Layer : Transfer 5 – 10 g air dried organic layer material to a dried, tared moisture tin and weigh. Dry at 105 °C (lid removed) for 24 hours.

Remove tin from oven, close with lid, cool in desiccator and weigh.

VI. Calculation

The moisture content in weight percentage is obtained by :

$$Moist\% = \frac{A - B}{B - tare\ tin} * 100$$

Where:

A : Weight of tared moisture tin and air-dried soil sample
B : Weight of tared moisture tin and oven-dried soil sample

The corresponding correction factor for analytical results or for amount of sample to be weighed in for analysis is:

$$\textit{moisture correction factor(MCF)} = \frac{100 + \textit{moist}\%}{100}$$

Note: when reporting the results of Carbonate Content (SA07), Total Organic Carbon (SA08), Total Nitrogen (SA09), Exchangeable acidity, Free H^+, exchangeable elements (SA10), Aqua Regia Extractable elements (SA11), Total elements (SA12), Acid Oxalate Extractable Fe and Al (SA13), the results on air-dry basis should be multiplied by the moisture correction factor (MCF) to obtain the result on oven-dry basis.

VII. Report

Report moisture content (in %) with 1 decimal place.

VIII. Reference

ISO 11465. 1993. Soil Quality – Determination of dry matter and water content on a mass basis – Gravimetric method. International Organization for Standardization. Geneva, Switzerland. 3 p. (available at www.iso.ch)

Soil Analysis Method 3 (SA03)

Determination of Particle Size Distribution

Particle Size Distribution	
Method sheet	SA03
Reference methods	ISO 11277
Method suitable for	Mineral Layer

I. Relevance in ICP Forests

Particle Size Distribution : Clay Percentage

Priority	**Level I**	**Level II**
Organic Layer	-	-
Mineral layer	Mandatory[1, 2]	Mandatory[1]

[1] if not determined in the first soil survey
[2] an estimation of clay content based on finger test is allowed

Particle Size Distribution : Silt Percentage

Priority	**Level I**	**Level II**
Organic Layer	-	-
Mineral layer	Optional	Mandatory[1]

[1] if not determined in the first soil survey

Particle Size Distribution : Sand Percentage

Priority	**Level I**	**Level II**
Organic Layer	-	-
Mineral layer	Optional	Mandatory[1]

[1] if not determined in the first soil survey

Particle Size Distribution : USDA-FAO texture Classification

Priority	**Level I**	**Level II**
Organic Layer	-	-
Mineral layer	Mandatory[1, 2]	Mandatory[1]

[1] if not determined in the first soil survey
[2] an estimation of granulometry based on finger test is allowed

II. Principle

Separation of the mineral part of the soil into various size fractions and determination of the proportion of these fractions. The analysis includes all soil material, i.e. including gravel and coarser material, but the procedure below is applied to the fine earth (< 2 mm) only. Of paramount importance in this analysis is the pretreatment of the sample aimed at complete dispersion of the primary particles. Therefore, generally, cementing materials (usually of secondary origin) such as organic matter, salts, iron oxides and carbonates such as calcium carbonate are removed. After shaking with a dispersing agent, sand (63 µm-2 mm) is separated from clay and silt with a 63 µm sieve (wet sieving). The clay (< 2 µm) and silt (2-63 µm) fractions are determined by the pipette method (sedimentation).

III. Apparatus

Sampling pipette (10 to 50 ml) with safety bulb and water reservoir, held in frame
Constant temperature room or thermoregulated bath (20 – 30 °C ± 0.5 °C)
Glass sedimentation cylinders (approx. diam. 50 mm, approx. length 350 mm) graduated 500 ml volume with rubber bungs or stirrer)
Stirrer and rod
Glass weighing vessels (with masses known to 0.0001 g)

Mechanical shaker (30 – 60 revolutions/min)
Sieves (2 mm – 63 µm)
Balance (accuracy 0.0001 g)
Drying oven
Stopwatch (accuracy 1 s)
Glass filter funnel capable of holding the 63 µm sieve
Wash bottle
Desiccator
650 ml glass beaker with cover glass, 100 ml measuring cylinder, 25 ml pipette
Hot plate or bunsen burner
Electrical conductivity meter (accuracy 0.1 dS/m)
Optional: Centrifuge and 300 ml centrifuge bottle

IV. Reagents

Hydrogen peroxide (H_2O_2), 30% volume fraction.
Dispersing agent: 3.3 % sodium hexametaphosphate and 0.7 % soda solution:
Dissolve 33 g sodium hexametaphosphate $(NaPO_3)_6$ and 7 g soda (Na_2CO_3) in water in a 1 l volumetric flask and make to volume. Both chemicals should be dried overnight at 105 °C prior to use. This solution is unstable and shall be replaced after one month.
Antifoaming agent (preferably octan-2-ol, alternatives are ethanol or methanol)
Calcium chloride solution ($CaCl_2$), conc. 1 mol/l
Hydrochloric acid (HCl), conc. 1 mol/l

V. Procedure

Test sample
Depending on the soil type, weigh 10 (clay) to 30 g (sand) soil of the air-dried soil (fraction < 2 mm). Place the sample in the 650 ml glass beaker or 300 ml centrifuge bottle.

Destruction of organic matter
Add 30 ml water to the test sample (add if necessary a few drops of octan-2-ol to allow thoroughly wetting). Add 30 ml of the 30 % hydrogen peroxide solution and mix using the glass or plastic rod (add if necessary a few drops of octan-2-ol to control foaming). Cover and leave overnight. The next day, place the vessel on hot plate or bunsen burner and warm. Control foaming with octan-2-ol and stir frequently. To avoid drying out, add water if necessary. Bring the suspension to a gentle boil until all signs of bubbling due to the decomposition of hydrogen peroxide have ceased. If undecomposed organic material is still present, cool the beaker and repeat the treatment with hydrogen peroxide.
If using a centrifuge bring the volume to 150 – 200 ml by addition of water. Centrifuge the bottle until obtaining a clear supernatant (recommended 15 min at a minimum relative centrifugal force (RCF) of 400 g) and remove this supernatant by decanting or by using a suction device.
If a centrifuge is not available the mineral residues may be flocculated by adding 25 ml of 1 mol/l calcium chloride solution, stirring and bringing to about 250 ml with water. Let stand until the supernatant is clear, then siphon or decant this from the residue. Add another 250 ml of water and repeat the washing procedure until the dark residues of the decomposed organic matter have gone (if using this method, take care to check the electrical conductivity (next step) before adding the salt).

Removal of soluble salts and gypsum
After destruction of organic matter add water until obtaining a soil:water ratio of 1:4 – 1:6 (v:v). Shake for 1 h using shaking machine. Centrifuge to obtain a clear supernatant and measure electrical conductivity (E_c) on this supernatant. If E_c > 0.4 dS/m soluble salts and gypsum is present in considerable amounts and have to be removed. Remove the supernatant, add 250 ml water and shake for 1 h. Centrifuge and measure electrical conductivity again. Repeat this washing procedure until E_c < 0.4 dS/m.

Removal of carbonates

A distinction is made on basis of the presence or absence of calcium carbonate:

(1) Calcareous soils: $pH(H_2O) > 6.8$

(2) Non-calcareous soils: $pH(H_2O) < 6.8$

Where the carbonate content is greater than about 2 % mass fraction, add to the washed, centrifuged soil (above) 4 ml of 1 mol/l hydrochloric acid for each percent of carbonate, plus an excess of 25 ml of acid. Make up to about 250 ml with water, and place the suspension on the water bath at about 80 °C for 15 min, stirring the suspension from time to time. Leave the suspension to stand overnight. If the soil flocculates sufficiently to leave a perfectly clear supernatant, then this can be siphoned off or decanted, otherwise centrifugation and decantation will be necessary. Repeat the washing and decantation with water until the E_c of the supernatant is less than 0,4 dS/m.

If the carbonate content is less than about 2 % mass fraction, then only an initial 25 ml of 1 mol/l hydrochloric acid solution is required. It is recommended, therefore, that 20 ml of 1 mol/l calcium chloride solution is added at the same time as the acid. The rest of the procedure is identical as for a higher carbonate content.

Note: *if the carbonate content is that high that the results of the particle size distribution become unreliable, this should be mentioned in the Data Accompanying Report.*

Dispersion

Add sufficient water to the vessel so that the total volume is between 150 ml and 200 ml, shake the contents until all the soil is in suspension, and add 25 ml of dispersing agent from a pipette. Shake the bottle for 18 h on the end-over-end shaker.

Wet sieving at 63 µm

Place a 63 µm aperture sieve in the large glass funnel, and place the funnel in the stand so that the neck of the funnel is inside one of the 500 ml sedimentation tubes. Transfer the dispersed suspension from the centrifuge bottle quantitatively onto the sieve, and wash the soil using a jet of water from the wash-bottle until the water runs clear. The total volume of the
washings should not exceed 500 ml.

Remove the sieve from the funnel and wash the residue on the sieve into an evaporating dish by means of a gentle spray from the wash-bottle. To alleviate sieve blockage, use the glass or plastic rod and rubber sleeve. Place this dish in an oven between 105 °C and 110 °C until the residue is dry. Record the mass to 0.0001 g (mf_s).

Wash any particles adhering to the inside of the funnel into the sedimentation tube. Make up the suspension in the sedimentation tube to 500 ml with water.

Calibration

Calibration sampling pipette

Clean and dry the pipette thoroughly and immerse the tip in water. Draw water into the pipette into the safety bulb. Drain off the water in the safety bulb through the outlet tube. Drain the pipette into a weighing bottle of known mass, and determine the internal volume of the pipette. Repeat this exercise three times and take the average of the three volumes as the internal volume of the pipette to the nearest 0.05 ml (V_c ml).

Calibration dispersing agent

Pipette 25 ml of dispersing agent solution into one of the glass sedimentation tubes, and fill the tube to the 500 ml mark with water. Mix the contents of the tube thoroughly. Place the tube in the constant temperature environment, and leave the tube for at least 1 h. Between any of the times at which samples may be taken from the sampling tube (Table SA03-1), take a sample (V_c ml) of the dispersing agent solution from the sedimentation tube using the sampling pipette. Drain the pipette into a weighing vessel of known mass, and dry the contents of the vessel between 105 °C and 110 °C. Allow the vessel to cool in the desiccator and determine the mass of the residue in the vessel to 0.0001 g (m_r). Follow this procedure each time a new batch of dispersing agent is prepared.

Sedimentation

Place the sedimentation tube in the constant temperature environment. Agitate (at least 30 times/min for a minimum of 2 min) the contents of the sedimentation tube vigorously, either by means of the stirrer, or by inserting a bung in the tube, followed by end-over-end shaking. Replace the tube upright in the constant-temperature environment and start the timer.

About 15 s before a sample is to be taken (Table SA03-1), lower the pipette, with the tap of the safety bulb closed, vertically into the soil suspension, and centrally in the sedimentation tube, until the tip is the appropriate depth (± 1 mm) below the suspension surface (Table SA03-1). Take care to disturb the suspension as little as possible, and complete the operation within about 10 s. Open the tap of the safety bulb and withdraw a sample of the suspension such that the pipette and a part of the safety bulb are full. This sampling operation shall take about 10 s. Withdraw the pipette from the suspension so that the tip of the pipette is clear of the top of the sedimentation tube. Run the surplus present in the safety bulb into a small beaker by the outlet tube. Wash with water from the water reservoir until no suspension remains in this part of the system.

Place a weighing vessel of known mass (to 0.0001 g) under the tip of the pipette and open the tap so that the contents of the pipette are delivered to the vessel. Wash any suspension left on the inner walls of the pipette into the vessel by allowing water from the water reservoir to run through the system. Place the weighing vessel and contents in the oven between 105 °C and 110 °C, and evaporate to dryness. Cool the vessel in the desiccator, weigh the vessel and its contents to the nearest 0.0001 g, and determine the mass of the residue the nearest 0.0001 g (ms_1). Clean the outside of the pipette of any adhering sediment, and take the other sample (fraction < 2 µm), in accordance with the times given in Table SA03-1, using the same pipetting procedure given above. Call the additional sample masse ms_2.

Table SA03-1 Pipette sampling times and fraction at different temperatures

Temperature (°C)	Time (after mixing) of starting sampling operation	
	Fraction : < 63 µm Sampling depth 200 mm ± 1 mm	Fraction : < 2 µm Sampling depth 100 mm ± 1 mm
20	56 s	7 h 44 min 16 s
21	54 s	7 h 34 min 04 s
22	53 s	7 h 23 min 53 s
23	52 s	7 h 13 min 13 s
24	51 s	7 h 03 min 02 s
25	49 s	6 h 52 min 50 s
26	48 s	6 h 44 min 02 s
27	47 s	6 h 35 min 42 s
28	46 s	6 h 26 min 53 s
29	45 s	6 h 18 min 33 s
30	44 s	6 h 09 min 45 s

VI. Calculation

Fractions < 63 µm

Calculate the mass of solids in suspension in 500 ml (mf_1 , mf_2) in grams, for each pipette sampling time from the equation:

Mass < 63 µm in 500 ml : $mf_1 = ms_1 (500/V_c)$
Mass < 2 µm in 500 ml : $mf_2 = ms_2 (500/V_c)$

where:
mf_x is the mass (g) of solid in suspension in 500 ml;
ms_x is the mass (g) of material from the xth pipette sampling;
V_c is the calibrated volume of the pipette.

Each fraction however, still contains a part of dispersing agent, which has to be corrected. The mass of solid material in 500 ml of dispersant solution, m_d, in grams, is given by:

Mass dispersing agent in 500 ml : : $m_d = m_r\ (500/V_c)$

where:
m_r is the mass of residue, in grams;
V_c is the calibrated volume of the pipette, in millilitres.

This gives the final fraction masses:

Clay	Mass fraction < 2 µm	=	$mf_2 - m_d$
Silt	Mass fraction 2 – 63 µm	=	$mf_1 - mf_2$

Fraction 63 µm - 2 mm
Mass of the fraction 63 µm - 2 mm = mf_s

Proportion of fraction
The method of calculation assumes that the sample mass is the sum of the constituent fractions, and not the mass of the test sample. The mass of sample < 2 mm is thus the sum of the masses of the fractions obtained by wet sieving at 63 µm and the masses of the fractions obtained by calculation. Denote this total sample mass as m_t in grams.

Calculate the proportion in each fraction <2 mm as follows:
Proportions = mass of fraction/m_t

VII. Report

It is an agreed convention that the percentage of each particle size grade is reported on the basis of oven-dry (or air dry) soil free of organic matter (1 decimal place).

Note: With this calculation, the clay, silt and sand fractions are obtained in percentage of the sum of the constituent fractions (minus carbonate and organic matter which have been removed).

USDA-FAO texture classification is based on the USDA-FAO textural triangle (FAO, 1990) as shown in Figure 1.

VIII. References

ISO 11277. 1998. Soil Quality – Determination of particle size distribution in mineral soil material – Method by sieving and sedimentation. International Organization for Standardization. Geneva, Switzerland. 30 p. (available at www.iso.ch)

FAO. 1990. Guidelines for soil description, 3rd (revised) edition.

Soil Analysis Method 4 (SA04)

Determination of Bulk Density

Bulk Density	
Method sheet	SA04
Reference methods	ISO 11272
Method suitable for	Mineral Layer

I. Relevance in ICP Forests

Priority	**Level I**	**Level II**
Organic Layer	-	-
Mineral layer	Mandatory[1,2]	Mandatory[2]

[1] for Level I, bulk density may also be obtained by using pedo-transfer functions
[2] only mandatory in non-stony soils

II. Principle

The dry bulk density (BD) is the ratio between the mass of oven dry soil material and the volume of the undisturbed fresh sample. The ISO defines dry bulk density as the ratio of the oven-dry mass of the solids to the volume (the bulk volume includes the volume of the solids and of the pore space) of the soil.

Non-gravely soils (when coarse fragments content < 5%)

Several methods can be applied for the determination of bulk density, going from simple methods such as digging out holes of known volume to sophisticated gamma radiometry methods. The recommended method (core method) uses steel cylinders of known volume (100 cm^3, 400 cm^3) that are driven in the soil vertically or horizontally by percussion. Sampling large volumes results in smaller relative errors but requires heavy equipment. The method cannot be used if stones or large roots are present or when the soil is too dry or too hard.

Soils with high stone or root content or when the soil is too dry or too hard

In these conditions it is advised to use measuring methods based on the following principle (excavation method): a hole on a horizontal surface is dug and then filled with a material with a known density (e.g. sand which packs to a calibrated volume or water separated from the soil material by an elastic membrane). The obtained soil from the hole, is dried to remove the water and the dry mass is weighed. Methods measuring the volume of clods or aggregates should be avoided because they tend to underestimate macroporosity.
The volumetric percentage of the coarse fragments needs to be determined in order to calculate the bulk density of the fine earth.

Stony soils

Soils with a high content of coarse gravel (2 – 6 cm) and/or the presence of stones (6 – 20 cm) and boulders (> 20 cm), have a low volume of fine earth. Core samplers normally used in forest monitoring are not able to representatively collect stones or large portions of coarse fragments in the field. In these cases, the above recommended excavation method will produce good results but may be considered very expensive, time-consuming and destructive. So, alternatively, a combined approach is described where the quantity of bulk density of both fine earth and coarse fragments (SA05) has to be estimated / sampled in the field.

Methods are according to the prevailing conditions (i.e. coarse fragment content and size) at each individual sampling site:
- In case of coarse fragment content of more than 5 %, the fine earth fraction must be sieved and

weighed. Its volume must then be determined either directly or indirectly by establishing the coarse fragment volume. Furthermore, the density of the coarse fragments (specific weight) must be known or established.

- In case of content of coarse fragments > 20 mm, representative sampling is no longer possible with a core sampler. Then the coarse fragment content must be determined by additional sampling using a shovel or spade and/or estimations in the soil profile.
- In case of coarse fragments content of > 60 mm, representative volume sampling is not possible and sampling with mini-core samplers is combined with an estimation in the profile pit.

In the analysis each method or each combined method leads to the determination of (partially) different parameters which means that different calculation formulas are needed.

Note: The determination of the bulk density of the fine earth is incorrect when the sample contains significant portions of roots in addition to the coarse fragment portions. In these cases, this must be corrected

III. Apparatus

Core sample holders, thin-walled metal cylinders with a volume of 100 cm^3 to 400 cm^3, a steel cap for driving into the soil, and a driver
(or root auger, hollow stem auger, AMS core sampler with liner or alike)
Oven (heated and ventilated, temperature 105 ± 2 °C)
Desiccator
Balance (accuracy 1/1000 of measured value).
Spade, shovel
Metal sieves (2 mm, 20 mm, 60 mm)

IV. Reagents

No reagents.

V. Procedure

Case 1: Non-gravely soils (when coarse fragments content < 5%)

Press or drive a core sample holder of known volume without deflection and compaction into either a vertical or horizontal soil surface far enough to fill the sampler. Carefully remove the sample holder and its contents to preserve the natural structure, and trim the soil extending beyond each end of the sample holder with a straight-edged knife or sharp spatula. The soil sample volume is thus equal to the volume of the sample holder. Take at least five core samples from each soil layer. Place the holders containing the samples in an oven at 105 °C until constant mass is reached (minimum 48 h). Remove the samples from the oven and allow them to cool in the desiccator. Weigh the samples on the balance immediately after removal from the desiccator (m_t). Control mass is reached when the differences in successive weighings of the cooled sample, at intervals of 4 h, do not exceed 0,01% of the original mass of the sample.

Case 2: Mineral soil with a coarse fragment content of more than 5% that can be sampled with a core sampler or any other representative sampler (coarse fragments < 20 mm)

The mineral soil sample is collected in the field with core samplers from the undisturbed soil. In the laboratory the sample is then dried at a temperature of 105 °C for at least 48 hours to constant weight and weighed.
The sample is then passed through a 2 mm metal sieve and the sieve residue washed in order to break down clumpy fine earth material and to rinse off earth adhering to the stones. The washed sieve residue (= coarse fragment portion) is shaken into a beaker, dried at a temperature of 105°C in a drying oven and then weighed.

Case 3: Mineral soil, which cannot be sampled with a core sampler or any other representative sampler (coarse fragments > 20 mm)

Case 3.1.: Combination of representative volume sampling with a core sampler and estimation of coarse fragments > 20 mm

The mineral soil sample is collected in the field with core samplers from the undisturbed soil. In the laboratory the sample is dried at a temperature of 105 °C for at least 48 hours to constant weight and weighed.
The sample is passed through a 2 mm metal sieve and the sieve residue washed in order to break down clumpy fine earth material and to rinse off earth adhering to the stones. The washed sieve residue (= coarse fragment portion) is shaken into a beaker, dried at a temperature of 105 °C in a drying oven and then weighed. After that, the sieve residue is passed through a 20 mm sieve and the 2 – 20 mm sieve fraction (fine and medium gravel) weighed.
For the coarse fragment portion > 20 mm an estimation from the profile must be available.

Case 3.2.: Combination of representative volume sampling with a core sampler, disturbed sample and estimation of coarse fragments more than 60 mm at the profile

The mineral soil sample is collected in the field with a core sampler from the undisturbed soil. In addition, a larger sample volume, which must be representative for the coarse fragment fraction 2 – 60 mm (gravel), is collected with a shovel or a spade. In the laboratory the two samples are then dried at a temperature of 105 °C for at least 48 hours to constant weight and weighed.
The core sample is then passed through a 2 mm metal sieve and the sieve residue is washed in order to break down clumpy fine earth material and wash off earth adhering to the stones. The washed sieve residue (= coarse fragment portion) is shaken into a beaker, dried in a drying oven at a temperature of 105 °C and then weighed.
The spade sample is also dried at a temperature of 105 °C to constant weight and then weighed. The spade sample is then passed through a 2 mm sieve and the sieve residue through a 60 mm sieve. The coarse fragment fraction 2 – 60 mm obtained in this way is weighed. For the coarse fragment content > 60 mm an estimation from the profile pit must be available.

Case 4: Representative volume sampling not possible, Sampling with mini-core samplers

With core sampler caps or mini-core samplers ($n \geq 5$) several samples are taken from the undisturbed soil. In addition, a larger sample volume, which must be representative for the coarse fragment fraction 2 – 60 mm, is collected with a shovel or a spade.
In the laboratory the core sampler caps together with their contents are dried at a temperature of 105 °C for at least 48 hours to constant weight and then weighed together. The empty weight of the core sampler caps is then deducted from the total weight.
The spade sample is dried at a temperature of 105 °C for at least 48 hours to constant weight and then weighed. The sample is then passed through a 2 mm sieve and the sieve residue through a 6 mm sieve. The sieve residue is then passed through a 60 mm sieve as well. The fractions obtained < 2 mm (fine earth), 2 – 6 mm (fine gravel) and 6 – 60 mm (medium and coarse gravel) are weighed.

Alternative to the combined approach of case 2 till case 4 in soils with high stone or root content or if the soil is too dry or too hard
In case of gravely or stony soils an alternative excavation method consist of excavating a quantity of soil, drying and weighing it, and determining the volume of the excavation by filling it with sand (cf. ISO 11272 – **excavation method**). Note that the excavation method measures the total dry bulk density

VI. Calculation

Case 1: Non-gravely soils (when coarse fragments content < 5%)

In case of measurements, the bulk density of the fine earth (BD_{fe}) is approximately equal to the bulk density of total soil. The bulk density (BD_s) the for *non-gravely soils* is calculated as follows:

$$BD_s = BD_{fe} = \frac{M_s}{V_s} \qquad \textit{(equation SA04.01)}$$

where:

BD_s = Bulk Density of the sample (kg/m^3)
BD_{fe} = Bulk Density of the fine earth (kg/m^3)
M_s = Mass of the sample (kg)
V_s = Volume of the sample (m^3)

Case 2: Mineral soil with a coarse fragment content of more than 5% that can be sampled with a core sampler or any other representative sampler (coarse fragments < 20 mm)

In case of measurement with a core sampler, the bulk density of the fine earth of gravely soils (BD_{fe}) is calculated as follows:

$$BD_{fe} = \frac{M_{fe}}{V_{fe}} = \frac{M_s - M_{cf}}{V_s - V_{cf}} = \frac{M_s - M_{cf}}{V_s - \frac{M_{cf}}{\rho_{cf}}} \qquad \textit{(equation SA04.02)}$$

where:

BD_{fe} = Bulk density of the fine earth (kg/m^3)
M_{fe} = Mass of the fine earth taken with core sampler (kg)
V_{fe} = Volume of the undisturbed fine earth (m^3)
M_s = Mass of the soil sample with gravel taken with core sampler (kg)
M_{cf} = Mass of the coarse fragments taken with the core sampler (kg)
V_s = Volume of core sampler (m^3)
V_{cf} = Volume of the coarse fragments taken with the core sampler (kg)
ρ_{cf} = Density of the coarse fragments (approximated by 2650 kg/m^3)

The fine earth stock (FES) is the amount (kg) of fine earth in the soil layer under consideration expressed per ha. In stony soils, a correction for the volume of coarse fragments is required. It is calculated as follows:

$$FES = BD_{fe} \times d \times 10 \times \left(1 - \frac{V_{cf}}{V_s}\right) = BD_{fe} \times d \times 10 \times \left(1 - \frac{M_{cf}}{\rho_{cf} \times V_s}\right) \qquad \textit{(equation SA04.03)}$$

where:

FES = Fine earth stock (t/ha)
BD_{fe} = Bulk density of fine earth (kg/m^3)
d = Thickness of the sampled layer (m)
V_{cf} = Volume of coarse fragment taken with core sampler (respectively core of root auger) (m^3)
M_{cf} = Mass of coarse fragment taken with core sampler (respectively core of root auger) (kg)

ρ_{cf} = Density of the coarse fragments (approximated by 2650 kg/m^3)
V_s = Volume of core sampler (m^3)

Notes:
- *If the core sampler sample cakes strongly as a consequence of drying, it might make sense to pulverise the sample with a crusher prior to sieving. The big stones should be removed beforehand. .*
- *In the case of non-cohesive soil (sand), there is no need to wash or dry the stones.*

Case 3: Mineral soil, which cannot be sampled with a core sampler or any other representative sampler (coarse fragments > 20 mm)

Case 3.1. Combination of representative volume sampling with a core sampler and estimation of coarse fragments > 20 mm

The bulk density of the fine earth (BD_{fe}) is calculated using equation SA04.02.

The FES is calculated as follows:

$$FES = BD_{fe} \times d \times 10 \times \left(1 - \frac{V_{cf>20}}{100} - \frac{M_{cf(2-20)}}{\rho_{cf} \times V_s} \right)$$ *(equation SA04.04)*

where:
FES = Fine earth stock (t/ha)
BD_{fe} = Bulk density of fine earth (kg/m^3)
d = Thickness of the sampled layer (m)
$M_{cf(2-20)}$ = Mass of coarse fragment between 2 and 20 mm taken with core sampler (respectively core of root auger) (kg)
$V_{cf>20}$ = Percentage volume of coarse fragment of the fraction > 20 mm estimated at the profile (%)
ρ_{cf} = Density of the coarse fragments (approximated by 2650 kg/m^3)
V_s = Volume of core sampler (m^3)

Notes: see Case 2

Case 3.2. Combination of representative volume sampling with a core sampler, disturbed sample and estimation of coarse fragments more than 60 mm at the profile

The bulk density of the fine earth (BD_{fe}) is calculated using equation SA04.02.

The fine earth stock (FES) is calculated as follows:

$$FES = BD_{fe} \times d \times 10 \times \left(1 - \frac{V_{cf>60}}{100} - \frac{M_{ds(2-60)}}{BD_{cf}} \times \frac{BD_{fe}}{M_{ds} - M_{ds(2-60)} + BD_{fe} \times \frac{M_{ds(2-60)}}{\rho_{cf}}} \right)$$

(equation SA04.05)

where:
FES = fine earth stock (t/ha)

BD_{fe} = bulk density of fine earth (kg/m^3)
d = thickness of the sampled layer (m)
$M_{ds(2-60)}$ = mass of coarse fragment between 2 and 60 mm of the disturbed sample (kg)
$V_{cf>60}$ = percentage volume of coarse fragment > 60 mm estimated at the profile (%)
ρ_{cf} = bulk density of the coarse fragments (approximated by 2650 kg/m^3)
M_{ds} = total mass of the disturbed sample (kg)

Notes: see Case 2

Case 4: Representative volume sampling not possible, Sampling with mini-core samplers

From the weight of the sample < 6 mm and the weight of the coarse fragment fraction 2 mm – 6 mm, factor f, which is approximately the coarse fragment portion in the core sampler cap, is calculated as follows:

$$f = \frac{M_{ds(2-6)}}{M_{ds(<6)}} \quad \textit{(equation SA04.06)}$$

where:
$M_{ds(2-6)}$ = mass of coarse fragment of the fraction 2 - 6 mm of the disturbed sample (kg)
$M_{ds(<6)}$ = mass of the sample < 6 mm in the aliquot of the disturbed sample (kg)

For the coarse fragment content > 60 mm an estimation from the profile must be available.

The bulk density of the fine earth (BD_{fe}) is calculated using the following formula:

$$BD_{fe} = \frac{M_{TOT}MINI \times (1 - f)}{V_{TOT}MINI - \frac{M_{TOT}MINI \times f}{\rho_{cf}}} \quad \textit{(equation SA04.07)}$$

where:
M_{TOT}MINI = Mass of mini-core sampler (kg)
V_{TOT}MINI = Volume of mini-core sampler (m^3)
ρ_{cf} = Density of the coarse fragments (kg/m^3) (approximated by 2650 kg/m^3)

The fine earth stock (FES) is calculated using equation SA04.05.

VII. Report

The dry bulk density (BD) is recorded in kg · m^{-3} (kg/m^3) with no decimal places.

In the case of stony or gravely soils the bulk density of the fine earth fraction (< 2 mm) should be reported together with the coarse fragment content (vol %) (See also SA05).

Furthermore, the bulk density of the coarse fragments should be known, but this may be approximated as 2650 kg.m^{-3}. In the case that pedotransfer functions are used (Level I), the calculation procedure should be reported as well.

Note that the "excavation method" described in ISO11272, asks for the total dry bulk density of the soil, while in this programme the bulk density of the fine earth should be reported.

VIII. Reference

ISO 11272. 1993. Soil Quality – Determination of dry bulk density. International Organization for Standardization. Geneva, Switzerland. 10 p. (available at www.iso.ch)

DIN ISO 11272, Normenausschuß Wasserwesen (NAW) in the Dt. Inst. für Normung e.V. [Eds.] (2001): Bodenbeschaffenheit - Bestimmung der Trockenrohdichte (Soil composition, Determination of bulk density)

W. Riek, B. Wolff (2006): Evaluierung von Verfahren zur Erfassung des Grobbodenanteils von Waldböden – Erarbeitung von Empfehlungen für die Anwendung dieser Verfahren im Rahmen der Bodenzustandserhebung im Wald (BZE II)“. Eberswalde (Evaluation of methods to determine the coarse fragment portion of forest soils – Drawing up recommendations for the use of these methods in forest soil surveys)

Soil Analysis Method 5 (SA05)

Determination of Coarse Fragments

Coarse Fragments	
Method sheet	SA05
Reference methods	ISO 11464, ISO 11277
Method suitable for	Mineral horizons

I. Relevance in ICP Forests

Priority	**Level I**	**Level II**
Organic Layer	-	-
Mineral layer	Mandatory[1]	Mandatory

[1] for Level I, coarse fragments content may also be obtained by estimation

II. Principle

The abundance of coarse fragments can be measured in the laboratory, but is usually estimated during routine soil profile description (see Mikkelsen et al., 2006). When the estimation is based on such a visual observation, one should take into account the volume of the macropores (packing pores between the stones) which is often underestimated.

The most straightforward way to determine the volumes in the field of stones and boulders is by digging pits. This method, however, encounters practical problems such as hard manual work and destructive sampling. The 'Finnish method' or 'rod penetration method' is described here as an example of a non-destructive method. This method estimates the proportion (*volume %)* of coarse gravel, stones and boulders (coarse fragments > 2 cm) in the 0 – 30 cm mineral layer by pushing a graduated metal rod down through the organic layer and as far as possible into the mineral soil.

Coarse fragments are separated from the fine earth fraction during the preparation of soil samples (SA01). The content of coarse fragments, *cf. (weight %),* is determined by weighing the residue left on a 2 mm sieve after washing and drying in the laboratory.

III. Apparatus

Field estimation: The 'Finnish method' or 'rod penetration method'
graduated metal rod (diameter 10 mm, length 80 – 100 cm)

Laboratory measurement
No apparatus, using data obtained in preparation of soil sample (SA01).

IV. Reagents

Field estimation: The 'Finnish method' or 'rod penetration method'
No reagents.

Laboratory measurement
No reagents, using data obtained in preparation of soil sample (SA01).

V. Procedure

Field estimation: The 'Finnish method' or 'rod penetration method'

The volume of stones is estimated in the 0-30 cm mineral soil layer. A steel rod (d = 10 mm, length = 80…100 cm, with a tip of hard metal, gradation lines at 10 cm intervals, see Fig. 1) is pushed down

(through the organic layer) into the mineral soil with sufficient force that the rod will stop if it comes into contact with a stone of 2 cm or larger (moderate push). The measuring rod is pushed down into the mineral soil at e.g. 20 or 30 systematically located (using a tape measure or even paces) points. The depth of penetration is measured with respect to the surface of the ground. If there is an organic layer present, then its thickness has to be measured using the rod or by taking a sample of the organic layer and measuring its thickness, and then subtracted from the penetration depth. In Finland, penetration is measured and organic layer samples are taken at the same time. The average penetration value and stoniness of the 0-30 cm mineral soil layer is calculated as follows (only 5 points in this example):

Penetration depth (cm)	**Organic layer thickness (cm)**	**Penetration depth – organic layer thickness (cm)**	**Penetration in the ≤30 layer (cm)**
12	2	10	10
40	4	36	30
4	4	0	0
35	3	32	30
22	5	17	17
			Average = 17.4

The great advantage of the rod method is that a large number of measurements can be made easily and quickly over the whole plot. The inaccuracy and other drawbacks of the method outweigh the lack of representability involved in measuring (estimating) stoniness in a very restricted number of soil pits.

Figure 1: Penetration rod

Figure 2: Tip of the penetration rod

Laboratory measurement

No procedure, using data obtained in preparation of soil sample (SA01).

VI. Calculation

Field estimation

0 – 30 cm layer

*Volume of stones (%) = 83 - 2.75 * average penetration (cm)* *[Equation SA05.01]*

The volume of stones in the example = 83 - 2.75 * 17.4 = 35 % in the 0-30 cm layer. According to equation SA05.01, the volume of stones is 0.5 % when the average penetration into the mineral soil is 30 cm, and volume of stones is 83 % when the average penetration is 0 cm.

It is possible to estimate the stoniness of thinner layers if the empirical relationship between penetration depth and volumetric stone percentage remains the same. The relevant equations are as follows:

0-10 cm layer

*Volume of stones (%) = 83 - 8.25 * average penetration (cm) for the layer*

0-20 cm layer

*Volume of stones (%) = 83 - 4.125 * average penetration (cm) for the layer.*

The constant maximum depth of each penetration should be set so that it reaches the target mineral soil depth, i.e. 30, 20 or 10 cm, through the thickest possible organic layer. On upland soils an extra 10 cm is commonly added to the target depth, i.e. there is a target depth of 40 cm if the studied layer is 0-30 cm, or to 30 cm if the layer is 0-20 cm.

Note: *Equation SA05.01 is based on a very limited material [Finnish till (morainic) soils] and it has not been tested on other soils, and in some respects it is somewhat illogical (see Eriksson and Holmgren, 1996). It is therefore of utmost importance that the equation is calibrated locally before it can be applied on other soil types..*

Laboratory measurement

The content of coarse fragments, *cf (weight%),* is determined by weighing the residue left on a 2 mm sieve after washing and drying according to:

$$cf(weight\%) = \frac{soil\ \ fraction > \ 2\ mm}{weight\ of\ the\ total\ oven\ dry\ soil} \cdot \times 100$$

In order to convert the content by weight to an expression by volume, the bulk density of both the coarse fragments and the fine earth should be determined.

$$cf\,(vol\%) = \frac{\mathrm{BD_s}}{\mathrm{BD_{cf}}} \cdot cf\,(weight\%)$$

where:

BD_s = Bulk density of the total soil (kg/m^3)
BD_{cf} = Bulk density of the coarse fragments (approximated by 2650 kg/m^3)
cf(vol%) = Volumetric percentage of coarse fragments in the soil (%)
cf(weight%) = Weight percentage of coarse fragments in the soil (%)

VII. Report

The amount of coarse fragments (stones and gravel with a diameter> 2 mm) has to be reported for the individual mineral layers in volume %.

Note: *The Rod penetration method only allows reporting for the 0 – 10 cm, 0 – 20 cm or 0- 30 cm layer and for the coarse fragments > 2 cm*

VIII. References

Eriksson, C.P., Holmgren, P. 1996. Estimating stone and boulder content in forest soils – evaluating the potential of surface penetration methods. Catena 28: 121 – 134.

ISO 11464. 1994. Soil Quality – Pretreatment of samples for physico-chemical analysis. International Organization for Standardization. Geneva, Switzerland. 9 p. (available at www.iso.ch)

ISO 11277. 1998. Soil Quality – Determination of particle size distribution in mineral soil material – Method by sieving and sedimentation. International Organization for Standardization. Geneva, Switzerland. 30 p. (available at www.iso.ch)

Mikkelsen, J. Cools, N., Langohr, R. 2006 Guidelines for Forest Soil Profile Description, adapted for optimal field observations within the framework of the EU Forest Focus Demonstration Project. BIOSOIL. Partly based on the 4th edition of the Guidelines for Soil Profile Description and Classification (FAO, In Press).

Tamminen, P. 1991. Kangasmaan ravinntunnusten ilmaiseminen ja viljavuuden alueellinen vaihtelu Etelä-Suomessa. Summary: Expression of soil nutrient status and regional variation in soil fertility of forested sites in Southern Finland. Folia Forestalia 777: 1-40.

Viro, P., 1947. Metsämaan raekoostumus ja viljavuus varsinkin maan kivisyyttä silmällä pitäen. Summary: The mechanical composition and fertility of forest soil taking into consideration especially the stoniness of the soil. Communicationes Instituti Forestalis Fenniae 35, 115.

- 1952. Kivisyyden määrittämisestä. Summary: On the determination of stoniness. Communicationes Instituti Forestalis Fenniae 40, 23.
- 1958. Suomen metsämaiden kivisyydestä. Summary: Stoniness of forest soil in Finland. Communicationes Instituti Forestalis Fenniae 49, 45

Soil Analysis Method 6 (SA06)

Determination of Soil pH

pH	
Method sheet	SA06
Reference methods	ISO 10390
Method suitable for	Organic Layer; Mineral Layer

I. Relevance in ICP Forests

$pH(CaCl_2)$

Priority	**Level I**	**Level II**
Organic Layer	Mandatory	Mandatory
Mineral layer	Mandatory	Mandatory

$pH(H_2O)$

Priority	**Level I**	**Level II**
Organic Layer	Optional	Optional
Mineral layer	Optional	Optional

II. Principle

The pH of the soil is potentiometrically measured in the supernatant suspension of a 1:5 soil : liquid (v/v) mixture. This liquid is made up of a 0.01 mol/l solution of calcium chloride in water $pH(CaCl_2)$ or water $pH(H_2O)$.

III. Apparatus

End-over-end shaking machine
pH meter with appropriate electrode
Thermometer (accuracy 1 °C)
Sample bottle (capacity at least 50 ml) with cap
Spoon

IV. Reagents

Water (grade 2)
Calcium chloride ($CaCl_2$), conc. 0.01 mol/l
make a solution of 1.47 g $CaCl2.2H_2O$/liter water
pH buffer solutions

V. Procedure

Preparation of the suspension
Take a representative sample (at least a volume of 5 ml) of the air-dried soil (fraction < 2 mm). Place the test sample in the sample bottle and add five times its volume of calcium chloride solution (pH-$CaCl_2$) or water (pH-H_2O). Shake or mix the suspension vigorously, for 5 min, using the mechanical shaker or mixer, and wait for 2 h.

Calibration of pH meter
Calibrate the pH-meter as prescribed in the manufacturer' s manual, using the buffer solutions.

pH measurement
Adjust the pH-meter as indicated in the manufacturer' s manual. Measure the temperature of the suspension and take care that the temperature of the buffer solution and the soil suspension does not

differ by more than 1 °C. Shake the suspension thoroughly just before measurement of the pH. Measure the pH in the settling suspension. Read the pH after stabilisation is reached.

VI. Calculations

No calculations.

VII. Report

Note the recorded values to one decimal place.

VIII. Reference

ISO 10390. 1994. Soil Quality – Determination of pH. International Organization for Standardization. Geneva, Switzerland. 5 p. (available at www.iso.ch)

Soil Analysis Method 7 (SA07)

Determination of Carbonate Content

Carbonates	
Method sheet	SA07
Reference methods	ISO 10693
Method suitable for	Organic Layer, Mineral Layer

I. Relevance in ICP Forests

Priority	**Level I**	**Level II**
Organic Layer	Mandatory (if pH-$CaCl_2$ > 5.5)	Mandatory (if pH-$CaCl_2$ > 5.5)
Mineral layer	Mandatory (if pH-$CaCl_2$ > 6)	Mandatory (if pH-$CaCl_2$ > 6)

II. Principle

The soil sample is treated with a strong acid. The volume of the carbon dioxide produced is measured by using a calcimeter (Scheibler unit), and is compared with the volume of carbon dioxide produced by pure calcium carbonate.

III. Apparatus

Calcimeter (Scheibler unit)
Analytical balance (accuracy 0.0001 g)
Reaction vessels (capacity 150 ml)
Plastic cups (which can pass through the neck of the reaction vessel)
Tong
Watch glass

IV. Reagents

Distilled water
Hydrochloric acid (HCl), conc. 4 mol/l
Dilute 340 ml of concentrated hydrochloric acid (ρ = 1 ,I 9 g/ml) to 1000 ml with water.
Calcium carbonate ($CaCO_3$), pure.

V. Procedure

Preparation
The mass of the test portion is determined based on the carbonate content. For a preliminary test on carbonate content, add some hydrochloric acid to a portion of the soil on a watch glass. The carbonate content of the sample can be estimated on the basis of the intensity and duration of effervescence (Table SA07-1). Determine from table SA07-1 the mass of test portion (air-dried soil fraction < 2 mm).

Table SA07-1 Mass of test portion for determination of carbonate content based on intensity of effervescence

Intensity of effervescence	**Carbonate content (g/kg)**	**Mass of test sample (g)**
None or only limited	< 20	10
Clear, but for a short time	20 – 80	5
Strong, for a long time	80 – 160	2.5
Very strong, for a long time	> 160	≤1[1]

[1] use sample that is crushed to a particle size of less than 250 µm

Measurement

Transfer the sample into the reaction vessels and add 20 ml of water. Fill the plastic cup with 7 ml of hydrochloric acid and place this, using tongs in the reaction vessel containing the test portion. Take care that there is no contact between the hydrochloric acid and the soil before the reaction vessel is connected to the calcimeter (Scheibler unit). Warm the reaction vessel by hand.

Connect the reaction vessel to the calcimeter. Carefully add the hydrochloric acid from the cup to the soil by tilting the reaction vessel at an angle. The gas produced will lower the water level in the tube on the right and at the same time will raise the water level in the tube on the left. Shake for 5 min and note the volume when it no longer varies. If it still varies, continue shaking until the volume is stable, but not longer than 1 h. At the end of the shaking period, bring the water level in both tubes to the same height and measure the volume of gas in the calibrated tube with an accuracy of 0.1 ml.

Calibration

Determinations of samples, blanks and the calcium carbonate used as standard material, shall be performed simultaneously in a room where temperature and pressure do not vary too much during the measurement.

Weigh the standards of 0.200 g and 0.400 g of calcium carbonate, transfer these amounts into the reaction vessels and add 20 ml of water. For the blank determinations, use reaction vessels containing 20 ml of water.

VI. Calculations

$$w(CaCO_3) = 1000 \times \frac{m_2 (V_1 - V_3)}{m_1 (V_2 - V_3)}$$

$w(CaCO_3)$ = carbonate content of sample (g/kg) on basis of air dried soil
m_1 = mass (g) of test sample
m_2 = mean mass (g) of standards
V_1 = volume (ml) of CO_2 produced by test sample
V_2 = mean volume (ml) of CO_2 produced by standards
V_3 = volume change (ml) in blank determinations (can be negative)

VII. Report

The results of the carbonate (g/kg) must be reported without decimals on the basis of oven-dried soil.

VIII. Reference

ISO 10693. 1994. Soil Quality – Determination of carbonate content - Volumetric method. International Organization for Standardization. Geneva, Switzerland. 7 p. (available at www.iso.ch)

Soil Analysis Method 8 (SA08)

Determination of Organic Carbon Content

Organic Carbon	
Method sheet	SA08
Reference methods	ISO 10694
Method suitable for	Organic Layer, Mineral Layer

I. Relevance in ICP Forests

Priority	**Level I**	**Level II**
Organic Layer	Mandatory	Mandatory
Mineral layer	Mandatory	Mandatory

II. Principle

The carbon present in the soil is oxidised to carbon dioxide (CO_2) by heating the soil to at least 900 °C in a flow of oxygen-containing gas that is free from carbon dioxide. The amount of carbon dioxide released is then measured by titrimetry, gravimetry, conductometry, gas chromatography or using an infrared detection method, depending on the apparatus used.
When the soil is heated to a temperature of at least 900 °C, any carbonates present are completely decomposed.

Total organic carbon can be determined directly or indirectly. Direct determination consists of previous removal of any carbonates present by treating the soil with hydrochloric acid. Indirect determination consists of a correction of the total carbon content for the carbonates present.

III. Apparatus

Glassware
Analytical balance (accuracy 0.0001 or 0.00001 g)
Apparatus for determination of total carbon content (temperature at least 900 °C)
Crucibles proper for the apparatus

IV. Reagents

Combustion gas - chemicals and catalysts proper to the apparatus
Calibration substances
Hydrochloric acid (HCl), conc. 4 mol/l

V. Procedure

Laboratory sample
Use sample of air-dried soil (fraction < 2 mm) of known moisture and carbonate content.

Calibration of the apparatus
Calibrate the apparatus as described in the relevant manual using the calibration substances.

Direct determination of organic carbon content
Add an excess of hydrochloric acid (4 mol/l) to the crucible containing a weighed quantity of air-dried soil and mix. Wait 4 h and dry the crucible for 16 h at a temperature of 60 °C to 70 °C. The amount of test portion taken for analysis depends on the expected carbon content and on the apparatus used. Weigh out m_1 g of the air-dried sample in a crucible. Carry out the analyses in accordance with the manufacturer's manual for the apparatus.

Indirect determination of organic carbon content
The procedure is identical to the direct determination of organic carbon content, without adding hydrochloric acid. The measured total carbon content is calculated according to the amount of test portion taken for analysis which depends on the expected total carbon content and on the apparatus used. Weigh out m_1 g of the air-dried sample in a crucible. Carry out the analyses in accordance with the manufacturer's manual for the apparatus.

VI. Calculation

Direct determination of organic carbon content
The organic carbon content (on basis of air-dried soil) is obtained by :

$$w_{C,o} = 1000 \times \frac{m_2}{m_1} \times 0.2727$$

where

$w_{C,o}$	=	Organic carbon content (g/kg) on basis of air-dried soil
m_1	=	Mass (g) of test portion
m_2	=	Mass (g) of released CO_2
0.2727	=	Conversion factor for CO_2 to C

Indirect determination of organic carbon content
The total carbon content (on basis of air-dried soil) is obtained by :

$$w_{C,t} = 1000 \times \frac{m_2}{m_1} \times 0.2727$$

where

$w_{C,t}$	=	Total carbon content (g/kg) on basis of air-dried soil
m_1	=	Mass (g) of test portion
m_2	=	Mass (g) of released CO_2
0.2727	=	Conversion factor for CO_2 to C

Calculate the organic carbon content of the sample using a correction for carbonates. The organic carbon content (on basis of air dried soil) is calculated by:

$$w_{C,o} = w_{C,t} - (0.12 \times w_{CaCO_3})$$

where

$w_{C,o}$	=	Organic carbon content (g/kg) on basis of air-dried soil
$w_{C,t}$	=	Total carbon content (g/kg) on basis of air-dried soil
0.12	=	Conversion factor
w_{CaCO3}	=	Carbonate content (g/kg) on basis of air-dried soil

VII. Report

Report organic carbon content (in g/kg) with 1 decimal place on the basis of oven-dried soil.

VIII. Reference

ISO 10694. 1995. Soil Quality – Determination of organic and total carbon after dry combustion (elementary analysis). International Organization for Standardization. Geneva, Switzerland. 7 p. (available at www.iso.ch)

Soil Analysis Method 9 (SA09)

Determination of Total Nitrogen Content

Total Nitrogen	
Method sheet	SA09A
Reference methods	ISO 13878
Method suitable for	Organic Layer, Mineral Layer

I. Relevance in ICP Forests

Priority	**Level I**	**Level II**
Organic Layer	Mandatory	Mandatory
Mineral layer	Mandatory	Mandatory

II. Principle

The nitrogen content of a soil is determined by heating to a temperature of at least 900 °C in the presence of oxygen gas. Mineral and organic nitrogen compounds are oxidised and/or volatilised. The combustion products are oxides of nitrogen (NO_x) and molecular nitrogen (N_2). After transforming all nitrogen forms into N_2, the content of total nitrogen is measured using thermal conductivity.

III. Apparatus

Laboratory glassware
Analytical balance (accuracy 0.0001 or 0.00001 mg)
Apparatus for determination of total nitrogen content (temperature at least 900 °C)
Crucibles proper for the apparatus

IV. Reagents

Combustion gas - chemicals and catalysts proper to the apparatus
Calibration substances

V. Procedure

Laboratory sample
Use fraction of air-dried soil (fraction < 2 mm) of known moisture content. If a soil mass of less than 2 g is required for nitrogen analysis, mill a representative subsample further, to pass a sieve of an aperture specified in the manufacturer's manual to ensure sufficient test reproducibility.

Calibration of the apparatus
Calibrate the apparatus as described in the relevant manual using the calibration substances.

Determination of total nitrogen content
The amount of test sample for analysis depends on the expected total nitrogen content and on the apparatus used. Weigh out m_1 g of the air-dried sample or subsample into a crucible. Carry out the analyses in accordance with the manufacturer's manual for the apparatus.
Normally the primary results are given as milligrams nitrogen (X_1) or a mass fraction of nitrogen (X_2), expressed as a percentage, referred to the mass of air-dry soil used (m_1).

VI. Calculation

Calculate the total content of nitrogen (wN), in milligrams per gram, on the basis of the air-dried soil according to the following equations:

- For primary results given in milligrams of nitrogen:

$$w_N = \frac{X_1}{m_1}$$

- For primary results, given as percent mass fraction of nitrogen:

$$w_N = 10.X_2$$

where

w_N : total nitrogen content (g/kg) on basis of air-dried soil
m_1 : mass (g) of test portion
X_1 : primary result as milligrams N
X_2 : primary result as percentage N

VII. Report

Report total nitrogen (in g/kg) with 1 decimal place on the basis of oven-dried soil.

VIII. Reference

ISO 13878. 1998. Soil Quality – Determination of total nitrogen content by dry combustion ("elemental analysis"). International Organization for Standardization. Geneva, Switzerland. 5 p. (available at www.iso.ch)

Total Nitrogen Modified Kjeldahl method	
Method sheet	SA09B
Reference methods	ISO 11261
Method suitable for	Organic Layer, Mineral Layer

I. Relevance in ICP Forests

Priority	Level I	Level II
Organic Layer	Mandatory	Mandatory
Mineral layer	Mandatory	Mandatory

II. Principle

The modified Kjeldahl method determines the total nitrogen content (including ammonium-N, nitrate-N, nitrite-N and organic N) of a soil. The method is based on a Kjeldahl digestion, but instead of selenium (Kjeldahl method) titanium dioxide is used as the catalyst.

III. Apparatus

Digestion flasks or tubes (50 ml)

Digestion stand

Distillation apparatus

Burette (intervals of 0.01ml or smaller)

IV. Reagents

- Salicylic acid / Sulfuric acid: Dissolve 25g of salicylic acid in 1 litre of concentrated sulfuric acid (ρ = 1.84 g/cm^3)
- Potassium sulfate catalyst mixture: Grind and thoroughly mix the following substances;
  - 200 g of potassium sulfate
  - 6 g of copper (II) sulfate pentahydrate
  - 6 g of titanium dioxide with the crystal structure of anatase
- Sodium thiosulfate pentahydrate: Crush the crystals of Sodium thiosulfate pentahydrate until they form a powder that passes through a sieve with an aperture of 0.25mm
- Sodium hydroxide: c(NaOH) = 10 mol/l
- Boric acid solution: $\rho(H_3BO_3)$ = 20g/l
- Mixed indicator: Dissolve 0.1g of bromocresol green and 0.02 g of methyl red in 100 ml of ethanol
- Sulfuric acid: c (H^+) = 0.01 mol/l

V. Procedure

Place a test portion from 0.2g (expected N-content 0.5%) to 1g (expected N-content of 0.1%) of the air-dried soil sample in the digestion flask.

Add 4 ml of salycylic/sulfuric acid and swirl the flask until the acid is thoroughly mixed with the soil. Let the mixture stand for at least several hours (or overnight).

Add 0.5 g of sodium thiosulfate trough a dry funnel with a long stem that reaches down into the bulb of the digestion flask. Heat the mixture cautiously on the digestion stand until frothing has ceased.

Cool the flask and add 1.1g of the catalyst mixture, heat until the digestion mixture becomes clear.

Boil the mixture gently for up to 5 h. (in most cases a boiling period of 2h. is sufficient) so that the sulfuric acid condenses about 1/3 of the way up to the neck of the flask. Make sure that the temperature of the solution does not exceed 400°C.

Allow the flask to cool down after the digestion and add about 20ml of water slowly while shaking. Then swirl the flask to bring any insoluble material into suspension and transfer then the contents to the distillation apparatus. Rinse tree time with water to complete the transfer.

Add 5 ml of boric acid to a 100 ml conical flask. Place the flask under the condenser of the distillation apparatus, make sure that the end of the condenser dips into the solution.

Add 20 ml of sodium hydroxide to the funnel of the apparatus and run the alkali slowly into the distillation chamber.

Distil about 40 ml of the condensate and rinse the end of the condenser.

Add a few drops of indicator to the distillate and titrate with sulfuric acid to a violet endpoint or use a potentiometric titration with endpoint pH=5.

Notes:

- *Carry out a blank test in which the same procedure is performed without soil.*
- *A potentiometric titration is also possible (endpoint of titration should be pH=5).*
- *If steam distillation is used, a distillation rate up to about 25ml/min is applicable. Stop the distillation when about 100ml of distillate have been collected.*

VI. Calculation

The total nitrogen content is calculated by use of the following formula:

$$w_N = \frac{(V_1 - V_0) \times c(H^+) \times M_N}{m} \times \frac{100 + w_{H_2O}}{100}$$

Where

W_N	=	The total nitrogen content (mg/g = g/kg)
V_1	=	Volume of the sulfuric acid used in the titration of the sample (ml)
V_0	=	Volume of the sulfuric acid used in the titration of the blank sample (ml)
$c(H^+)$	=	Concentration of H+ in the sulfuric acid (moles/litre)
M_N	=	The molar mass of nitrogen (= 14 g/mol)
m	=	Mass of the air-dried soil sample (g)
w_{H2O}	=	Water content of the soil sample, based on oven-dried soil (% by mass)

VII. Report

Report total nitrogen in g/kg with 1 decimal place on the basis of oven-dried oil.

VIII. Reference

ISO 11261. 1995. Soil Quality – Determination of total nitrogen – Modified Kjeldahl method. International Organization for Standardization. Geneva, Switzerland. 4p. (available at www.iso.ch)

Soil Analysis Method 10 (SA10)

Determination of Exchangeable Cations (Al, Ca, Fe, K, Mg, Mn, Na),

Free H^+ and Exchangeable Acidity

Exchangeable acidity and exchangeable cations	
Method sheet	SA10
Reference Methods	ISO11260 & ISO 14254
Method suitable for	Organic Layer, Mineral Layer

I. Relevance in ICP Forests

Exchangeable cations (Ca, Mg, K, Na, Al, Fe, Mn)

Priority	Level I	Level II
Organic Layer	Mandatory[1]	Mandatory[1]
Mineral layer	Mandatory	Mandatory

[1] in calcareous soil, this parameter is optional

Determination of free H^+ acidity

Priority	Level I	Level II
Organic Layer	Mandatory[1]	Mandatory[1]
Mineral layer	Mandatory	Mandatory

[1] in calcareous soil, this parameter is optional

Exchangeable acidity

Priority	Level I	Level II
Organic Layer	Mandatory[1]	Mandatory[1]
Mineral layer	Mandatory	Mandatory

[1] in calcareous soil, this parameter is optional

II. Principle

The soil is first saturated with respect to barium by treating the soil one single time with a 0,1 mol/l barium chloride solution.

Concentrations of the exchangeable basic cations sodium, potassium, calcium and magnesium and the exchangeable acid cations iron, manganese, aluminium are determined in the 0.1 mol/l barium chloride extract of the soil using spectrometry.

To determine free acidity, the 0.1 mol/l extract is titrated with a 0.05 mol/l NaOH solution up to pH = 7.8. Determination of the free H+ acidity is realised using a method in which sodium fluoride is added to the soil extract before the titration (Aluminium ions are complexed and only the H+ acidity is detected during the titration process).

Note: *the reference method deviates from ISO ISO11260 & ISO 14254 in the sense that one single barium chloride extraction must be used in stead of three extractions*

Alternatively the free H^+ acidity can be determined by the "German calculation method" based on the pH of the barium chloride solution before and after extraction (König et al. 2005). The free acidity is subsequently calculated based on the sum of the acid cations and the free H^+.

III. Apparatus

Centrifuge + centrifuge tubes
Mechanical shaker

Laboratory glassware
Magnetic stirrer
pH-meter
Burette
Atomic Absorption Spectrometer (AAS) / Flame Emission Spectrometer (FES) / Inductively Coupled Plasma Spectrometer (ICP)

IV. Reagents

Barium chloride ($BaCl_2$) solution, conc. 0.1 mol/l
Sodium hydroxide (NaOH) solution, conc. 0.05 mol/l
Sodium fluoride (NaF) solution, conc. 1 mol/l
pH buffer solutions
Calibration substances

V. Procedure

Laboratory sample
Use 2.5 g air-dried soil (particle size < 2 mm) of known moisture content.

Leaching procedure
Place the laboratory sample in a 50 ml centrifuge tube. Add 30 ml barium chloride solution and shake for 2 hours. Centrifuge at 3000 g for 10 min. Transfer the supernatant liquid to a 100 ml volumetric flask. Make up to the volume of the volumetric flask with barium chloride solution and mix and filter the extract. Retain the extract for analysis (Volume V).

Note: According to ISO11260 & ISO 14254 three $BaCl_2$ extractions should be done and each time shaken for 1 hour.

Determination of exchangeable cations (Ca, Mg, K, Na, Al, Fe, Mn)
Measure the exchangeable cations in the extract using one of the spectrometric determination methods.

Determination of free H^+
Pipette 25 ml of the extract (Volume V_s). Add 1.25 ml of the sodium fluoride (1 mol/l) solution. Titrate with the sodium hydroxide (0.05 mol/l) solution to a pH value of 7.8. Titrate a blank in the same way.

Note: *If 25 ml is not sufficient for the titration, new $BaCl_2$ extract, in accordance to ISO 11260, should be obtained and used.*

Determination of exchangeable acidity
Pipette 25 ml of the extract into a container of sufficient capacity to also receive the electrodes of the pH-meter. Insert the electrodes and titrate with the sodium hydroxide (0.05 mol/l) solution until a pH value of 7.8 is reached and remains stable for 30 s. Repeat the procedure for a blank 0.1 mol/l $BaCl_2$ solution extract.

Note: If 25 ml is not sufficient for the titration, new $BaCl_2$ extract, in accordance to ISO 11260, should be obtained and used.

VI. Calculation

Determination of exchangeable cations (Ca, Mg, K, Na, Al, Fe, Mn)

Calculation according to apparatus taking into account following equivalent weights in g/mol:

Na^+	= 22,99	Ca^{2+}	= 20,04	Fe^{3+}	= 18,62	Al^{3+}	= 8,99
K^+	= 39,10	Mg^{2+}	= 12,16	Mn^{2+}	= 27,47	H^+	= 1,01

Determination of exchangeable acidity
The total exchangeable acidity on basis of air-dried soil is given by:

$$E_A = \frac{(V_a - V_B) . c_{NaOH} . 100 . V}{V_s . m}$$

where

E_A : total exchangeable acidity (cmol/kg) of the soil on basis of air-dried soil
V_A : volume NaOH (ml) used for the test sample
V_B : volume NaOH (ml) used for the blank
c_{NaOH} : concentration of NaOH (mol/l)
V_S : volume (ml) pipetted for analysis
m : mass (g) of the laboratory sample
V : final volume (ml) of the extract

Determination of free H^+
For free H^+ acidity use the same equation as for exchangeable acidity but use the volumes V_a and V_b for the volume NaOH used in the titration for free acidity.

Alternative method for the determination of free H^+ ("German" calculation method)

Calculation of the Proton equivalent per gram soil:

$$H^+(\mu mol/g) = \frac{(10^{-pH_p} - 10^{-pH_0}) * V * 1000}{m * 0{,}88} - \frac{c(Al) * V}{m * M(Al) * \left(1 + \frac{10^{-pH_p}}{10^{-5{,}85}}\right)}$$

Or

$$H^+(\mu mol/g) = \frac{(10^{-pH_p} - 10^{-pH_0}) * V * 1000}{m * 0{,}88} - \frac{c(Al) * V}{m * M(Al) * F}$$

where F = the Ulrich/Prenzel factor. Values of the F factor for different pH values can be read from Table SA10-1.

H^+	=	Free H^+ in µmol/g
pH_P	=	pH-value of the $BaCl_2$ extract after the leaching procedure
pH_0	=	pH-value of the pure $BaCl_2$-extract
V	=	Final Volume of the extract in ml (100 ml)
m	=	Mass of the laboratory sample in g (2.5 g)
c(Al)	=	Concentration of the Aluminium in the $BaCl_2$ extract in mg/l
M(Al)	=	Molecular weight of Aluminium in g/mol (26,98 g/mol)
F	=	Ulrich/Prenzel factor (cf. Table SA10-1)

Note:
As alternative method, the exchangeable acidity can be calculated as the sum of the exchangeable acid cations (Al, Fe, Mn, H).

Table SA10-1: The Ulrich/Prenzel factor (F) for a range of pH_p values (König and Fortman, 1996)

pH	F	pH	F	pH	F	pH	F	pH	F	pH	F
		4,6	18,8	4,1	57,2	3,6	179	3,1	563	2,6	1774
		4,59	19,2	4,09	58,5	3,59	1 83	3,09	576	2,59	1816
		4,58	19,6	4,08	59,9	3,58	187	3,08	590	2,58	1858
		4,57	20,1	4,07	61,3	3,57	192	3,07	604	2,57	1900
		4,56	20,5	4,06	62,7	3,56	196	3,06	618	2,56	1943
		4,55	21	4,05	64,1	3,55	201	3,05	632	2,55	1993
		4,54	21,4	4,04	65,6	3,54	205	3,04	647	2,54	2035
		4,53	21,9	4,03	67,1	3,53	210	3,03	662	2,53	2084
		4,52	22,4	4,02	68,6	3,52	215	3,02	677	2,52	2134
		4,51	22,9	4,01	70,2	3,51	220	3,01	693	2,51	2183
		4,50	23,4	4	71,8	3,5	225	3	709	2,5	2233
		4,49	23,9	3,99	73,5	3,49	230	2,99	721	2,49	2289
		4,48	24,4	3,98	75,1	3,48	235	2,98	743	2,48	2341
		4,47	25	3,97	76,9	3,47	241	2,97	757	2,47	2401
		4,46	25,5	3,96	78,6	3,46	246	2,96	778	2,46	2451
		4,45	26,1	3,95	80,4	3,45	252	2,95	792	2,45	2511
		4,44	26,7	3,94	82,3	3,44	258	2,94	813	2,44	2571
		4,43	27,3	3,93	84,2	3,43	264	2,93	827	2,43	2631
		4,42	27,9	3,92	86,2	3,42	270	2,92	848	2,42	2691
		4,41	28,5	3,91	88,1	3,41	276	2,91	870	2,41	2751
		4,4	29,2	3,9	90,1	3,4	283	2,9	891	2,4	2821
		4,39	29,8	3,89	92,2	3,39	289	2,89	912	2,39	2881
		4,38	30,5	3,88	94,3	3,38	296	2,88	933	2,38	2961
		4,37	31,2	3,87	96,5	3,37	303	2,87	954	2,37	3021
		4,36	31,9	3,86	98,7	3,36	310	2,86	976	2,36	3091
		4,35	32,6	3,85	101	3,35	317	2,85	997	2,35	3161
		4,34	33,4	3,84	103	3,34	325	2,84	1024	2,34	3241
		4,33	34,1	3,83	106	3,33	332	2,83	1046	2,33	3311
		4,32	34,9	3,82	108	3,32	340	2,82	1067	2,32	3391
		4,31	35,7	3,81	111	3,31	348	2,81	1095	2,31	3471
		4 3	36,5	3,8	113	3,3	356	2,8	1117	2,30	3551
4,8	12,2	4,29	37,3	3,79	116	3,29	364	2,79	1145	2,29	3631
4,79		4,28	38,2	3,78	118	3,28	373	2,78	1173	2,28	3721
4,78		4,27	39	3,77	121	3,27	381	2,77	1202	2,27	3801
4,77	13	4,26	39,9	3,76	124	3,26	390	2,76	1230	2,26	3891
4,76	13,3	4,25	40,8	3,75	127	3,25	399	2,75	1258	2,25	3981
4,75	13,6	4,24	41,7	3,74	130	3,24	408	2,74	1286	2,24	4071
4,74	13,9	4,23	42,7	3,73	133	3,23	418	2,73	1315	2,23	4171
4,73	14,2	4,22	43,9	3,72	136	3,22	430	2,72	1350	2,22	4271
4,72	14,5	4,21	44,7	3,71	139	3,21	438	2,71	1378	2,21	4371
4,71	14,8	4,20	45,1	3,70	142	3,20	448	2,70	1413	2,20	4471
4,7	15,1	4,19	46,7	3,69	146	3,19	458	2,69	1442	2,19	4571
4,69	15,5	4,18	47,3	3,68	149	3,18	469	2,68	1477	2,18	4681
4,68	15,8	4,17	48,9	3,67	152	3,17	480	2,67	1512	2,17	4791
4,67	16,1	4,16	50	3,66	156	3,16	491	2,66	1548	2,16	4901
4,66	16,5	4,15	51,1	3,65	159	3,15	502	2,65	1583	2,15	5001
4,65	16,8	4,14	52,3	3,64	163	3,14	514	2,64	1618	2,14	5131
4,64	17,2	4,13	53,5	3,63	167	3,13	526	2,63	1654	2,13	5251
4,63	17,6	4,12	54,7	3,62	170	3,12	538	2,62	1695	2,12	5371
4,62	18	4,11	56	3,61	175	3,11	551	2,61	1731	2,11	5501
4,61	18,4	4,10	57,2	3,60	179	3,10	563	2,60	1774	2,10	5621

VII. Report

Report (in cmol(+)/kg) total exchangeable acidity, exchangeable cations and free H^+ with 2 decimal places on the basis of oven-dried soil.

VIII. References

ISO 11260. 1994. Soil Quality – Determination of effective cation exchange capacity and base saturation level using barium chloride solution. International Organization for Standardization. Geneva, Switzerland. 10 p. (available at www.iso.ch)

ISO 14254. 1994. Soil Quality – Determination of exchangeable acidity in barium chloride extracts. International Organization for Standardization. Geneva, Switzerland. 5 p. (available at www.iso.ch)

König and Fortmann 1996. Probenvorbereitungs-, Untersuchungs- und Element-bestimmungsmethoden des Umweltlabors der Niedersächsischen Forstlichen Versuchsanstalt und des Zentrallabors II des Forschungszentrums Waldökosysteme, Teil 4: Probenvorbereitungs- und Untersuchungsmethoden, Qualitätskontrolle und Datenverarbeitung; Berichte des Forschungszentrums Waldökosyst. B, Bd. 49, Untersuchungsmethode Boden AKEG1.1

König, N. and Bartners, H. Eds. 2005. Eine Handbuch Forsliche Analytik. Loseblatt-Sammlung der Analysemethoden im Forestbereich, Herausgegeben vom Gutacherausschuss Forstliche Analytik. 433 pg. (Method A3.2.1.3)

Soil Analysis Method 11 (SA11)

Aqua Regia Extractant Determinations

P, Ca, K, Mg, Mn,

Cu, Pb, Cd, Zn,

Al, Fe, Cr, Ni, S, Hg, Na

Aqua Regia extractant determinations P, Ca, K, Mg, Mn, Cu, Pb, Cd, Zn, Al, Fe, Cr, Ni, S, Hg, Na	
Method sheet	SA11
Reference methods	ISO 11466
Method suitable for	Organic Layer, Mineral Layer

I. Relevance in ICP Forests

Aqua Regia extractant determinations (P, Ca, K, Mg, Mn)

Priority	**Level I**	**Level II**
Organic Layer	Mandatory	Mandatory
Mineral layer	Optional	Optional

Aqua Regia extractant determinations (Cu, Pb, Cd, Zn)

Priority	**Level I**	**Level II**
Organic Layer	Mandatory	Mandatory
Mineral layer	Mandatory	Mandatory

Aqua Regia extractant determinations (Al, Fe, Cr, Ni, S, Hg, Na)

Priority	**Level I**	**Level II**
Organic Layer	Optional	Optional
Mineral layer	Optional	Optional

II. Principle

The dried Sample is extracted with a hydrochloric/nitric acid mixture by standing for 16 h at room temperature, followed by boiling under reflux for 2 h. The extract is then clarified and made up to volume with nitric acid. Elements are determined by spectrometry.

III. Apparatus

Analytical balance (accuracy 0.001 g)
Desiccator (2 l)
Reaction vessel (250 ml)
Reflux condenser
Absorption vessel, non return type, containing 15 ml of nitric acid (0.5 mol/l) (only necessary for determination of mercury)
Roughened glass beads or antibumping granules
Temperature-controlled heating apparatus
Funnel (diam. approx. 110 cm)
Volumetric flask (110 ml)
Filter paper (diam. 150 mm, pore size approx. 8 µm)
Atomic Absorption Spectrometer (AAS) / Flame Emission Spectrometer (FES) / Inductively Coupled Plasma Spectrometer (ICP) / Colorimeter

IV. Reagents

Water (grade 2)
Hydrochloric acid (HCl) concentration 12 mol/l, $\rho \approx 1.19$ g/ml
Nitric acid (HNO_3) concentration 15.8 mol/l, $\rho \approx 1.42$ g/ml
Nitric acid (HNO_3) concentration 0.5 mol/l

V. Procedure

Laboratory sample
Weigh 3,000 g air-dried soil (particle size < 2 mm) of known moisture content in the 250 ml reaction vessel.

Note: *Because we are interested in the easily available elements, it is not allowed to mill the < 2mm sample. This deviates from the ISO norm.*

Aqua regia extraction
Moisten with about 0.5 ml to 1.0 ml of water and add, while mixing, 21 ml of hydrochloric acid followed by 7 ml of nitric acid (15.8 mol/l), drop by drop if necessary, to reduce foaming. Connect the condenser (and the absorption vessel) to the reaction vessel, and allow to stand for 16 h at room temperature to allow for slow oxidation of the organic matter in the soil.

The amount of aqua regia is sufficient only for oxidation of about 0.5 g of organic carbon. If there is more than 0.5 g of organic carbon in the 3 g subsample, proceed as follows. Allow the first reaction with the aqua regia to subside. Then add an extra 1 ml of nitric acid (15.8 mol/l) 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 time, and allow any reaction to subside before proceeding further.

Raise the temperature of the reaction mixture slowly until reflux conditions are reached and maintain for 2 h, ensuring that the condensation zone is lower than 1/3 of the height of the condenser, then allow to cool.

Allow the reaction vessel to stand so that most of any insoluble residue settles out of suspension. (Add the contents of the absorption vessel to the reaction vessel, via the condenser, rinsing both the absorption vessel and condenser with a further 10 ml of nitric acid (0.5 mol/l)). Decant the relatively sediment-free supernatant carefully onto a filter paper, collecting the filtrate in a 100 ml volumetric flask. Allow all the initial filtrate to pass through the filter paper, then wash the insoluble residue onto the filter paper with a minimum of nitric acid (0.5 mol/l). Collect this filtrate with the first. before proceeding further. The extract thus prepared is ready for the determination of trace elements, by an appropriate method.

Determination of elements (P, Ca, K, Mg, Mn, Cu, Pb, Cd, Zn, Al, Fe, Cr, Ni, S, Hg, Na)
Measure the elements cations in the extract using one of the spectrometric determination methods.
Note: ISO 11047 can be used as a guideline for the determination of Cd, Cr, Cu, Pb, Mn, Ni and Zn.

VI. Calculation

Determination of elements (P, Ca, K, Mg, Mn, Cu, Pb, Cd, Zn, Al, Fe, Cr, Ni, S, Hg, Na)
Calculation according to apparatus.

VII. Report

Report aqua regia extract determinations (mg/kg) with 1 decimal place on the basis of oven-dried soil.

Note*: Laboratories which have the possibility to determine the Cu content up to 2 decimal places and the Hg content up to 3 decimal places, are given the opportunity to report accordingly.*

VIII. Reference

ISO 11466. 1995. Soil Quality – Extraction of trace elements soluble in *aqua regia*. International Organization for Standardization. Geneva, Switzerland. 6 p. (available at www.iso.ch)

ISO 11047. 1998. Soil Quality – Determination of cadmium, chromium, cobalt, copper, lead, manganese nickel and zinc. Flame and electrothermal atomic absorption spectrometric methods. International Organization for Standardization. Geneva, Switzerland. 6 p. (available at www.iso.ch)

Soil Analysis Method 12 (SA12)

Determination of Total Elements

Ca, Mg, Na, K, Al, Fe, Mn