Determination of Nutrients

Determination of Kjeldahl nitrogen

Introduction

This document is developed in the project 'Horizontal'. It is the result of desk study "Determination of total phosphorus and nitrogen and fractions" in the project and aims at the description of the to determination of Kjeldahl nitrogen according to the Kjeldahl procedure in soil, sludge, biowaste and related waste. After discussion with all parties concerned in CEN and selection of a number of test methods described in this study will be developed further as an modular horizontal method and validated in the project 'Horizontal' .

Until now test methods determining properties of materials were often prepared in Technical Committees (TC's) working on specific products or specific sectors. In those test methods often steps as sampling, extraction, release or other processing, analyses, etc were included. In this approach it was necessary to develop, edit and validate similar procedural steps over and over again for each other product.. Consequently this resulted in a lot of duplicate work. To avoid such duplication of work for parts of a testing procedure often was referred to parts of test methods from other TC's. However following problems are often encountered while using references in this way: 1).The referenced parts are often not edited in a way that they could easily be referred to, 2). the referenced parts are often not validated for the other type of material and 3) the updates of such test standards on products might lead to inadequate references.

In the growing amount of product and sector oriented test methods it was recognised that many steps in test procedures are or could be used in test procedures for many products, materials and sectors. It was supposed that, by careful determination of these steps and selection of specific questions within these steps, elements of the test procedure could be described in a way that they can be used for all materials and products or for all materials and products with certain specifications.
Based on this hypothesis a horizontal modular approach is being investigated and developed in the project 'Horizontal'. 'Horizontal' means that the methods can be used for a wide range of materials and products with certain properties. 'Modular' means that a test standard developed in this approach concerns a specific step in a test procedure and not the whole test procedure (from sampling to analyses) but only.

The use of modular horizontal standards implies the drawing of test schemes as well. Before executing a test on a certain material or product to determine certain characteristics it is necessary to draw up a protocol in which the adequate modules are selected and together form the basis for the test procedure.

This standard is a module, for determining nutrients in solid materials and liquids. This module concerns with the digestion method according to Kjeldahl and the determination of the Kjeldahl nitrogen in soil, sludge, biowaste and related waste .

The other horizontal modules that will be available in due time are to be found in the informative annex C which contains a brief overview of the modules that are or will be worked out in the project 'Horizontal.'

The texts of the chapters 1 -11 are normative ; annexes are normative or informative, as stated in the top lines of the annexes.

1 Normative references

ISO 11464 Soil quality – Pretreatment of samples for physico-chemical analysis
ISO 11465 Soil quality – Determaination of dry matter and water content on a mass basis – gravimetric method
EN 12880 Characterisation of sludge – Determination of dry residue and water content
CEN/TC 292 WI 29292030 Characterisation of waste – Preparation of test portions from the laboratory
EN 13342 Characterization of sludges - Determination of Kjeldahl nitrogen

2 Scope and working area

This standard is to determine Kjeldahl nitrogen according to the Kjeldahl procedure in soil, sludge, biowaste and related waste. Nitrate and nitrite are not included. Compounds with special chemical N-bonding (N-N, N-O and heterocycles) are not digested entirely.

3 Principle

The dried and homogenised material is digested in a suitable Kjeldahltube with sulfuric acid. To rise the temperature potassium sulfate is added and titanium dioxid/copper sulfate is used as a catalyst. After adding sodium hydroxide to the digestion solution the produced ammonium from all nitrogen species is evaporated by distillation as ammonia. This is condensed in a conical flask with boric acid solution. The amount is titrated against indicator with sulfuric acid.

4 Reagents

4.1 General
All reagents shall be of analytical grade. Use water of grade 2 complying with ISO 3696

4.2 Sulfuric acid, ρ = 1.84 kg /l

4.3 Potassium sulfate catalyst mixture
Grind and thoroughly mix 200 g of potassium sulfate, 6g of copper sulfate pentahydrate and 6 g of titanium dioxide, with the crystal structure of anatase.

4.4 Sodium hydroxide, c (NaOH) = 10 mol/l

4.5 Boric acid solution , ρ = 20g /l

4.6 Mixed inicator
Dissolve 0.1 g of bromocresol green and 0.02 g of methyl red in 100 ml ethanol.

4.7 Sulfuric acid, $c(H^+)$ = 0.01 mol/l

4.8 Ammonium sulfate NH_4SO_4

5 Apparatus

5.1 General
Usual laboratory equipment is needed

5.2 Kjeldahl digestion flasks or tubes, of nominal volume 50 ml, suitable for digestion stand (6.2). To use the semimikro- or the makroproceeding respective flasks or tubes are to be used.

5.3 Digestion stand, suitable for digestion of samples with sulfuric acid at temperature near to 400 °C and fit to evaporate the fume

5.4 Distillation apparatus, e.g. of the Parnas-Wagner type or other suitable distillattion apparatus with steam generator

5.5 Burette, graduated in intervals of 0.01 ml or smaller.

Note 1: Modern Kjeldahl apparatus combine digestion and/or distillation and titration in an automated method. See instructions of the manufacturer. These apparatus are usable.

6 Calibration samples

Calibration substances with known and unchangeable content of nitrogen are used to control the digestion and the apparatus. This may be: acetanilid, l-asparaginacid, sulfanilacid or other aminoacids with known nitrogen content. Besides these substances certified reference materials are used to control the whole procedure

7 Pretreatment of test samples

All samples shall be pretreated according to the special standard in the field of soil, sludge, biowaste and related waste, so that they are dry, homogenuous and of a defined grain size.

Note 2: During the drying procedure there is taken care not to loose amounts of ammonium-N and/or nitrate-N. Therefore, excessive drying (105 °C) should be avoided. Rapid microwave drying may be a good choice.

8 Working instructions

Note 3: Homogeneity of the laboratory sample and the test sample has to be guaranteed.

8.1 Digestion

Place a test portion of the dried and grinded sample, of about 0.2 g (exspected nitrogen content $\approx$ 0.5 %) to 1g (exspected nitrogen content $\approx$ 0.1 %) or undried sample with the corresponding dry matter to the nearest of 0,1% accuracy in the digestion flask or tube (5.2). Add 5 ml sulfuric acid (4.2) and swirl until the acid is thoroughly mixed with the sample. Allow the mixture to stand for cooling. Add 1.1 g of the catalyst mixture (4.3) and heat until the digestion mixture becomes clear. Boil the mixture gently for up to 5 h so that the sulfuric acid condenses about 1/3 of the way

up to the neck of the flask or the end of the tube. Ensure that the temperature of the solution does not exceed 400 °C .

Note 4: The amount of sulfuric acid may be adopted (see Annex B)

Note 5: The time of boiling period may be different and depends on the sample material. The solution has to be clear at the end of boiling.

Note 6: The amount of test material and added chemicals can be changed in the ratio described in the working instructions. The semimicro and the macro version of the Kjeldahl procedure are suitable for some materials.

8.2 Titration

After completion of the digestion step, allow the flask or tube to cool and add 20 ml of water slowly while shaking. Then swirl the flask or tube to bring any insoluble material into suspension and transfer the contents to the distillation apparatus (5.4). Rinse three times with water to complete the transfer. Add 5 ml of boric acid (4.5) to a 200 ml conical flask and and place the flask under the condenser of the distillation apparatus in such a way that the end of the condenser dips into the solution. Add 20 ml of sodium hydroxide (4.4) to the funnel of the apparatus and run the alkali slowly into the distillation chamber. Distil about 100 ml of condensate (the amount for quantitative results depends on the dimensions of the apparatus), rinse the end of the condenser, add a few drops of mixed indicator (4.6) to the distillate and titrate with sulfuric acid (4.7) to a violet endpoint.

Note 7: The best way of distillation is steam distillation. A rate of up to 25 ml/min is applicable. Stop the distillation when 100 ml of distillate have been collected.

8.3 Blank test

Carry out a blank test in which the same procedure is performed without sample. Notify the consumption of sulfuric acid in the blank test and in the tests of the samples. Maybe you have to rise the amount of sulfuric acid to have quantitative results.

8.4 Quality control

To test the distillation apparatus run a distillation with ammonium sulfate (4.9) only. Certified reference material is used to control the whole method.

Note 8: Modern Kjeldahl apparatus use the digestion tubes for distillation and the addition of chemicals is programmed. The distillation is done automatically. A potentiometric titration with an endpoint of pH = 5.0 is possible.

9 Calculations and expression of results

The content of nitrogen, (w_N), in milligrams per gram, is calculated using the formula:

$$w_N = \frac{(V_1 - V_0) \times c(H^+) \times M_N}{m} \times \frac{100 + w_{H2O}}{100}$$

where

V_1 is the volume, in ml, of the sulfuric acid (4.7) used in the titration of the sample

V_0 is the volume, in millilitres, of the sulfuric acid (4.7) used in the titration of the blank test

$c(H^+)$ is the concentration of H^+ in the sulfuric acid (4.7) in moles per litre (e.g. if 0.01 mol/l sulfuric acid is used, $c(H^+) = 0.02$ mol/l)

M_N is the molar mass of nitrogen, in grams per mole (=14)

m is the mass of test sample

w_{H2O} is the water content, expressed as a percentage in mass on the basis of oven dried material according to the standard of the spezial material

10 Validation and Precision

The work to produce the validation/precision data has to be done in future (see Annex B)

11 Testreport

The test report is formulated according to the spezial conditions of accredidation (EN 41001 or EN 17025)

Annex A
(informative)

The amount of sulfuric acid may be different in relation to the material of analysis. This is calculated according to the table 1 and is adapted.

Table 1 Amounts of sulfuric acid consumption by various materials during Kjeldahl digestion (Bremner 1960)

Material	Acid consumption ml of 36 mol/l H_2SO_4/ g of material
Soil organic C	10,0
Soil organic matter	5.8

Al_2O_3	1,63
Fe_2O_3	1,04
Clay	0,60
$CaCO_3$	0,55
Silt	0,33
Sand	0
Salicylic acid	6,76
$Na_2S_2O_3$	0,58
Reduced Fe	1,50

The amount of sulfuric acid has to be investigated with different materials to yield the highest digestion efficiency, see Annex B.

Annex B

(informative)

Validation and precision data

Within the work of CEN TC 308 a ring test was organized with four samples of different sludges. The following results were obtained:

Sample No.	Number of participants	Nitrogen content, Average, g/kg m_T	s_r mg/kg	s_r %	s_R mg/kg	s_R %
1	4	28,48	2,13	12,65	2,15	16,84
2	4	21,87	3,5	21,47	3,22	16,3
3	4	27,52	0,7	7,13	1,07	9,81
4	4	16,28	0,65	4,79	0,718	13,58

These results were obtained on the basis of only four laboratories and this does not agree with EN 5725.

In the future the validation has to be done with three samples of different soils, three samples of different sludges, three different samples of biowaste and three different samples of to biowaste related wastes with different contents of nitrogen each. These samples have to be dry samples.

The repeatability and the reproducibility are calculated from the results of the round robin studies with the factor 2,8 .

Annex C

(informative)

Bibliography

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