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ISO 6495

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Animal feeding stuffs — Determination of water-soluble chlorides content

Aliments des animaux — Détermination de la teneur en chlorures solubles dans l'eau

Foreword

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International Standards are drafted in accordance with the rules given in the ISO/IEC Directives, Part 3.

Draft International Standards adopted by the technical committees are circulated to the member bodies for voting. Publication as an International Standard requires approval by at least 75 % of the member bodies casting a vote.

International Standard ISO 6495 was prepared by Technical Committee ISO/TC 34, *Agricultural food products*, Subcommittee SC 10, *Animal feeding stuffs*.

This second edition of ISO 6495 cancels and replaces the first edition (ISO 6495:1980), which has been technically revised.

Annex A of this International Standard is for information only.

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Animal feeding stuffs — Determination of water-soluble chlorides content

1 Scope

This International Standard specifies a method for the determination of the water-soluble chlorides content, expressed as sodium chloride, of animal feeding stuffs.

The method is applicable to animal feeding stuffs.

2 Normative references

The following normative documents contain provisions which, through reference in this text, constitute provisions of this International Standard. For dated references, subsequent amendments to, or revisions of, any of these publications do not apply. However, parties to agreements based on this International Standard are encouraged to investigate the possibility of applying the most recent editions of the normative documents indicated below. For undated references, the latest edition of the normative document referred to applies. Members of ISO and IEC maintain registers of currently valid International Standards.

ISO 3696, *Water for analytical laboratory use — Specification and test methods.*

ISO 6498, *Animal feeding stuffs — Preparation of test samples.*

3 Principle

The chlorides present in a test portion are dissolved in water. The solution is clarified if the product contains organic matter. It is then slightly acidified with nitric acid and the chlorides are precipitated as silver chloride by means of standard volumetric silver nitrate solution. The excess silver nitrate is titrated with a standard volumetric solution of ammonium thiocyanate or potassium thiocyanate.

4 Reagents

Use only reagents of recognized analytical grade.

4.1 Water, complying with at least grade 3 in accordance with ISO 3696.

4.2 Acetone.

4.3 *n*-Hexane.

4.4 Nitric acid, $\rho_{20}(HNO_3)$ = 1,38 g/ml.

4.5 Activated carbon, free from chlorides and not capable of adsorbing chlorides.

4.6 Ammonium iron(II) sulfate , saturated solution.

Prepare from $NH_4Fe(SO_4)_2 \cdot 12H_2O$.

4.7 Carrez I solution .

Dissolve in water, 10,6 g of potassium hexacyanoferrate(II) trihydrate [$K_4Fe(CN)_6 \cdot 3H_2O$]. Dilute to 100 ml with water.

4.8 Carrez II solution.

Dissolve in water, 21,9 g of zinc acetate dihydrate [$Zn(CH_3COO)_2 \cdot 2H_2O$] and add 3 ml of glacial acetic acid. Dilute to 100 ml with water.

4.9 Ammonium thiocyanate or **potassium thiocyanate**, standard volumetric solution, $c(NH_4SCN)$ = 0,1 mol/l or $c(KSCN)$ = 0,1 mol/l.

4.10 Silver nitrate, standard volumetric solution, $c(AgNO_3)$ = 0,1 mol/l.

5 Apparatus

Usual laboratory apparatus and, in particular, the following.

5.1 Rotary shaker, operating at a rotation frequency of approximately 35 min^{-1} to 40 min^{-1}.

5.2 One-mark volumetric flasks, of capacities 200 ml and 500 ml.

5.3 Pipettes, of appropriate capacity.

5.4 Burettes.

5.5 Analytical balance, capable of weighing to the nearest 0,001 g.

6 Sampling

Sampling is not part of the method specified in this International Standard. A recommended sampling method is given in ISO 6497 [1].

It is important that the laboratory receive a sample which is truly representative and has not been damaged or changed during transport or storage.

7 Preparation of test sample

Prepare the test sample in accordance with ISO 6498.

If solid, grind the laboratory sample (usually 500 g) so that it passes completely through a sieve with 1 mm apertures. Mix thoroughly.

8 Procedure

NOTE If it is required to check whether the repeatability limit is met (see 10.2), carry out two single determinations in accordance with 8.1 to 8.5.

8.1 Procedure selection

If the test sample is free from organic matter, proceed in accordance with 8.2.

If the test sample contains organic matter, proceed in accordance with 8.3, unless the test sample concerns cooked feeding stuffs, flax cakes and flour, products rich in flax flour, and other products rich in mucilage or in colloidal substances (e.g. dextrinated starch). In this case proceed in accordance with 8.4.

8.2 Preparation of test solution of samples free from organic matter

Weigh, to the nearest 0,001 g, a test portion (mass m) of the prepared test sample of not more than 10 g presumed to contain not more than 3 g of chlorides. Transfer to a 500 ml volumetric flask (5.2) and add 400 ml of water at a temperature of approximately 20 °C.

Mix for 30 min in the rotary shaker (5.1), dilute to the mark with water (resulting volume V_i), then mix and filter.

Proceed in accordance with 8.5.

8.3 Preparation of test solution of samples containing organic matter, excluding the products listed in 8.4

Weigh approximately 5 g (mass m) of the prepared test sample to the nearest 0,001 g and transfer to a 500 ml volumetric flask (5.2). Add 1 g of activated carbon (4.5), 400 ml of water at a temperature of approximately 20 °C, and 5 ml of Carrez I solution (4.7). Stir, then add 5 ml of Carrez II solution (4.8).

Mix for 30 min on the rotary shaker (5.1). Dilute to the mark with water (resulting volume V_i), mix and filter.

Proceed in accordance with 8.5.

8.4 Cooked feeding stuffs, flax cakes and flour, products rich in flax flour, and other products rich in mucilage or in colloidal substances

Weigh approximately 5 g (mass m) of the prepared test sample to the nearest 0,001 g and transfer to a 500 ml volumetric flask (5.2). Add 1 g of activated carbon (4.5), 400 ml of water at a temperature of approximately 20 °C, and 5 ml of Carrez I solution (4.7). Stir, then add 5 ml of Carrez II solution (4.8).

Mix for 30 min on the rotary shaker (5.1). Dilute to the mark with water (resulting volume V_i) and mix.

Decant (if necessary, centrifuge). By means of a pipette (5.3), transfer 100 ml of the supernatant liquid to a 200 ml volumetric flask (5.2).

Mix with acetone (4.2), dilute to the mark with the same solvent, mix and filter.

8.5 Titration

By means of a pipette (5.3), transfer to a conical flask an aliquot portion of between 25 ml and 100 ml (volume V_a) of the filtrate. The aliquot portion shall not contain more than 150 mg of chloride.

Dilute, if necessary, to a volume of not less than 50 ml with water. Add 5 ml of nitric acid (4.4), 2 ml of ammonium iron(III) sulfate solution (4.6) and 2 drops of ammonium thiocyanate solution or potassium thiocyanate solution (4.9) from a burette (5.4) filled to the zero mark.

NOTE The remainder of the solution is to be used for titration of the excess silver nitrate.

Add from a burette (5.4), silver nitrate solution (4.10) until the reddish-brown tint disappears, then add an excess of 5 ml (resulting volume V_{s1}). Shake vigorously to coagulate the precipitate. If necessary, 5 ml of *n*-hexane (4.3) may be added to assist coagulation.

Titrate the excess silver nitrate with ammonium thiocyanate solution or potassium thiocyanate solution (4.9) until a reddish brown tint, persisting for at least 30 s, develops (resulting volume V_{t1}).

8.6 Blank test

Carry out a blank test in parallel with the determination, using the same procedure and the same reagents, but omitting the test portion.

9 Expression of results

Calculate the water-soluble chlorides content, expressed as sodium chloride, by the equation:

$$w_{wc} = \frac{M[(V_{s1} - V_{s0})c_s - (V_{t1} - V_{t0})c_t]}{m} \times \frac{V_i}{V_a} \times f \times 100\ \%$$

where

w_{wc} is the water-soluble chlorides content, expressed as sodium chloride, as percentage by mass, of the test sample;

M is the molar mass, in grams per mole, of sodium chloride (M = 58,44 g/mol);

V_{s1} is the volume, in millilitres, of the silver nitrate solution added for the titration of the test solution (8.5);

V_{s0} is the volume, in millilitres, of the silver nitrate solution added for the titration of the blank solution (8.6);

c_s is the concentration, in moles per litre, of the silver nitrate solution (4.10);

V_{t1} is the volume, in millilitres, of the ammonium thiocyanate solution or potassium thiocyanate solution used in the titration of the test solution (8.5);

V_{t0} is the volume, in millilitres, of the ammonium thiocyanate solution or potassium thiocyanate solution used in the titration of the blank solution (8.6);

c_t is the concentration, in moles per litre, of the ammonium thiocyanate or potassium thiocyanate solution (4.9);

m is the mass, in milligrams, of the test portion;

V_i is the volume, in millilitres, of the test solution prepared in 8.2, 8.3 and 8.4 (V_i = 500 ml);

V_a is the volume, in millilitres, of the aliquot portion of filtrate taken (see 8.5);

f is a dilution factor:

f = 2 for cooked feeding stuffs, flax cakes and flour, products rich in flax flour and other products in mucilage or in colloidal substances (see 8.4);

f = 1 for other animal feeding stuffs (see 8.2 and 8.3).

Report the result[1)] to the nearest:

0,05 % for water-soluble chlorides contents less than 1,5 %;

0,1 % for water-soluble chlorides contents greater than or equal to 1,5 %.

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1) Expressed as percentage by mass [% (m/m)].

10 Precision

10.1 Interlaboratory test

Details of an interlaboratory test on the precision of the method are summarized in annex A. The values derived from this interlaboratory test may not be applicable to concentrations ranges and matrices other than those given.

10.2 Repeatability

The absolute difference between two independent single test results, obtained using the same method on identical test material in the same laboratory by the same operator using the same equipment within a short interval of time, will in not more than 5 % of cases exceed the repeatability limit (r) derived from the equation:

$$r = 0{,}134(\overline{w}_{\mathrm{wc}})^{0{,}521}$$

where

r is the repeatability limit, expressed as percentage by mass;

$\overline{w}_{\mathrm{wc}}$ is the mean of the two single test results, expressed as percentage by mass.

10.3 Reproducibility

The absolute difference between two single test results, obtained using the same method on identical test material in different laboratories by different operators using different equipment, will in not more than 5 % of cases exceed the reproducibility limit (R) derived from the equation:

$$R = 0{,}552\ \% + 0{,}135\ \overline{w}_{\mathrm{wc}}$$

where

R is the reproducibility limit, expressed as percentage by mass;

$\overline{w}_{\mathrm{wc}}$ is the mean of the two single test results, expressed as percentage by mass.

11 Test report

The test report shall specify:

— all information necessary for the complete identification of the sample;

— the sampling method used, if known;

— the test method used, with reference to this International Standard;

— all operating details not specified in this International Standard, or regarded as optional, together with details of any incidents which may have influenced the test results;

— the test result(s) obtained; or

— if the repeatability has been checked, the final quoted result obtained.

Annex A
(informative)

Results of interlaboratory test

An interlaboratory test was organized by ISO/TC 34/SC 10, *Animal feeding stuffs*, in 1987 and carried out in accordance with ISO 5725:1986[2]. The final statistical analysis was carried out in accordance with ISO 5725-2:1994[4]. In the test 24 laboratories participated. Samples of corn gluten feed, finished mixed feed stuff, fish meal, mixed feed stuff concentrate (2 types), premixed feed stuff and yeast were investigated.

Table A.1 — Statistical results of the interlaboratory test

Parameter	Sample[a]						
	1	2	3	4	5	6	7
Number of laboratories retained after eliminating outliers	23	23	23	23	23	23	23
Mean water-soluble chlorides content, dry matter, %[b]	3,22	0,105	1,56	14,1	3,56	2,54	1,07
Repeatability standard deviation (s_r), %[b]	0,11	0,018	0,05	0,20	0,10	0,08	0,03
Repeatability coefficient of variation, %	3,5	17,2	3,5	1,4	2,9	3,0	2,9
Repeatability limit (r) ($r = 2{,}8 \times s_r$), %[b]	0,308	0,050	0,140	0,560	0,280	0,224	0,084
Reproducibility standard deviation (s_R), %[b]	0,22	0,22	0,37	0,88	0,50	0,30	0,16
Reproducibility coefficient of variation, %	7	210	24	6	14	12	15
Reproducibility limit (R) ($R = 2{,}8 \times s_R$), %[b]	0,616	0,616	1,036	2,464	1,400	0,840	0,448

[a] Sample 1: fish meal;
Sample 2: corn gluten feed;
Sample 3: yeast;
Sample 4: premixed feed stuff;
Sample 5: mixed feed stuff concentrate;
Sample 6: mixed feed stuff concentrate;
Sample 7: finished mixed feed stuff.

[b] Expressed as a percentage by mass [% (m/m)].

Bibliography

[1] ISO 6497, *Animal feeding stuffs — Sampling.*

[2] ISO 5725:1986, *Precision of test methods — Determination of repeatability and reproducibility for a standard test method by inter-laboratory tests* (now withdrawn).

[3] ISO 5725-1:1994, *Accuracy (trueness and precision) of measurement methods and results — Part 1: General principles and definitions.*

[4] ISO 5725-2:1994, *Accuracy (trueness and precision) of measurement methods and results — Part 2: Basic method for the determination of repeatability and reproducibility of a standard measurement method.*