Commission Regulation (EU) No 51/2013 of 16 January 2013 amending Regulation (EC) No 152/2009 as regards the methods of analysis for the determination of constituents of animal origin for the official control of feed (Text with EEA relevance)

COMMISSION REGULATION (EU) No 51/2013

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amending Regulation (EC) No 152/2009 as regards the methods of analysis for the determination of constituents of animal origin for the official control of feed

(Text with EEA relevance)

THE EUROPEAN COMMISSION,

Having regard to the Treaty on the Functioning of the European Union,

Having regard to Regulation (EC) No 882/2004 of the European Parliament and of the Council of 29 April 2004 on official controls performed to ensure the verification of compliance with feed and food law, animal health and animal welfare rules[(1)], and in particular Article 11(4) thereof,

Whereas:

(1) Article 7(1) of Regulation (EC) No 999/2001 of the European Parliament and of the Council of 22 May 2001 laying down rules for the prevention, control and eradication of certain transmissible spongiform encephalopathies[(2)] provides that the feeding to ruminants of protein derived from animals is prohibited. That prohibition is extended to animals other than ruminants and restricted, as regards the feeding of those animals with products of animal origin, in accordance with Annex IV to that Regulation.

(2) Article 11(1) of Regulation (EC) No 1069/2009 of the European Parliament and of the Council of 21 October 2009 laying down health rules as regards animal by-products and derived products not intended for human consumption and repealing Regulation (EC) No 1774/2002[(3)] prohibits the feeding of terrestrial animals of a given species other than fur animals with processed animal protein derived from the bodies or parts of bodies of animals of the same species, as well as the feeding of farmed fish with processed animal protein derived from the bodies or parts of bodies of farmed fish of the same species.

(3) Commission Regulation (EC) No 152/2009 of 27 January 2009 laying down the methods of sampling and analysis for the official control of feed[(4)] sets out in its Annex VI the methods of analysis for the determination of constituents of animal origin for the official control of feed. The microscopic method, which is currently the only method validated to detect the presence of animal proteins in feed, is able to distinguish the presence of constituents derived from terrestrial animals from the presence of constituents derived from fish, but unable to quantify with a sufficient level of accuracy the amount of animal constituents present in feed, and therefore should not be used for this purpose.

Changes to legislation: *There are currently no known outstanding effects for the Commission Regulation (EU) No 51/2013. (See end of Document for details)*

(4) A new method of detection of animal constituents based on polymerase chain reaction (PCR) was validated by the EU reference laboratory for animal proteins in feedingstuffs. An implementation study, organised with the national reference laboratories of the Member States, proved that the new method is sufficiently robust to be used as an official control method in the Union. This new method is able to detect the presence of animal constituents in feed, and also able to identify the species origin of these constituents. The use of this new method in combination with or in replacement of, as appropriate, the microscopic method would be very valuable for the control of the correct implementation of the feeding prohibitions laid down in Regulations (EC) No 999/2001 and (EC) No 1069/2009.

(5) Annex VI to Regulation (EC) No 152/2009 should therefore be replaced accordingly.

(6) The measures provided for in this Regulation are in accordance with the opinion of the Standing Committee on the Food Chain and Animal Health and neither the European Parliament nor the Council have opposed them,

HAS ADOPTED THIS REGULATION:

Article 1

Annex VI to Regulation (EC) No 152/2009 is replaced by the text set out in the Annex to this Regulation.

Article 2

This Regulation shall enter into force on the twentieth day following that of its publication in the *Official Journal of the European Union*.

This Regulation is binding in its entirety and directly applicable in all Member States.

***Changes to legislation:** There are currently no known outstanding effects for the Commission Regulation (EU) No 51/2013. (See end of Document for details)*

ANNEX

ANNEX VI

METHODS OF ANALYSIS FOR THE DETERMINATION OF CONSTITUENTS OF ANIMAL ORIGIN FOR THE OFFICIAL CONTROL OF FEED

1. PURPOSE AND SCOPE

The determination of constituents of animal origin in feed shall be performed by light microscopy or polymerase chain reaction (PCR) in accordance with the provisions laid down in this Annex.

These two methods make it possible to detect the presence of constituents of animal origin in feed materials and compound feed. However, they do not make it possible to calculate the amount of such constituents in feed materials and compound feed. Both methods have a limit of detection below 0,1 % (w/w).

The PCR method makes it possible to identify the taxonomic group of constituents of animal origin present in feed materials and compound feed.

These methods shall apply for the control of the application of the prohibitions laid down in Article 7(1) and Annex IV to Regulation (EC) No 999/2001 and in Article 11(1) of Regulation (EC) No 1069/2009.

Depending on the type of feed being tested, these methods may be used, within one single operational protocol, either on their own or combined together in accordance with the standard operating procedures (SOP) established by the EU reference laboratory for animal proteins in feedingstuffs (EURL-AP) and published on its website[(5)].

2. METHODS

2.1. **Light microscopy**

2.1.1. *Principle*

The constituents of animal origin which may be present in feed materials and compound feed sent for analysis are identified on the basis of typical and microscopically identifiable characteristics like muscle fibres and other meat particles, cartilage, bones, horn, hair, bristles, blood, feathers, egg shells, fish bones and scales.

2.1.2. *Reagents and equipment*

2.1.2.1. Reagents

2.1.2.1.1. Concentrating agent

2.1.2.1.1.1. Tetrachloroethylene (specific gravity 1,62)

2.1.2.1.2. Staining reagent

2.1.2.1.2.1. Alizarin Red solution (dilute 2,5 ml 1M hydrochloric acid in 100 ml water and add 200 mg Alizarin Red to this solution)

2.1.2.1.3. Mounting media

2.1.2.1.3.1. Lye (NaOH 2,5 % w/v or KOH 2,5 % w/v)

2.1.2.1.3.2. Glycerol (undiluted, viscosity: 1 490 cP)

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2.1.2.1.3.3. Norland ® Optical Adhesive 65 (viscosity: 1 200 cP) or a resin with equivalent properties for permanent slide preparation

2.1.2.1.4. Mounting media with staining properties

2.1.2.1.4.1. Lugol solution (dissolve 2 g potassium iodide in 100 ml water and add 1 g iodine while frequently shaking)

2.1.2.1.4.2. Cystine reagent (2 g lead acetate, 10 g NaOH/100 ml water)

2.1.2.1.4.3. Fehling's reagent (prepared before use from equals parts (1/1) of two stock solutions A and B. Solution A: dissolve 6,9 g copper (II) sulphate pentahydrate in 100 ml water. Solution B: dissolve 34,6 g potassium sodium tartrate tetrahydrate and 12 g NaOH in 100 ml water)

2.1.2.1.4.4. Tetramethylbenzidine/Hydrogen peroxide. (dissolve 1 g 3,3',5,5' tetramethylbenzidine (TMB) in 100 ml glacial acetic acid and 150 ml water. Before use, mix 4 parts of this TMB solution with 1 part 3 % hydrogen peroxide)

2.1.2.1.5. Rinsing agents

2.1.2.1.5.1. Ethanol ≥ 96 % (technical grade)

2.1.2.1.5.2. Acetone (technical grade)

2.1.2.1.6. Bleaching reagent

2.1.2.1.6.1. Commercial sodium hypochlorite solution (9 - 14 % active chlorine)

2.1.2.2. Equipment

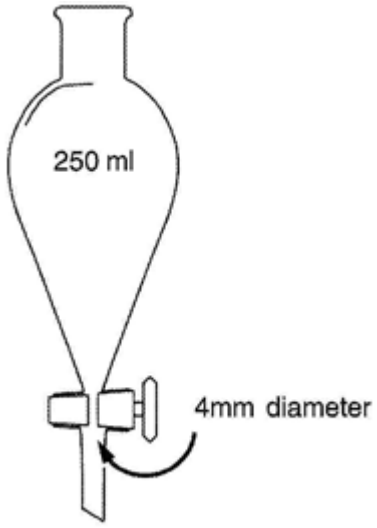
2.1.2.2.1. Analytical balance with an accuracy of 0,001 g

2.1.2.2.2. Grinding equipment: mill - or mortar

2.1.2.2.3. Sieves with square meshes of 0,25 mm and 1 mm width

2.1.2.2.4. Conical glass separation funnel with a content of 250 ml with Teflon or ground glass stopcock at the base of the cone. Stopcock opening diameter shall be ≥ 4mm. Alternatively, a conical bottomed settling beaker may be used provided the laboratory has demonstrated that detection levels are equivalent to that obtained using the conical glass separation funnel.
Separation funnel

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2.1.2.2.5. Stereomicroscope covering at least a 6,5× to 40× final magnification range

2.1.2.2.6. Compound microscope covering at least a 100× to 400× final magnification range with transmitted light bright field. Polarised light and differential interferential contrast can additionally be used

2.1.2.2.7. Standard laboratory glassware

2.1.2.2.8. Equipment for slide preparation: classical microscope slides, hollow slides, coverslips (20 × 20 mm), tweezers, fine spatula

2.1.3. *Sampling and sample preparation*

2.1.3.1. Sampling

A representative sample, taken in accordance with the provisions laid down in Annex I shall be used.

2.1.3.2. Precautions to be taken

In order to avoid laboratory cross-contamination, all reusable equipment shall be carefully cleaned before use. Separation funnel pieces shall be disassembled before cleaning. Separation funnel pieces and glassware shall be pre-washed manually and then washed in a washing machine. Sieves shall be cleaned by using a brush with stiff synthetic hairs. A final cleaning of sieves with acetone and compressed air is recommended after sieving of fatty material like fishmeal.

2.1.3.3. Preparation of samples other than fat or oil

2.1.3.3.1. Sample drying: samples with a moisture content > 14 % shall be dried prior to handling.

2.1.3.3.2. Sample pre-sieving: it is recommended to pre-sieve at 1 mm pelleted feeds and kernels and to subsequently prepare and analyse the two resulting fractions as distinct samples.

2.1.3.3.3. Sub-sampling and grinding: at least 50 g of the sample shall be sub-sampled for analysis and subsequently ground.