

Commission Implementing Regulation (EU) 2020/1560 of 26 October
2020 amending Annex VI to Regulation (EC) No 152/2009 laying
down the methods of analysis for the determination of constituents of
animal origin for the official control of feed (Text with EEA relevance)

ANNEX

Annex VI to Regulation (EC) No 152/2009 is amended as follows:

- (1) point 2.1.1. is replaced by the following:

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, milk globules, lactose crystals, feathers, egg shells, fish bones and scales.

- (2) point 2.1.2.1.3.2, is replaced by the following:

Glycerol (undiluted, viscosity: 1 490 cP) or a mounting medium with equivalent properties for non-permanent slide preparation.

- (3) point 2.1.2.2.2, is replaced by the following:

Grinding equipment: knife or rotor mill. If a rotor mill is used, mill sieves $\leq 0,5$ mm shall be prohibited.

- (4) point 2.1.2.2.3, is replaced by the following:

Sieves with square meshes of 0,25 mm and 1 mm width. With the exception of sample pre-sieving, the diameter of the sieves should not exceed 10 cm to avoid loss of materials. Calibration of sieves is not required.

- (5) the following points are added in point 2.1.2.2:

2.1.2.2.9. Laboratory oven

2.1.2.2.10. Centrifuge

2.1.2.2.11. Filter paper: qualitative cellulose filter (pore size 4-11 μm).

- (6) point 2.1.3.1, is replaced by the following:

Sampling

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

- (7) point 2.1.3.3.1, is replaced by the following:

Sample drying: samples with a moisture content > 14 % shall be dried prior to handling according to Annex III to this Regulation.

- (8) point 2.1.3.3.2, is replaced by the following:

Sample pre-sieving: in order to collect information on possible environmental contamination of the feed, it is recommended to pre-sieve at 1 mm pelleted feeds and kernels and to subsequently prepare, analyse, and report separately on the two resulting fractions, which must be considered as distinct samples.

- (9) the last paragraph of point 2.1.3.3.4 is replaced by the following:

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The sediment shall be collected on a filter paper placed into a funnel to allow the separation of the remaining TCE while avoiding fat deposition into the sediment. The sediment shall be dried. It is recommended to subsequently weigh the sediment (accurate to 0,001 g) to control the sedimentation step. Lastly, the sediment shall be sieved at 0,25 mm and the two resulting fractions shall be examined, unless sieving is not deemed necessary.

- (10) the first sentence of point 2.1.4.1 is replaced by the following:

Microscopic slides shall be prepared from the sediment and, depending on the operator's choice, from either the flotata or the raw material.

- (11) point 2.1.4.2, including its Diagrams 1 and 2, is replaced by the following:

‘Observation flowchart for the detection of animal particles in compound feed and feed material

The prepared microscopic slides shall be observed in accordance with the observation flowcharts laid down in diagrams 1 and 2.

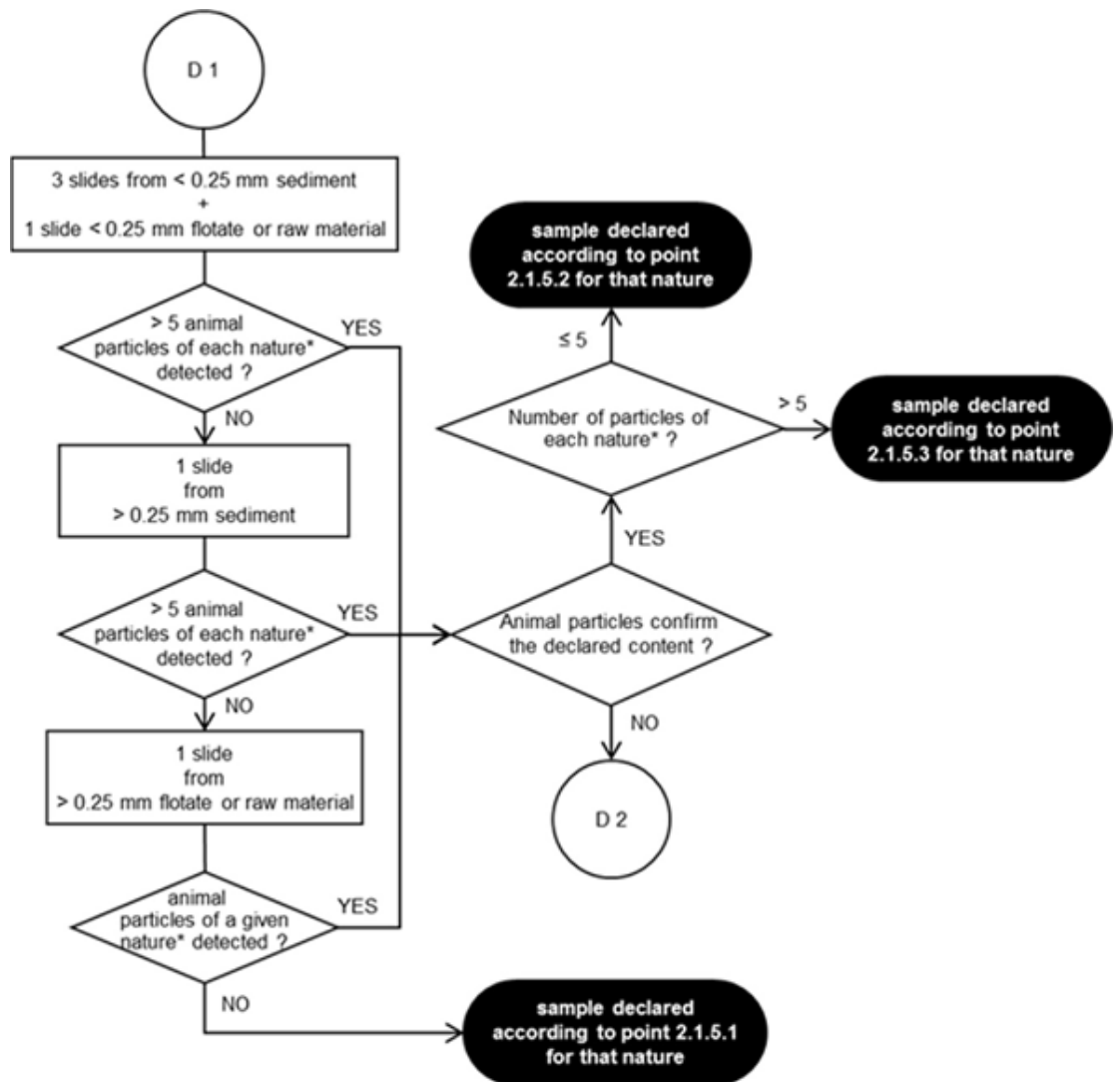
The microscopic observations shall be conducted using the compound microscope on the sediment and, depending on the operator's choice, either on the flotata or on the raw material. The stereomicroscope may be used in addition to the compound microscope for the coarse fractions. Each slide shall be screened entirely at various magnifications. Precise explanations on how to use the observation flowcharts are detailed by a SOP established by the EURL-AP and published on its website.

The minimum numbers of slides to be observed at each step of the observation flowcharts shall be strictly respected, unless the entire fraction material does not permit to reach the stipulated slide number, for instance when no sediment is obtained. No more than 6 slides per determination shall be used for recording of the number of particles.

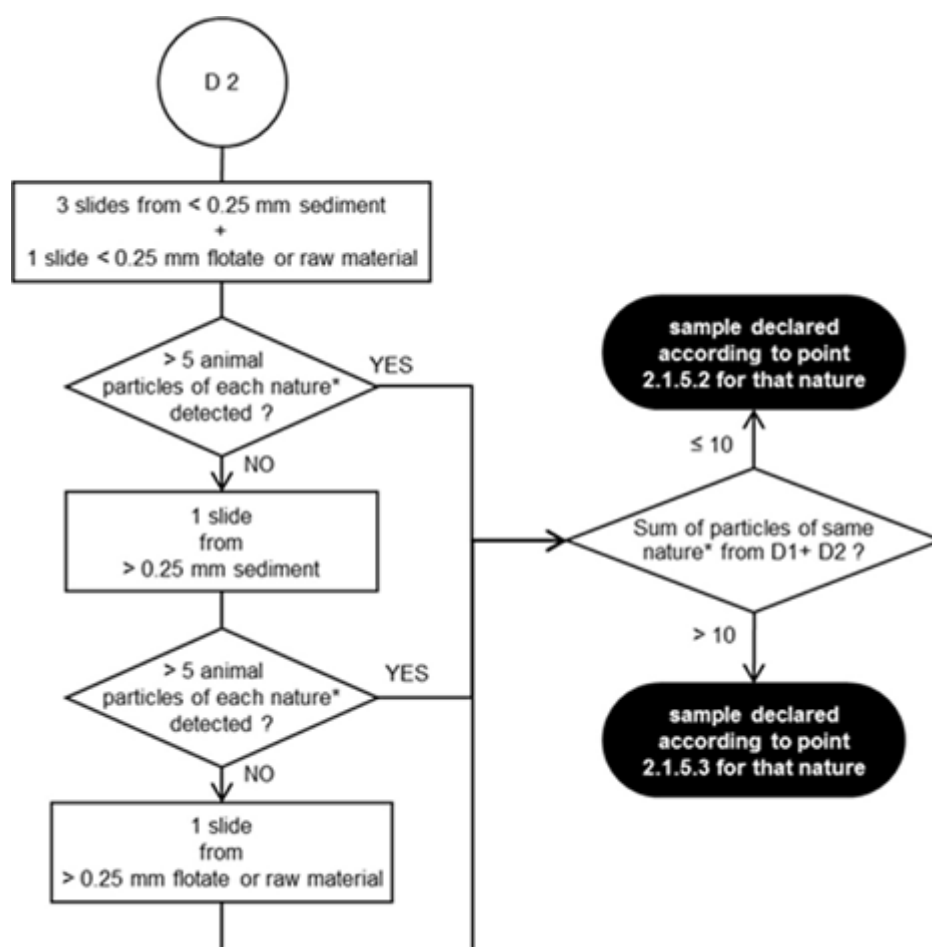
When additional slides are prepared on the flotata or the raw material using a more specific mounting medium with staining properties, as laid down in point 2.1.2.1.4, to further characterise structures (e.g. feathers, hairs, muscle or blood particles) which have been detected on slides prepared by other mounting media, as laid down in point 2.1.2.1.3, the number of particles shall be counted based on a number of slides per determination not exceeding 6, including the additional slides with a more specific mounting medium.

In order to facilitate the identification of the particles' nature and origin, the operator may use support tools like decision support systems, image libraries and reference samples.

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(12) point 2.1.4.3 is replaced by the following:

Number of determinations

Determinations shall be performed on different sub-samples of 50 g each.

If following the first determination carried out in accordance with the observation flowchart laid down in diagram 1, no animal particles are detected, no additional determination is necessary and the result of the analysis shall be reported using the terminology laid down in point 2.1.5.1.

If, following the first determination carried out in accordance with the observation flowchart laid down in diagram 1, one or more animal particles of a given nature (i.e. terrestrial vertebrate or fish) are detected, and the nature of the particles found confirms the declared content of the sample, no second determination is necessary. If the number of the animal particles of a given nature detected during this first determination is higher than 5, the result of the analysis shall be reported per animal nature using the terminology laid down in point 2.1.5.3. Otherwise, the result of the analysis shall be reported per animal nature using the terminology laid down in point 2.1.5.2.

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In other cases, including when no declaration of content has been provided to the laboratory a second determination shall be carried out from a new sub-sample.

If, following the second determination carried out in accordance with the observation flowchart laid down in diagram 2, the sum of the animal particles of a given nature detected over the two determinations is higher than 10, the result of the analysis shall be reported per animal nature using the terminology laid down in point 2.1.5.3. Otherwise, the result of the analysis shall be reported per animal nature using the terminology laid down in point 2.1.5.2.

(13) point 2.1.5 is replaced by the following:

Expression of the results

When reporting the results, the laboratory shall indicate on which type of material the analysis has been carried-out (sediment, flotate or raw material). The reporting shall clearly indicate how many determinations have been carried-out and if sieving of the fractions prior to slide preparation, in accordance with the last paragraph of point 2.1.3.3.4., was not performed.

The laboratory report shall at least contain information on the presence of constituents derived from terrestrial vertebrates and from fish.

The different situations shall be reported in the following ways.

2.1.5.1. No animal particle of a given nature detected:

- “As far as was discernible using a light microscope, no particle derived from terrestrial vertebrates was detected in the submitted sample.”
- “As far as was discernible using a light microscope, no particle derived from fish was detected in the submitted sample.”

2.1.5.2. Between 1 and 5 animal particles of a given nature detected when only one determination has been performed, or between 1 and 10 particles of a given nature detected in case of two determinations (the number of detected particles is below the decision limit established in the standard operating procedures (SOP) of the EU reference laboratory for animal proteins in feedingstuffs (EURL-AP) and published on its website⁽¹⁾):

When only one determination has been performed:

- “As far as was discernible using a light microscope, no more than 5 particles derived from terrestrial vertebrates were detected in the submitted sample. The particles were identified as ... [bone, cartilage, muscle, hair, horn...]. This low level presence is below the decision limit established for this microscopic method.”
- “As far as was discernible using a light microscope, no more than 5 particles derived from fish were detected in the submitted sample. The particles were identified as ... [fishbone, fish scale, cartilage, muscle, otolith, gill...]. This low level presence, is below the decision limit established for this microscopic method.”

When two determinations have been performed:

- “As far as was discernible using a light microscope, no more than 10 particles derived from terrestrial vertebrates were detected over the two determinations in the submitted sample. The particles

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were identified as ... [bone, cartilage, muscle, hair, horn...]. This low level presence is below the decision limit established for this microscopic method.”

- “As far as was discernible using a light microscope, no more than 10 particles derived from fish were detected over the two determinations in the submitted sample. The particles were identified as ... [fishbone, fish scale, cartilage, muscle, otolith, gill...]. This low level presence is below the decision limit established for this microscopic method.”

Additionally:

- In case of sample pre-sieving, the laboratory report shall mention in which fraction (sieved fraction, pelleted fraction or kernels) the animal particles have been detected insofar as the detection of animal particles only in the sieved fraction may be the sign of an environmental contamination.
- When only animal particles which cannot be categorised as either terrestrial vertebrates or fish are detected (e.g. muscle fibres), the report shall mention that only such animal particles were detected and that it cannot be excluded that they originate from terrestrial vertebrates

- 2.1.5.3. More than 5 animal particles of a given nature detected when only one determination has been performed, or more than 10 particles of a given nature detected in case of two determinations:

When only one determination has been performed:

- “As far as was discernible using a light microscope, more than 5 particles derived from terrestrial vertebrates were detected in the submitted sample. The particles were identified as ... [bone, cartilage, muscle, hair, horn...].”
- “As far as was discernible using a light microscope, more than 5 particles derived from fish were detected in the submitted sample. The particles were identified as ... [fishbone, fish scale, cartilage, muscle, otolith, gill...].”

When two determinations have been performed:

- “As far as was discernible using a light microscope, more than 10 particles derived from terrestrial vertebrates were detected over the two determinations in the submitted sample. The particles were identified as ... [bone, cartilage, muscle, hair, horn...].”
- “As far as was discernible using a light microscope, more than 10 particles derived from fish were detected over the two determinations in the submitted sample. The particles were identified as ... [fishbone, fish scale, cartilage, muscle, otolith, gill...].”

Additionally:

- In case of sample pre-sieving, the laboratory report shall mention in which fraction (sieved fraction, pelleted fraction or kernels) the animal particles have been detected insofar as the detection of animal particles only in the sieved fraction may be the sign of an environmental contamination.

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- When only animal particles which cannot be categorised as either terrestrial vertebrates or fish are detected (e.g. muscle fibres), the report shall mention that only such animal particles were detected and that it cannot be excluded that they originate from terrestrial vertebrates.

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(1) <http://eurl.craw.eu/>

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