



Determination of Rice chaffs (*Oryza sativa*) in Feedstuff, IAG-Method A9



International Association of Feedstuff Analysis
Section Feedstuff Microscopy



1. Objective and field of application

The method is used for both qualitative and quantitative estimation of Rice chaffs (*Oryza sativa* ?) in animal feedingstuff.

2. Principle

Rice chaff content is determined by macroscopic and microscopic identification of the chaffs and chaff fragments. Quantification is done by weighing the amount of identified chaffs and fragments in sieved fractions of the sample.

3. Reagents

3.1 Embedding agents

3.1.1 Chloral hydrate ($\beta = 60\%$)

3.1.2 Water

The reagents listed may be replaced by others which yield comparable results.

4. Equipment and accessories

4.1 Optical equipment

4.1.1 Stereo microscope (up to 70x magnification); recommended additional equipment: image support system

4.1.2 Compound microscope (up to 400x magnification); recommended additional equipment: polarization, phase contrast, image support system

4.1.3 Magnifier (up to 10x magnification)

4.2 Mortar and pestle

4.3 Sieve fitted with square meshes of width of 0.5 mm

4.4 Analytical balance (accuracy 0.001 g)

4.5 Additional laboratory equipment is listed in supporting document (9)

4.6 Reference material

5. Procedure

The examination is performed in non-pelleted feedingstuff. Pelleted feedingstuff have to be depelleted before examination (4.2; 8.1).

Qualitative determination of the rice chaffs is performed macroscopically and microscopically considering rice chaffs and rice chaff fragments in both the sieve fractions > 0.5 mm and ≤ 0.5 mm.



Quantitative estimation is performed by selecting and weighing of rice chaffs and rice chaff fragments with a particle size > 0.5 mm out of the laboratory sample or an aliquot of it.

5.1 Preparation of the laboratory sample

Non-pelleted feedstuff (at least 10g) are weighed (4.4) and fractionated according to supporting document (9., 5.3.1). The obtained fractions > 0.5mm and ≤ 0.5mm are weighed (4.4).

5.2 Identification of rice chaffs

Rice chaffs are identified based on characteristic features of both the whole chaffs and their fragments. The identification of rice chaffs and rice chaff fragments may be facilitated through comparison with reference material (4.6) and existing descriptions (10).

Lemma and palea of rice can be distinguished from those of other types of grains by their longitudinal and transversal striation. Using a compound microscope (4.1.2) one can recognise the deeply emarginated and highly silicified epidermal cells, up to 500 µm long hairs and a hypodermis that consists of 2-3 rows of fibre cells. The parenchyma and the inner epidermis have very thin cell walls. Fragments of rice chaffs can be easily recognised in polarised light.

5.3 Quantitative estimation of the rice chaff content

The quantitative estimation of rice chaffs and fragments is performed using the sieve fractions > 0.5 mm. Material identified as rice chaff fragments is separated from the sample and weighed (4.4). The result is expressed as milligram rice chaffs and fragments per kilogram feedstuff (mg/kg) (6.1).

An aliquot of the fraction may be used if necessary.

6. Calculation and report

6.1 Calculation

The amount of rice chaffs and fragments in mg/kg feedstuff (original sample) is calculated using the following formula:

$$C = \frac{BC \times 1000}{E} \text{ [mg/kg]}$$

C = amount of rice chaffs and fragments in mg/kg feedstuff

BC = selected rice chaffs and fragments in the examined fraction or an aliquot of it [mg]

E = total weight of the laboratory sample or an examined aliquot of the laboratory sample [g]



6.2 Report

6.2.1 Negative result:

As far as was discernible using a microscope, rice chaffs or rice chaff fragments (*Oryza sativa* ?) were not found in the submitted sample.

6.2.2 Positive result:

As far as was discernible using a microscope ... mg rice chaffs or rice chaff fragments (*Oryza sativa* ?) /kg feedstuff were found in the submitted sample. For quantitative estimation rice chaffs and rice chaff fragments > 0.5 mm are considered.

6.2.3 Possible supplement to the report:

The sieve fraction > 0.5 mm amounts to xx % of the laboratory sample.

7. Validation

not applicable

8. Remarks

- 8.1 For the identification of rice chaffs in pelleted feedstuff the use of supporting document (9.; 5.2.2.2 or 5.2.2.3) is recommended for depelleting the sample.
- 8.2 According to their high content of silicates, rice chaffs should not be fed to animals
- 8.3 This method is also recommended for examining food and unprocessed feedstuff.

9. Supporting document

Sample Preparation for the Macroscopic and Microscopic Analysis, IAG-Method A1

10. Literature

- FREUND, H. (ed.), 1976: Handbuch der Mikroskopie in der Technik. Band VIII: Mikroskopie der Nahrungs- und Futtermittel, der Drogen und Genußmittel. Umschau-Verlag, Frankfurt/ Main
- GASSNER, G.; HOHMANN, B. UND DEUTSCHMANN, F., 1989: Mikroskopische Untersuchung pflanzlicher Lebensmittel. 5. Auflage, Gustav Fischer-Verlag, Stuttgart, New York, 66-70



- HAHN, H. UND MICHAELSEN, L., 1996: Mikroskopische Diagnostik pflanzlicher Nahrungs-, Genuß- und Futtermittel, einschließlich Gewürze. Springer-Verlag, Berlin, Heidelberg, New York
 - HUSS, W., 1969: Methode zur Bestimmung von Hülsen, Spelzen und Schalen. In: Tagungsprotokoll der Internationalen Arbeitsgemeinschaft für Futtermitteluntersuchung, Sektion Futtermittelmikroskopie, Münster
 - HUSS, W., 1970: Quantitative Bestimmung von Schalen, Hülsen, Spelzen in Pressfuttermitteln. In: Tagungsprotokoll der Internationalen Arbeitsgemeinschaft für Futtermitteluntersuchung, Sektion Futtermittelmikroskopie, Maastricht
 - HUSS, W., 1971: Quantitative Bestimmung von Haferspelzen in Mischfutterpellets durch Auslesen. In: Tagungsprotokoll der Internationalen Arbeitsgemeinschaft für Futtermitteluntersuchung, Sektion Futtermittelmikroskopie, Würzburg
 - MALKOMESIUS, E.; Nehring, K.; Claus, G. und Kummer, H., 1951: Die Untersuchung von Futtermitteln. In: Handbuch der landwirtschaftlichen Versuchs- und Untersuchungsmethodik (Methodenbuch, Band III, 2. Auflage, Hrsg. R. Herrmann), Neumann-Verlag, Radebeul und Berlin
 - MÉSZAROS, L. und Bihler, E., 1975: Atlas für die Mikroskopie von Nahrungsgrundstoffen und Futtermitteln. Teil 2: Stärkereiche Nahrungsgrundstoffe und deren Verarbeitungsprodukte. In: Handbuch der landwirtschaftlichen Versuchs- und Untersuchungsmethodik (Methodenbuch, Band XI, Hrsg. L. Schmitt), Verlag Neumann-Neudamm, Melsungen
 - RÖZSE, E., 2005: Atlas of Feed Microscopy. Part II: The Examination of Non-desired Materials in Feed. National Institut for Agricultural Quality Control, Budapest
 - VÖHRINGER, H., 1997: Leitfaden für die Futtermittel-Qualitätskontrolle durch Mikroskopie. Hoffmann-La Roche, 3. Fassung, Wien
- 10.1 This method has been developed by the International Association of Feedingstuff Analysis (IAG) – Section Feedingstuff Microscopy.
-