



## **Method for the Determination of poisonous plants in roughage, IAG-Method A8**



International Association of Feedstuff Analysis  
Section Feedstuff Microscopy



## 1. Objective and field of application

The method is used for the qualitative determination and estimation of poisonous plants in roughage.

Roughage is defined as fresh grass, hay, haylage and straw material.

## 2. Principle

Poisonous plants in roughage are determined by macroscopic and/or microscopic identification using the morphologic characteristics. For estimating their amount in the sample, the identified plant material in the roughage is weighed.

## 3. Reagents

3.1 Staining reagent

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

## 4. Equipment and accessories

4.1 Optical equipment

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

4.1.2 Magnifier (up to 10 x magnification)

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

4.2 Analytical balance (accuracy 0.01 g)

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

4.4 Reference material

## 5. Procedure

*From the plant material all those plant parts are selected which show the morphologic characteristics of poisonous species.*

5.1 Identification

Important for the identification of the plant material is the habitus of the plant parts in a whole (length of the stem, branching, edged or grooved stem etc.), as well as obtained parts of leaves, flowers and fruits.



For the identification stereomicroscope (4.1.1) and a magnifier (4.1.2) are used  
Suspicious shrivelled plant material can be conditioned in hot water before starting the morphologic identification. In several cases this method allows better identification, because sometimes the material returns into its natural plant habitus.

For the identification of poisonous plants: literature, annex descriptions and reference material is used.

## 5.2 Quantitative estimation

At least 500 g of an original sample is used to select all plant parts of suspicious plants. The whole examined sample and the selected plant parts are weighed separately (4.2)

## 6. Calculation and report

### 6.1 Calculation

The amount of selected plant material in g/kg feedingstuff (original sample) is calculated using the following formula:

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

**C** = amount of component in feedingstuff [g/kg]

**BC** = selected fragments of component in the examined sample or an aliquot of it [g] with a precision of 0.1 g

**E** = total weight of the examined sample of it [g] with a precision of 0.1 g



## 6.2 Report

### 6.2.1 Negative result:

As far as was discernible by macroscopic and/or microscopic investigation no poisonous plant material/no plant material of X was found in the submitted sample.

### 6.2.2 Positive result:

As far as was discernible by macroscopic and/or microscopic investigation plant material of X was found in the submitted sample. The amount was estimated to x g/kg.

## 7. Validation

Not applicable

## 8. Remarks

- 8.1 Identification and quantitative estimation of poisonous plants are possible, if fresh and whole plants are present in the sample. In hay, especially in hay with a high degree of mechanic abrasion or crumbling, identification is more difficult as in most cases only small plant fragments are found. Nevertheless the finding of vegetative characteristics might enable identification. It is useful to know in which month the hay was harvested. Then conclusions can be drawn from the growth stage of the plants, e.g. flowering.
- 8.2 In case suspicious plant material is found, it is advisable to do further toxicological analysis, e.g. to look for pyrrolizidine alkaloids when suspect *Senecio* spp. are present.

## 9. Supporting document

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

## 10. Literature (exemplary)

FROHNE, D., JENSEN, U., 1998: Systematik des Pflanzenreichs; unter besonderer Berücksichtigung chemischer Merkmale und pflanzlicher Drogen. 5. Auflage, wissenschaftliche Verlagsges., Stuttgart

FROHNE, D., PFÄNDER, H. J., 2005: Poisonous Plants; A handbook for Doctors, Pharmacists, Toxicologists, Biologists and Veterinarians. Manson publishing Ltd., London

ROTH, L.; DAUNDERER, M. und KORMANN, K., 1994: Giftpflanzen-Pflanzengifte. 4. Auflage, ecomed verlagsgesellschaft, Landsberg



Rothmaler, W., 2009: Exkursionsflora von Deutschland, Band 2: Gefäßpflanzen Grundband, 19. Auflage; Band 3: Gefäßpflanzen Atlasband, 11. Auflage; Spektrum Akademischer Verlag, Heidelberg

LINKS:

[http://www.vetpharm.uzh.ch/reloader.htm?perldocs/toxysqry.htm?inhalt\\_c.htm](http://www.vetpharm.uzh.ch/reloader.htm?perldocs/toxysqry.htm?inhalt_c.htm)

## 11. Annex of some frequently occurring poisonous plants in European roughage

- 11.1 *Senecio* sp. is a genus of the Asteraceae family. The botanical name is deduced from the Latin word senex (= old person), probably, because of the white hairy pappus of the *Senecio* fruit, that looks like hairs of old people. *Senecio* is one of the species-richest genera of flowering plants. *S. jacobaea* L. (Tansy Ragwort, Common Ragwort), often found on pastures, is being named as the representative for many *S.* species. The plant is 30-100 cm tall and shows feathered leaves. The stem is strong and branched. The golden-yellow disc-and radiant flowers yield uphold umbel clusters. Flowering: July-August/September.

The genus is known for containing pyrrolizidin alkaloids, which are changed into metabolites in the liver. These react irreversibly with DNA and other macro molecules, causing liver damage or even the death of the animals. Important is the total amount of pyrrolizidin alkaloid intake, no matter of time. All parts of the plant, fresh as well as dried, are toxic.

- 11.2 Common St. Johnswort (*Hypericum perforatum* L.) belongs to the Hyperaceae family. The plant is 30-60 cm tall. The leaves are oval and standing towards each other. The uphold stem shows two exalted length edges. The flowers are golden-yellow with asymmetric, black dotted crown leaves. They yield rich flowering clusters. The calyx and crown leaves consist of black, lined and tapped oil glands, which cause red colouring when grinded. Flowering: June-September/October.

Common St. Johnswort contains hypericin. This chemical causes continuous photosensibility of the skin. Horses and other farm animals are highly sensitive. Unpigmented or poorly haired skin areas can be damaged.

Qualitative test for hypericin: A plant part with dark spots is moistened with a drop of chloral hydrate (3.1.1). A dark red area is observed, if the dark spots are secretion glands.

- 11.3 Autumn Crocus (*Colchicum autumnale* L.) belongs to the Liliaceae family and mainly occurs in meadows. Its purple or pink flowers appear from August until October. The leaves are lanceolate, 20 - 30 cm long and appear from early



spring until July bearing the seed capsules between them. They contain small brownish-black seeds with a sticky appendix.

All plant parts contain colchicine and other poisonous alkaloids. Fresh plants as well as dried are toxic. Colchicine inhibits mitosis and cell division and damages the capillary vessels. It causes sickness, diarrhoea and paralysis of the central nervous system. Death occurs due to respiratory arrest. There is a cumulative effect if several low doses of colchicine are taken in.

- 11.4 Tansy (*Tanacetum vulgare* L.) belongs to the Asteraceae family. The plant is 60 – 120 cm tall and has a remarkable odour, somewhat like camphor. The leaves are alternate and feathery. The flower heads look like yellow, flat buttons, growing in clusters from July to October.

The whole plant contains volatile oils; first of all, the neurotoxic thujone. Ingestion of Tansy could cause *obnubliation*, mucosa irritation, kidney- and liver damage.

- 11.5 *Ranunculus* sp. is a genus of the family Ranunculaceae. Most plants of this genus are perennial herbs and have shiny, bright yellow flowers.

One representative is the Meadow Buttercup (*Ranunculus acris* L.). The herb is 30 – 100 cm tall with flowers from May to September. The flowers have 5 glossy, yellow, overlapping petals. The leaves are digitated and deeply cut.

Fresh and ensilaged plants are toxic. They contain protoanemonin, which can cause severe skin and gastrointestinal irritation. While drying, protoanemonin dimerizes to the non toxic anemonin. As *Ranunculus* has an acrid taste, intoxication of livestock is uncommon, but could happen where overgrown pastures occur.

This method has been developed by the International Association of Feedstuff Analysis (IAG) – Section Feedstuff Microscopy