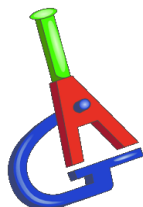


# Newsletter

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## President's address

As the (still) president of the IAG feed Microscopy Association, I am pleased to address this letter to all our members.

The board and active members are pleased to keep up the long-lived informal IAG association with such essential tasks as organizing and highlighting 3 Proficiency Tests (on composition, impurities and animal proteins), hosting a rewarding Annual Conference and Workshop (in Namur in 2024) and modernizing our communication and functioning.

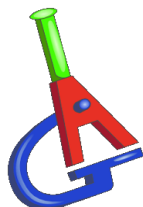
On the topic of animal proteins, IAG Feed microscopy collaborates closely with the EURL-AP. The members of the IAG have an essential role to play as they are the actors at the front, the day-to day analysts who must apply in routine the harmonized protocols with high efficiency, security and exactness. Often, they also are the interface between production and control; common-sense and acute knowledge shall be combined to advice competent authorities as well as stakeholders.

The feed and food microscopists can help in the field of checking the composition and purity, detecting fraud, estimating problematic alteration, or contaminations.

Knowing how feedingstuffs and feed ingredients are produced, handled and combined, disclosing how they look like, what they are made of, and may involuntarily contain, but also understanding some of their physical properties, is as important as having some ideas about their nutritional and, finally, economical value.

Having this combination of knowledge helps to apprehend new problems and, sometimes, solve a puzzle.

Organising IAG Annual Meetings and workshops, as well as keeping the network active, performing PTs, and searching for training possibilities is the task of the association. Doing so, it offers to all our members, from the technician to the professor, the possibility to increase this combination of knowledge.



Finally, sharing experiences and problems is making our professional life more interesting and, hopefully, easier.

Please enjoy reading of this Newsletter which is an update on our speciality and activities!

See you in Fribourg in May 2025

Yours sincerely

Geneviève Frick, IAG president

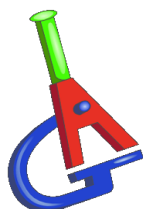
## IAG Meeting 2025 in Fribourg, Switzerland

Dear Colleagues,

We are delighted to invite you to the IAG Annual Meeting 2025 co-organised by the IAG board and Agroscope in Switzerland. The meeting will take place in the city of Fribourg and at the Agroscope Institute in Posieux from 20 to 22 May 2025.

The registration to the meeting is yet open by way of the online [registration form](#) until **1 April 2025** as a deadline.





## IAG Proficiency tests in 2024

### IAG PT 2024 Determination of the composition of a mixed feed for dairy cows

The ring test was organized as the annual proficiency test for the composition of feeding stuffs by microscopy. It was presented by the IAG (International Association for Feeding stuffs Analysis, Section Feeding stuffs microscopy) and organized by the LUFA Nord-West in Oldenburg. In particular this ring test is about the determination of the composition of a mixed feed for ruminants. One mixed feed for dairy cows (Sample No. 1/2024) was offered to the participants. The sample had to be analysed according to the IAG method A2 'Method for the Identification and Estimation of Constituents in Animal Feeding stuff'.

17 laboratories from nine different countries (Austria, Belgium, Denmark, France, Germany, Poland, Slovak Republic, Switzerland, The Netherlands) participated in the ring test

#### Material:

The sample (Sample No. 1/2024) we offered to the participants of the ring test 2024 is a commercially available mixed feed for dairy cows, which was provided by a local producer of feeding stuffs. The sample was sent as a blank sample, that means without any declaration.

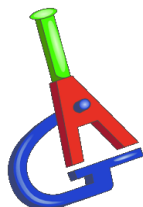
Raw material	Inclusion %
Rape seed products	44.0
Maize products	28.5
Beet pulp	10.0
Rye products	4.9
Wheat products	7.6

Tab 1: Declaration used for evaluation of results

Table 1 presents the summarized declaration as it is used for the evaluation of the results. The individual components were summarized to facilitate the evaluation. For example, maize stillage and maize were combined to maize products.

#### Results:

For detailed information of the single results of the participants please have a look at the report of the IAG-PT Composition, you can find it on the IAG-homepage in the member area → ["Reports"](#)



## Evaluation:

This IAG proficiency test was evaluated based on the uncertainty limits as they were decided in 2006 on the annual IAG meeting in Rostock (Tab 2).

Range	Uncertainty limits
< 2.0 %	traces
2.0 - 5.0 %	+/- 100 % r
5.1-10.0 %	+/- 5 % a
10.1-20.0 %	+/- 50 % r
20.1-50.0 %	+/- 10 % a
> 50 %	+/- 20 % r

Tab 2: Uncertainty intervals developed by IAG 2006 in Rostock.

Find the statistical summary in the enclosed table 3

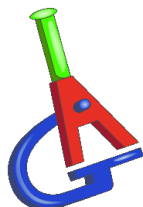
Ingredients	Target %	Range %	Min. %	Max. %	Average %	Median %	Over-estimation %	Under-estimation %	Correct %
Rapeseed products	44.0	34.0 – 54.0	15.0	70.4	36.7	31.0	17.6 (3)	52.9 (9)	29.5 (5)
Maize products	28.5	18.5 – 38.5	10.5	27.5	17.0	15.0	0.0 (0)	76.5 (13)	23.5 (4)
Beet pulp	10.0	5.0 – 15.0	5.0	30.0	14.6	15.0	47.1 (8)	0.0 (0)	52.9 (9)
Wheat products	7.6	2.6 – 12.6	0.0	33.0	16.0	17.1	47.1 (8)	5.9 (1)	44.2 (7)
Rye products	4.9	0.0 – 9.8	0.0	33.0	11.0	10.0	47.1 (8)	0.0 (0)	47.1 (8)

Tab 3: Statistics of results

Due to the nature of the sample, the depelletization of the pellets resulted in very fine sample material with a high proportion in the fraction < 0.5 mm. Sorting the sample was therefore only possible to a limited extent for a small part of the sample, as the vinasse and molasse caused the individual pieces to stick together strongly. A large part of the sample therefore had to be estimated.

## Summary:

This report shows the results of the IAG ring test 2024 "Determination of the composition of a mixed feed for dairy cows". One mixed feed for dairy cows (Sample No. 1/2024) was offered to the participants. The sample was sent without any declaration und had to be analysed according to IAG Method A2 'Method for the Identification and Estimation of Constituents in Animal Feeding stuff'. 17 laboratories participated in this proficiency test. The uncertainty intervals were calculated



according to the IAG Model decided on the annual IAG meeting in 2006 in Rostock. The sample seems to have been a difficult one for most of the participants. Only two of the 17 participants estimated the amounts of the included components correctly.

Many thanks to the **team of LUFA Nord-West in Oldenburg** organizing this interesting IAG PT on Composition 2024.

In the name of the IAG board the association is already looking forward to the presentation of the results of the upcoming IAG PT 2025 which will be on the annual meeting in Fribourg/Posieux (Switzerland)

## IAG Proficiency test on animal proteins in feed 2024

Since decades the International Association for Feedingstuff Analysis (IAG) section Feed Microscopy has been organizing collaborative studies for the evaluation of composition and the detection of animal constituents in feed. Since 2023 the board subcontracts this organization to the Walloon Agricultural Research Centre CRA-W (Gembloux, Belgium).





The CRA-W is notably hosting the European Union Laboratory for Animal Proteins detection in feedingstuff (EURL-AP), is ISO/CEN 17025 accredited for light microscopy and PCR detection of animal constituents in feed and ISO/CEN 17043\* accredited for the organization of proficiency tests for the detection of animal constituents.

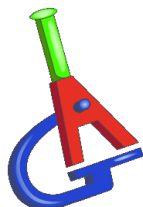
The present proficiency test was organized in the next framework. The use of processed animal by-products as ingredient for animal feedingstuffs within the European Union is regulated by the TSE Regulation EC N°999/2001 (European Commission, 2001), as amended. In particular, Article 7 imposes a prohibition to use processed animal proteins (PAPs) in the feeding of farmed animals (extended feed ban).

The objective of the present study was to assess the performance of the participants to detect the presence of PAPs in feed by the reference method using light microscopy as stated in Annex VI of Regulation EC 152/2009 (European Commission, 2009) imposing the methods of analysis for the determination of constituents of animal origin for the official control of feed as recently amended by Commission implementing Regulation (EU) No 2022/893 (European Commission, 2022) and related SOPs

### Sample set material and preparation

Four different sample materials were prepared for the study. Each participant received one entity of 40 g of each material. The sample set consisted of:

-  1 = Poultry feed
-  2 = Poultry feed + 0.1 % of bovine PAP + 0.1 % of bovine hairs
-  3 = Poultry feed + 1 % of Tenebrio molitor PAP
-  4 = Aquafeed



Both matrices, the poultry feed and the aquafeed, were commercial feeds from EU origin. The poultry feed was a complete feed for broilers and the aquafeed was a complete feed for trout. The poultry feed contained only plant material and minerals while the aquafeed contained poultry meal. The bovine PAP was a dedicated batch of produced material with a bone content of about 61 %. The bovine hairs were collected, cleaned and crushed into liquid nitrogen for size reduction prior to use. The insect \* The present study is outside the ISO/CEN 17043 accreditation Page 4 on 18 PAP used was a commercial insect protein meal produced from *Tenebrio molitor*; the meal was free from any other trace of animal origin (tested by light microscopy).

## Participants

A maximum of 60 participants was planned and in the end 56 registrations were recorded. From the 56 registered participants (Annex 1), 53 participants delivered their results. Among the 53 sets of received results, one was refused since the results were obtained by PCR and not by light microscopy. The present study is thus based on a final result set from 52 participants

## General results

Table 1 is summarising the results from all participants.

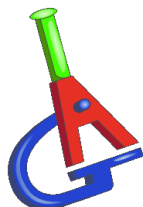
Table 1: General results; Numbers in brackets refer to the number of errors.

	TERR. VERT.		TERR. INVERT.		FISH		Global AC
	SE	SP	SE	SP	SE	SP	
1 = Poul feed		1.000		0.863 (7)		0.962 (2)	0.942
2 = Poul feed + 0.1% PAP + 0.1 % hair	0.962 (2)			0.941 (3)		0.808 (10)	0.903
3 = Poul feed + 1 % TM		0.981 (1)	0.863 (7)			0.962 (2)	0.935
4 = Aquafeed	0.962 (2)			1.000	0.962 (2)		0.974
AC	0.976		0.917		0.923		

Global accuracies range from 0.903 to 0.974 which are plenty satisfactory. Although possible improvement might be still expected (e.g. sample 2), it demonstrates the ability of a large majority of participants to implement the light microscopic method for the detection of animal remains in feed. Further insights will be presented in the detailed sample review.

About the three parameters studied (terr. vert., terr. invert. and fish), the results showed the best accuracy score for the detection of terrestrial vertebrates' materials (AC of 0.976 with only 5 errors), followed by the detections of fish material and terrestrial invertebrates, at very closed accuracy rates (respectively AC of 0.923 with 16 errors and AC of 0.917 with 17 errors).





The total number of errors was of 38 which represents a total rate of 6.1 % over all delivered results. Among this total number of errors, specificity issues were predominant: 14 PD for fish, 10 PD for terrestrial invertebrates and finally only 1 PD for terrestrial vertebrates.

Sensitivity issues were also noted. For terrestrial invertebrates, 7 ND were found. Failing at disclosing the presence of terrestrial vertebrates occurred also (4 ND) as well as for fish (2 ND).

For more detailed information of the single results of the participants please have a look at the report of the IAG-PT on animal proteins, you can find it on the IAG-homepage in the member area → ["Reports"](#)

## Discussion and conclusion

As from last year (Veys & Fumière, 2023), the study only focused on the assessment of the participating laboratories regarding the detection of animal remains in feed by light microscopy. Investigations on method parameters were excluded from the scope of the proficiency test. The reason is that according to the instructions EU participants were only asked to stick to the official method as per Annex VI of EC 152/2009 regulation in its current version and related SOPs, on the exception of specific derogations as stated in the instructions, while non-EU participants were not bound to this condition. The two derogations to the legal method were involving the conditions for grinding due to the limited amount of sample contained into each entity and the mandatory application of both single TCE and double PE/TCE sedimentations on all samples.

The sample set composition of the study aimed at mimicking real laboratory practice by using common feed matrices and commercially available sources of PAPs at realistic concentration levels.

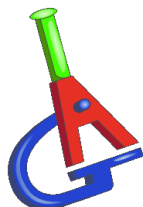
The global accuracies for the samples ranged from 0.903 to 0.974 which is mostly satisfactory and in line with past studies.

The detection capabilities of terrestrial vertebrates' material scored the best as confirmed by the parameter AC of 0.976, with only 5 errors. Nevertheless, the mention of animal hair presence is still subject to improvement.

Regarding the accuracies for terrestrial invertebrates' material (AC of 0.917 with 17 errors) both sensitivity and specificity problem are evenly impacting the performance of the network of participants. When compared to last year IAG proficiency assessment (Veys & Fumière, 2023) where the AC for that parameter was of 0.896, a trend to improvement is observed.

The accuracy related to fish material (AC of 0.923 with 16 errors) revealed that specificity problems were largely predominant over the sensitivity issues. This situation, associated with misidentifications, was not observed last year. Possible





explanations may be linked with the type of matrix used, the poultry feed, or the adulterant from sample 2, the bovine PAP. In 2016 a similar situation was noted in an EURL-AP proficiency test (Fumière et al., 2017) with a poultry feed containing seashell grits generating misidentification with fishbones. However, this assumption does not explain the rise of PD for fish into sample 2 only. Some bone particles from the used bovine PAP appeared as more elongated than usual with compact bone osteocytes with bilateral irradiating lacunae network responsible for a potential “fishy” aspect. This assumption is more plausible. Whether or not it may fully explain the current specificity issue is impossible, but it highlights the need for a broad histological knowledge, the key to avoid confusions leading to false positive results

Also on this side many thanks to the **team of the EURL-AP/CRA-W in Gembloux** organizing this interesting IAG PT on animal proteins 2024.

In the name of the IAG board the association is already looking forward to the presentation of the results of the upcoming IAG PT 2025 which will be on the annual meeting in Fribourg/Posieux (Switzerland).

## IAG Meeting 2024 in Namur, Belgium

The IAG meeting 2024 was organised in Namur, Belgium from 5-7 June by the colleagues of the CRA-W. Many thanks for the organisation of the meeting.

A summary of the meeting is presented here:

05/06/2024
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### Highlights on the CRA-W (G. Sinnaeve)

The organisation of the CRA-W was presented as well as their different tasks.

### Tour de table, Feed Survey, special requests (G. Frick)

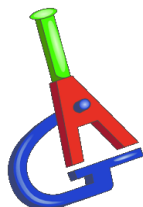
A round table was organised with cases and experiences of the participants.

### ISO 17025 accreditation for microscopy: what to expect? (J. Vancutsem)

An overview of the different chapters on accreditation of microscopic analyses was presented.

### Method of recover and microscopic evaluation of coarse fraction from granular and crumble feeds as the answer for question: Do we have whole grain inside pellets or crumble ? (P. Czajkowski)

A strategy on the preparation of samples for microscopic evaluation was presented. Methods for depelleting were proposed: examples for depelleting with a mortar and depelleting in water were illustrated.



Milk replacer. Microscopy as the proposition number one and base element to identification and control them "on plant" (P. Czajkowski)

Different strategies on the evaluation of milk replacer were proposed: visual, NIR, physical (solubility, centrifugation, microscopic). Images of blood plasma, hydrolysed wheat gluten, whey powder, whey protein concentrate, fat filled whey, wheat flour,... were presented.

Last reading of IAG method A10: Method for the detection and determination of macroscopically/microscopically detectable foreign substances in Feedingstuffs (R. Weiss)

A last reading of the method on foreign substances was done. The method has been published and approved.

IAG method A10: perspectives for validation and implementation (M. Zadavec)

The list of prohibited materials of Annex III of EU regulation 767/2009 was discussed. The IAG-publication on the monitoring of packaging materials was discussed.

06/06/2024
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IAG- AP PT results (P. Veys)

See earlier in the newsletter

Insect detection: natural contamination vs PAPs (A. Anselmo)

Several pictures of insect parts were presented. Several pest species were discussed (*Sitophilus*, *Tribolium*, *Ephestia*, *Acar*). When there is a natural contamination, different development stages are observed.

Matthews correlation coefficient (MCC) (P. Veys)

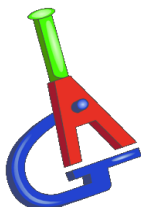
An alternative evaluation on sensitivity ( $\frac{TP}{TP+FN}$ ), specificity ( $\frac{TN}{TN+FP}$ ) and accuracy ( $\frac{TP+TN}{TP+TN+FP+FN}$ ) was presented. These are not suitable in the case of imbalanced classes. Alternatives as F<sub>1</sub>-score and MCC were discussed.

Artificial intelligence to detect bone fragments in microscopy images (G. van der Borg)

Experiments were set up for automated identification of bone fragments in sediments. A model has been developed that reached an AUROC (Area Under Receiver Operating Characteristic Curve) of 0.97. For the experiment a microfluidic channel has been developed where particles are flowing through.

AI: what to expect for the future (R. Weiss)

A first attempt on the differentiation of botanical impurities and the identification of animal proteins by using AI Zeiss Arivis Cloud was presented.



### Keyence presentation

A presentation and demonstration on digital microscopy was given.

### Practical workshop on botanical impurities

A workshop on botanical impurities was given.

07/06/2024
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### Past changes of feed ban: consequences (J. Vancutsem)

A historical overview on feed ban and the ABP-regulation was given. As current legislation exists of a compilation of several texts, a new building plan should be set up.

### What's new in legislation in food/feed microscopy (J. Vancutsem)

New legislation was presented: Implementing regulations 2024/771 and 2023/2782 on the analysis of ergot and botanical impurities. Also the differences between these regulations were presented. New legislation was published on the maximum levels of ergot in food in regulation 2023/915. A method on microplastics in water was published in delegated decision 2024/1441.

### Open discussion

An open discussion with following subjects was held:

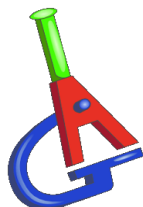
- Method / Protocol /sequence /AP for ruminant feed: the discussion was on the PE/TCE sedimentation
- Protocol for impurities (new legislation): it was discussed if the first analysis is > 2x the maximum level: is it still necessary to perform a 2<sup>nd</sup> analysis.
- Several questions on determination of certain particles by microscopy: there were questions on the use of polarization, the identification of blood products, invertebrates, fish and eggs.
- Dividing the sample when microscopy and PCR is needed (protocol?)

### Future proficiency tests and trainings

Following proficiency tests for 2025 are decided:

- A PT on animal proteins will be organised by CRA-W.
- A PT on composition of a compound feed will be organised by LUFA NW.
- A PT on botanical impurities/forbidden materials will be organised by AGES.
- A PT on packaging materials will be organised by WFSR.

The possibility of organising an external training on botanical impurities will be considered.



The possibility of organising a PT on toxic plants in hay will be considered.  
The possibility of organising a PT or training on fraud will be considered.

### IAG organisation and board selection

The election scheme was presented.

	2019	2021	2023	2025
President		election		election
Regulation affairs		election		election
Scientific officer	election		election	
Method reviser		election		election
Website manager	election		election	

There has been no re-election in 2024.

Julia Dietz changed of job and left the board.

Two new members joined the board: Lotte Hougs and Tommy Stojkovski.

We wish them a warm welcome to the board!

### Social Program during the IAG-meeting

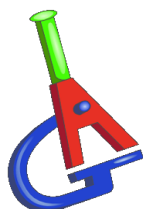
We had a fine guided tour in the citadel of Namur (Terra Nova) which we reached by cable car and the abbey of Marche-les-Dames where we had a nice dinner.



Citadel of Namur : Terra Nova



Abbey of Marche-les-Dames



## What's new in legislation in food/feed microscopy?

Commission Regulation (EU) 2024/1808 of 1 July 2024 amending Regulation (EU) 2023/915 as regards the application date of lower maximum levels for ergot sclerotia and ergot alkaloids in food was published.

Following a detailed examination of the information provided, it is concluded that the lower maximum levels are not yet achievable for ergot sclerotia in unprocessed rye grains (...) because of an increase in the prevalence of ergot sclerotia and ergot alkaloids in cereals due to climatic conditions.

It is therefore appropriate to defer the application of the lower maximum levels for ergot sclerotia in unprocessed rye grains for 1 year (...).

Annex I to Regulation (EU) 2023/915 is amended as follows:

Ergot sclerotia:

Unprocessed rye grains	Maximum level: 0,5g/kg - 0,2 g/kg as from 1 July 2025
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## IAG scientific contribution

### Scientific article in Italian Journal of Animal Science

The successful collaboration of 15 laboratories, primarily composed of IAG members, culminated in the publication of the article titled **"Survey among European and Canadian feed control units on monitoring packaging material residues in feed by microscopy analyses"**

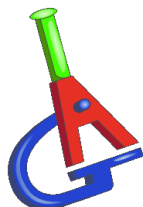
published in the Italian Journal of Animal Science

(<https://www.tandfonline.com/doi/epdf/10.1080/1828051X.2024.2370387?needAccess=true>).

Here is a summary:

Macro- and microscopic evaluation of feed includes detection of animal proteins, botanical ingredients and impurities, and prohibited ingredients such as packaging material (PM), according to Regulation (EC) 767/2009. In addition, detection of micro-plastics (possible degradation products of some of the PM) is getting attention. PM can harm animals or disturb their feed intake, pollute the environment, and are considered as undesired impurities in feeds. These materials do not consist of a definite molecule, group of molecules, living species or definite bodies. They can be plastic foil, hard plastic, metal pieces, paper, wood or some combination of materials. Their features (sharp, pointed) can be as important as the material itself. This is a typical topic for microscopy detection and evaluation. This short review presents the work done on detection of PM in 15 monitoring entities (institute, laboratories). Since 2011, some institutes have analysed more than 20 samples each year and the incidence of non-compliant samples will be presented here. Thirteen out of 15 entities have an active monitoring,





whereas others have passive surveillance (done while performing other microscopy analyses). The protocols used by the different entities depend on sample types and analysts, highlighting a need for harmonisation

## Case report

### A puzzling hay sample

*Geneviève Frick, Agroscope, Switzerland*

Last year, the Agroscope feed analysis group received a hay sample from a lot which seemed to be responsible for inducing sickness to horses. Several analyses were demanded, and the results led to the conclusion that no single cause could be determined for the problematic condition of this material.

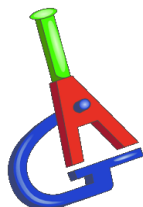
Here below, you will find the report and conclusions of the individual parameters. Some pictures are added for the visual part of the analyses.

Botanical impurity: general view of the fragments isolated from the hay



Numerous fragments of plant stems from the Apiaceae family: 16.9 g in 500 g analysed (3.38%). It is difficult to determine the species. There are poisonous plants in this family, notably hemlock, dwarf hemlock and poison hemlock, in the fresh state. Poisoning is possible by ingesting leaves from these species. No leaves were found, but if present, they are small and fragile when dried, and may not remain intact -nor selectable- in hay. It should be verified if the horses' symptoms can correspond with poisoning. Note that these stems are anyhow quite large and probably not desirable for horses to consume, even if they are not toxic.





### Mycotoxins:



Numerous grass leaves (26g in 500g) were observed with brown/black spots that could be due to bacterial infection or mould. Detection of mycotoxins was thought to be useful if the microbiological and chemical analyses did not identify the problem with the hay.

Analysis of 2 mycotoxins were done on the original sample and on the selected dotted leaves.

Results for Deoxynivalenol and Zearalenone: There are no recommended values for deoxynivalenol and zearalenone in hay or feed for horses. However, by comparing with the recommended values for other animals, it can be deduced that the levels found in this hay are not problematic. The concentrations were in the same order of magnitude in the selected leaves.

<b>Mycotoxin</b>	<b>Original sample</b>	<b>Selected leaves</b>
Deoxynivalenol	0.25 mg/kg	0.33 mg/kg
Zearalenone	10.00 µg/kg	<4.00 µg/kg

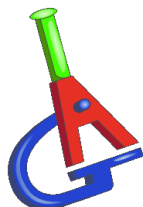
Soil fragments: 17.9 g in 500 g analysed (3.6%).  
Probably not a problem.



### Microbial quality:

The hay sample contains a fairly large number of mesophilic aerobic bacteria from group 2 (i.e. spoilage-indicating bacteria, which develops mainly during storage): 5.74E6 CFU/g, which represents an exceeding 2.9X the VDLUFA guideline value for hay (2E6 CFU/g for group 2 bacteria). This excess results in a microbiological quality score of "slightly reduced".





The yeast and mould content are unremarkable:

Product typical bacteria:  $6.38 \times 10^5$  cfu/g

Spoilage-indicating bacteria:  $5.74 \times 10^6$  cfu/g

Streptomyces: <1000 cfu/g

Moulds:  $5.52 \times 10^3$  cfu/g

Product typical moulds:  $2.76 \times 10^3$  cfu/g

Spoilage-indicating moulds:  $2.76 \times 10^3$  cfu/g

Mucorales: <100 cfu/g

Yeasts:  $4.44 \times 10^4$  cfu/g

Aerobic mesophilic germs:  $6.38 \times 10^6$  cfu/g

*Chemical and nutritional contents:*

All other results for this hay sample were considered to be normal.

Crude protein: 59.7 g/kg

Crude Fibre: 348 g/kg

Crude fat: 13.0 g/kg

Potassium: 14.8 g/kg

Magnesium: 1.08 g/kg

Manganese: 33.4 mg/kg

Sodium: 0.0950 g/kg

Phosphorus: 1.70 g/kg

Zinc: 18.1 mg/kg

Calcium: 3.92 g/kg

Copper: 4.54 mg/kg

Iron: 643 mg/kg

Crude ash: 72.3 g/kg

Water soluble carbohydrates : 95.8 g/kg

The laboratory is not expected to give any conclusion on the impact of the ingestion of the product, but even our knowledge on the quality and security of a feedingstuff was not sufficient to judge if a "cocktail" of different seemingly not highly significant spoilage could have a negative consequence on the physiology and health of horses.



## Microscopy as useful tool for the quality of feed

***Piotr Czajkowski, Reg Lab, Cargill Poland***

### Recover and evaluation of whole grains in pellets or crumbs

Digestibility of feedingstuffs is important for the animals and is part of the quality criteria for the farmers willing to buy a product.

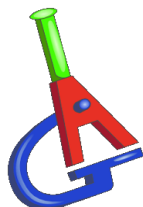
The feed producers may advertise on this criterion. Ingredients present as whole grains will show better digestibility if the grains are ground or, at least, broken.

How to check the proportion of broken grains in a finished pelleted or crumbed compound feed?



Most used whole grains:

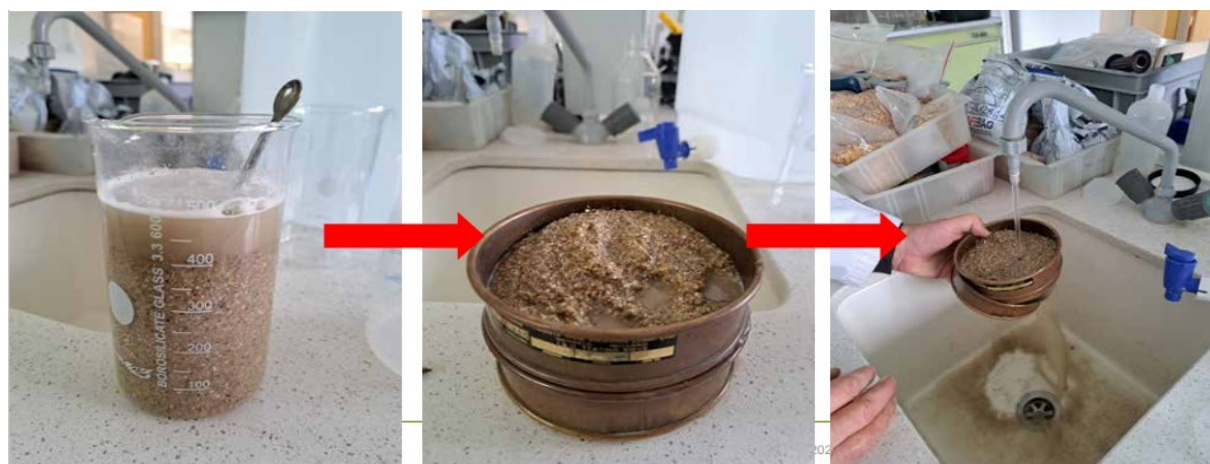




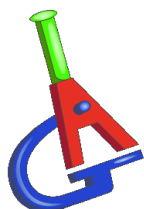
Method: 100 grams of pellets in 400-500 ml of water



After 1 hour when pellets have disintegrated in the water and no more visible pellet were observed → sieving was started under a running stream of water (a 2 mm sieve was used).







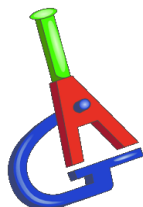
After complete drying, sieve again with 2mm sieve and analyse the coarse fraction



### Result of analysis:

			Result / 100 gram of sample	
nr lab	product feed for pig	Plnat	fraktion >2 mm	whole grain : quantity /100 g sample
1515...	Utmost Grower	1	5%	3 ( triticales - germ removed)
1515...	Utmost Starter	1	4,10%	3 (barley,rye,wheat)
1515...	Utmost Starter	1	4,40%	2 sztuki (rye)
1515...	Utmost Starter	1	4,30%	3 szutki ( barley,rye)
1516...	Utmost Finisher	2	2,60%	0
151...	Utmost Grower	2	2%	0
1516...	Utmost Finisher	2	2%	1 (rye)
1516...	Utmost Starter	2	3,20%	0
1516...	Utmost Finisher	2	2,40%	0

- Microscopic evaluation showed that most grains looking like whole grains



were partially damaged, what enabled penetration by digestive substance and then digestion

- The presented method shows possibility of microscopical evaluation of fraction above 2 mm → structure of the grains and answer about what amount of mass is not digestible.
- Additionally: Information about amount of fraction above 2 mm gives important information about grinding quality of component to be pelleted.

## Use of blood products for farmed insects?

Current discussions on an issue – the use of blood products for farmed insects – raised by the industry for which it is not easy to find an answer in the relevant feed legislation.

The facts:

Regulation 999/2001 states in Annex IV Chapter I the extension of the feed ban

→ **Figure 1**

02001R0999 — EN — 15.04.2024 — 061.001 — 46

### ▼ M48

#### ANNEX IV

#### ANIMAL FEEDING

#### CHAPTER I

##### Extensions of the prohibition provided for in Article 7(1)

In accordance with Article 7(2), the prohibition provided for in Article 7(1) shall be extended to the feeding:

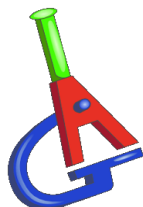
- (a) to ruminants of dicalcium phosphate and tricalcium phosphate of animal origin and compound feed containing these products;

### ▼ M69

- (b) to non-ruminant farmed animals, other than fur animals, of:

- (i) processed animal protein;
- (ii) blood products;
- (iii) hydrolysed protein of animal origin;
- (iv) dicalcium phosphate and tricalcium phosphate of animal origin;
- (v) feed containing the products listed in points (i) to (iv).

Figure 1: Reg. 999/2001 Annex IV Chapter I



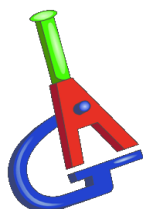
However, in chapter II derogations are mentioned. → **Figure 2**

As farmed insects are not mentioned in the [EURL-AP-SOP-operational-schemes-V5.1.pdf](#) and in the table of Regulation 999/2001, it is not clear if blood products are allowed to be fed to farmed insects.

CHAPTER II	
<i>Derogations from the prohibitions provided for in Article 7(1) and in Chapter I</i>	
In accordance with the first subparagraph of Article 7(3), the prohibitions provided for in Article 7(1) and in Chapter I shall not apply to the feeding to:	
(a) ruminants of:	
(i) milk, milk-based products, milk-derived products, colostrum and colostrum products;	
(ii) eggs and egg products;	
(iii) collagen and gelatine derived from non-ruminants;	
(iv) hydrolysed proteins derived from:	
— parts of non-ruminants, or	
— ruminant hides and skins;	
(v) compound feed containing the products listed in points (i) to (iv) above;	
(b) <b>non-ruminant farmed animals</b> of the following feed materials and compound feed:	
(i) hydrolysed proteins derived from parts of non-ruminants or from ruminant hides and skins;	
(ii) fishmeal and compound feed containing fishmeal which are produced, placed on the market and used in accordance with the general conditions laid down in Chapter III and the specific conditions laid down in Section A of Chapter IV;	
(iii) dicalcium phosphate and tricalcium phosphate of animal origin and compound feed containing such phosphates which are produced, placed on the market and used in accordance with the general conditions laid down in Chapter III and the specific conditions laid down in Section B of Chapter IV;	
(iv) <b>blood products derived from non-ruminants</b> and compound feed containing such blood products which are produced, placed on the market and used in accordance with the general conditions laid down in Chapter III and the specific conditions laid down in Section C of Chapter IV;	
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(c) aquaculture animals of the following feed materials and compound feed:	
(i) processed animal protein derived from non-ruminants, other than fishmeal and other than processed animal protein derived from farmed insects, and compound feed containing such processed animal protein, which are produced, placed on the market and used in accordance with the general conditions laid down in Chapter III and the specific conditions laid down in Section D of Chapter IV;	
(ii) processed animal protein derived from farmed insects, and compound feed containing such processed animal protein, which are produced, placed on the market and used in accordance with the general conditions laid down in Chapter III and the specific conditions laid down in Section F of Chapter IV;	
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(d) unweaned ruminants of milk replacers containing fishmeal and which are produced, placed on the market and used in accordance with specific conditions laid down in Section E of Chapter IV;	
(e) farmed animals of feed materials of plant origin and compound feed containing such feed materials contaminated with insignificant amount of bone spicules derived from unauthorised animal species. Member States may only use this derogation if they have carried out a risk assessment beforehand which has confirmed there is a negligible risk for animal health. That risk assessment must take into account at least the following:	
(i) the level of the contamination;	
(ii) the nature and the source of the contamination;	
(iii) the intended use of the contaminated feed;	
▼M69 ↓	
(f) poultry of the following feed materials and compound feed:	
(i) processed animal protein derived from porcine animals and compound feed containing such processed animal protein, which are produced, placed on the market and used in accordance with the general conditions laid down in Chapter III and the specific conditions laid down in Chapter IV, Section G;	
(ii) processed animal protein derived from farmed insects, and compound feed containing such processed animal protein, which are produced, placed on the market and used in accordance with the general conditions laid down in Chapter III and the specific conditions laid down in Chapter IV, Section F;	
(g) porcine animals of the following feed materials and compound feed:	
(i) processed animal protein derived from poultry and compound feed containing such processed animal protein, which are produced, placed on the market and used in accordance with the general conditions laid down in Chapter III and the specific conditions laid down in Chapter IV, Section H;	
(ii) processed animal protein derived from farmed insects, and compound feed containing such processed animal protein, which are produced, placed on the market and used in accordance with the general conditions laid down in Chapter III and the specific conditions laid down in Chapter IV, Section F.	

Figure 2: Reg. 999/2001 Annex IV Chapter II





After requesting the EURL-AP a very detailed and interesting answer from IPIFF was received:

### Blood products from non-ruminant are authorised for the farmed insects!

A guide was published the 17<sup>th</sup> of December by IPIFF after having been deeply examined by the Commission and the Member States. The attached picture is on page 21 of the guide.

#### → Figure 3














Substrates for insects intended for all applications (food, feed, technical uses) – Farmed insects qualify as 'farmed animals'- Article (3)(6) of Regulation (EC) 1069/2009	
Authorised	Prohibited
<b>Feed materials of vegetal origin</b> 	<b>'Feed Marketing' Regulation - Regulation (EC) No 767/2009 Annex III:</b> - Faeces and separated digestive tract content  - hide treated with tanning substances  - seeds and other plant-propagating materials (treated with plant protection products)  - wood or other materials derived from wood, which have been treated with wood preservatives  - waste derived from urban, domestic and industrial waste treatment  - packaging from agri-food products and parts thereof - protein products obtained from yeasts of the Candida variety cultivated on n-alkanes.
<b>Feed materials of animal origin</b>  <b>TSE Regulation (EU) No 999/2001, Article 7 and Annex IV, Chapter 1 and 2:</b> - Hydrolysed proteins, collagen and gelatine or blood products derived from non-ruminants (or parts of non-ruminants) (including compound feed containing such products) - Hydrolysed proteins from ruminant hide and skins - Dicalcium phosphate and tricalcium phosphate of animal origin (including compound feed containing such phosphates) - Fishmeal	<b>EU Animal By-Products (ABP) Regulation (EC) No 1069/2009:</b> - catering waste (Art. 11 (1) (b))  <b>TSE legislation - Regulation (EC) No 999/2001 (Annex IV, Chapter 1 and 2)</b> - Processed Animal Proteins (PAPs) referred to in Annex IV are prohibited: PAPs from ruminants, PAPs from poultry animals; PAPs from swine animals and PAPs from farmed insects and PAPs from other non-ruminants except fishmeal. - Blood products from ruminant animals - hydrolysed proteins of animal origin and derived from ruminants 
<b>Former Foodstuffs</b>  <b>TSE legislation - Regulation (EC) No 999/2001 Annex IV, Chapter II:</b> - Without meat and/or fish - only products containing the following ingredients of animal origin: eggs and egg products; milk, milk based-products and milk-derived products; honey; rendered fat; collagen; gelatine <i>*these ingredients must have been previously processed (either prior their intended use as food product or after being requalified as animal-by-product).</i> 	
<b>Residue limits for contaminants and requirements applying to feed additives</b> 	The 'Feed Marketing' Regulation (i.e. Regulation (EC) No 767/2009) provides that animals (including therefore insects) bred in the EU may be only be fed with <b>safe feed</b> . <b>Regulation (EC) No 396/2005</b> - maximum residue levels of <b>pesticides in feed</b> <b>Undesirable Substances Directive (i.e. Directive 2002/32/EC)</b>
<b>Feed additives</b> 	Only the feed additives which are authorised for all animal species may be used as feed ingredient for insects. - <b>Regulation (EC) No 1831/2003</b> . No specific additives for insects have been defined.

Figure 3: IPIFF Guide on Good Hygiene Practices

Enclosed is the link provided by IPIFF to this Guide ([Guide](#)) accessible and loadable on a website of the Commission (<https://food.ec.europa.eu/>).

**At this point, a big thank you to the Team of the EURL-AP, who are always helpful with good advice!**

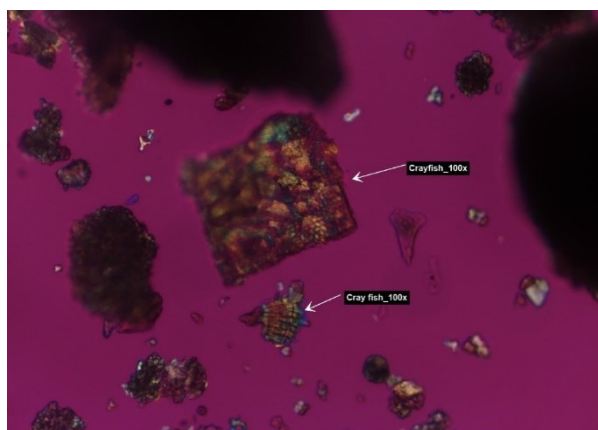
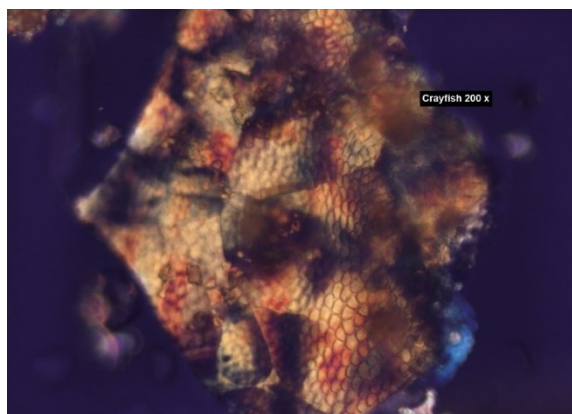
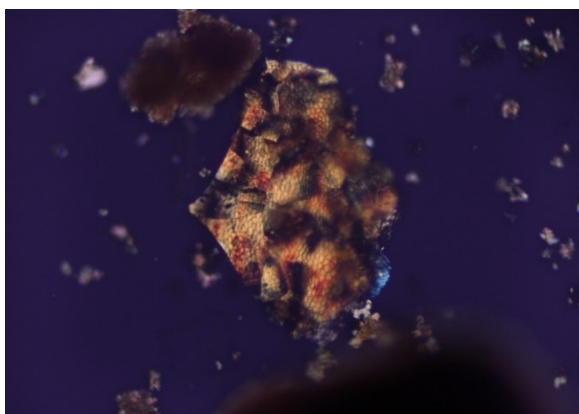




## Figures of unusual organisms

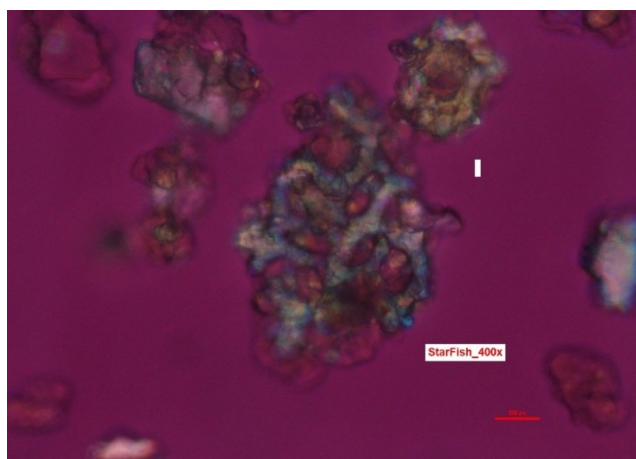
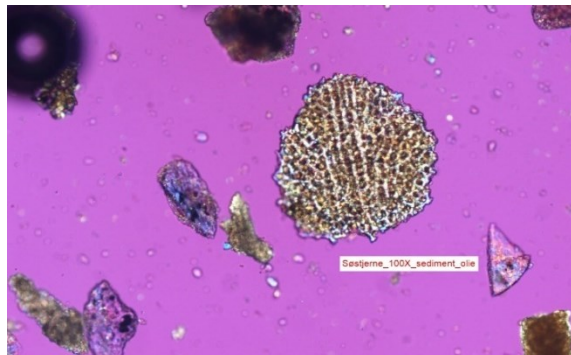
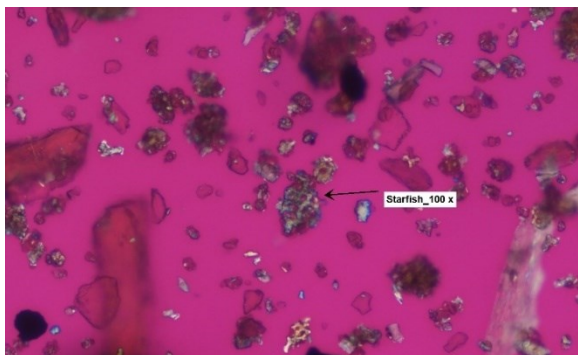
Thanks to Lotte to Hougs from The Danish Veterinary and Food Administration some pictures of crayfish and starfish particles which can be found in feedstuffs are presented.

Crayfish are freshwater crustaceans belonging to the infraorder Astacidea, which also contains lobsters (<https://en.wikipedia.org/wiki/Crayfish>).





Starfish or sea stars are star-shaped echinoderms belonging to the class Asteroidea (<https://en.wikipedia.org/wiki/Starfish>).



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