

# Animal feeding stuff: Global Standard for the Determination of Acid Detergent Fibre (ADF) and Lignin

Today the requirements for a well balanced compound feed have become higher and higher. A desirable fibre content in the feed yields better breeding and higher cost efficiency, but requires more analysis and quality control. For monogastric animals a proper proportioning of fibre fractions increases the utilisation of the compounded feed. For ruminants, fibre is an important part of the metabolism in the rumen. It is a determining factor for the hydrolysis of all nutritional ingredients in the feed.

For a number of years, livestock nutritionists have been using Acid Detergent Fibre (ADF) and Neutral Detergent Fibre (NDF) as indicators of dietary energy and intake, especially for ruminant rations. As a result these fibre fractions have replaced

crude fibre (CF) in ration formulations in many parts of the world. Today ADF and NDF values are frequently used to estimate the amount of forage that can be digested by animals; the total digestible nutrients and other energy values, as well as the relative feed value an index used to allocate the correct forage to specific animal performance; to price hay and to assess forage management, harvest and storage skills.

The detergent system of feed analysis (Fig. 1) was developed by Peter Van Soest at the United States Department of Agriculture in the 1960s<sup>(1)</sup> and is today one of the most important sets of feed assays in ruminant nutrition but also, increasingly, in non-ruminant research.

The concept behind detergent fibre

analysis is that plant cells can be divided into less digestible cell walls (comprising hemicellulose, cellulose and lignin) and mostly digestible cell contents (comprising starch and sugars). These two components can be separated by using two detergents: a neutral detergent and an acid detergent. Neutral Detergent Fibre is a good indicator of bulk and thus feed intake. Acid detergent fiber is a good indicator of digestibility and thus energy intake.

A harmonised global standardisation method for the determination of neutral detergent fibres has recently been published by the AOAC<sup>(2)</sup> and jointly by the International Organisation for Standardisation (ISO) and the European Committee for Standardisation (CEN)<sup>(3)</sup>.

Based on the AOAC Official Method 973.18<sup>(4)</sup> 'Fibre (Acid detergent) and lignin (H<sub>2</sub>SO<sub>4</sub>) in animal feed', ISO and CEN in 2008 also issued a fully validated standard entitled 'Animal feeding stuffs – Determination of acid detergent fibre content (ADF) and lignin (ADL)'<sup>(5)</sup>.

Since the 1970's, FOSS has been a pioneer in the determination of fibre fractions using Tecator Fibertec™ systems.

This report summarises the results of an international validation study for the EN ISO 13906:2008 standard, performed by FOSS.

## Inter-laboratory Study

Samples of sawdust, wheat, cattle feed, soy meal, clover silage and grass silage were selected for the study. Participants received blind duplicates of the samples numbered 1-12, without identifying the type of sample.

Details of the method are provided in the standard<sup>(5)</sup>, the FOSS application note, and the study report<sup>(6)</sup>.

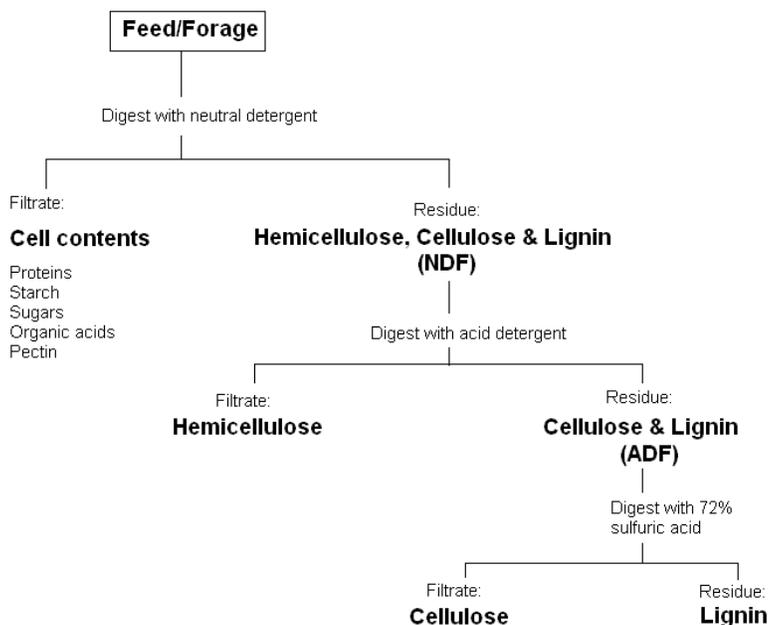


Fig. 1 : The detergent fibre system according to Van Soest



## Results and Discussion

Out of the 22 participating labs, four used conventional or other equipment and 18 used the FOSS Fibertec system. Results of the statistical evaluation for ADF can be seen in Table 1 and Fig. 2.

The performance data, in the form of relative errors (coefficient of variation of repeatability and reproducibility) is better than 10%, except for Sample 2 (see Fig. 2). These data are comparable with those obtained in the NDF method<sup>(3)</sup>.

The results of the statistical evaluation for the determination of acid detergent lignin are shown in Table 2.

Based on the obtained results it is recommended to report values lower than 1,5 % as < 1,5%.

## Conclusions

As the EN ISO 13906:2008 method is based on the AOAC Official Method 973.18 – a globally harmonised, validated protocol for the determination of Acid Detergent Fibre (ADF) and Lignin (ADL) has now become available.

This is of importance for the global trade of raw materials and compound feed, international research, the ISO 17025 accreditation of laboratories using the FOSS

Fibertec systems and for the development of NIR prediction models, but – more importantly – it enables official bodies to require fibre guarantees on livestock feed labels (depending on species) based on globally harmonised methods:

### **For ruminant feed labels:**

ADF and NDF guarantees

### **For non-ruminant feed labels:**

CF guarantees

### **For both ruminant and non-ruminant feed labels:**

ADF, NDF, and CF guarantees

*Cont'd on page 24*

Sample	Saw dust	Wheat	Cattle feed	Soy meal	Clover silage	Grass silage
Number of labs after elimination of outliers	19	18	18	17	17	20
Average ADF value, g/100 g	72,7	3,5	14,1	7,2	30,0	31,4
Repeatability standard deviation, $s_r$ , g/100 g	0,95	0,30	0,30	0,32	0,56	0,65
Coefficient of variation of repeatability, %	1,3	8,6	2,1	4,5	1,9	2,1
Repeatability limit $r$ (2,8 $s_r$ ), g/100 g	2,7	0,9	0,9	0,9	1,9	1,8
Reproducibility standard deviation, $s_R$ , g/100g	2,44	0,63	0,85	0,56	1,13	1,57
Coefficient of variation of reproducibility, %	3,4	17,7	6,1	7,7	3,8	5,0
Reproducibility limit $R$ (2,8 $s_R$ ), g/100 g	6,8	1,8	2,4	1,6	3,2	4,4

Table 1: Interlaboratory study results for the determination of ADF

Sample	Saw dust	Wheat	Cattle feed	Soy meal	Clover silage	Grass silage
Number of labs after elimination of outliers	20	14	20	15	18	19
Average ADL value, g/100 g	20,3	0,9	4,3	0,6	4,0	3,0
Repeatability standard deviation, $s_r$ , g/100 g	0,59	0,08	0,152	0,225	0,334	0,206
Coefficient of variation of repeatability, %	2,9	9,1	3,5	39,0	8,3	6,9
Repeatability limit $r$ ( $2,8 s_r$ ), g/100 g	1,7	0,2	0,4	0,6	0,9	0,6
Reproducibility standard deviation, $s_R$ , g/100g	0,75	0,19	0,53	0,30	0,52	0,48
Coefficient of variation of reproducibility, %	3,7	21,1	12,3	52,6	12,9	16,0
Reproducibility limit $R$ ( $2,8 s_R$ ), g/100 g	2,1	0,5	1,5	0,8	1,4	1,3

Table 2: Interlaboratory study results for the determination of ADL (lignin)

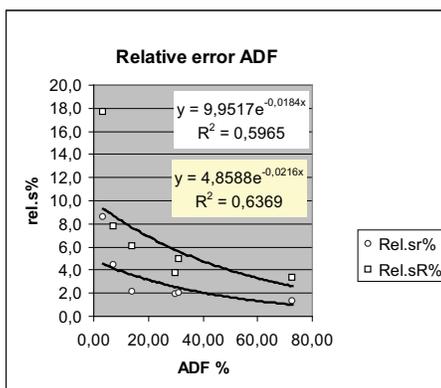


Fig. 2 Change in the coefficient of variation of repeatability and reproducibility as a function of the ADF content

Cont'd from page 23

## References

- (1) Goering, H.K. and Van Soest, P.J. (1970) Forage fibre analysis (apparatus, reagents, procedures, and some applications). USDA Agricultural Research Service Handbook No. 379. Washington, DC.
- (2) Official Methods of Analysis, AOAC INTERNATIONAL, Gaithersburg, MD, Official Method 2002.04
- (3) EN ISO 16472:2006 Animal feeding stuffs - Determination of amylase-treated neutral detergent fibre content (aNDF) – [www.iso.org](http://www.iso.org) / [www.cen.eu](http://www.cen.eu)
- (4) Official Methods of Analysis, AOAC INTERNATIONAL, Gaithersburg, MD, Official Method 973.18
- (5) EN ISO 13906:2008 Animal feeding stuffs – Determination of acid detergent fibre (ADF) and acid detergent lignin (ADL) contents – [www.iso.org](http://www.iso.org) / [www.cen.eu](http://www.cen.eu)
- (6) To be published in the Journal of the AOAC

by Dr. Jürgen Möller, FOSS ([jmr@foss.dk](mailto:jmr@foss.dk))

The new standard EN ISO 13906:2008 Animal feeding stuffs – Determination of acid detergent fibre (ADF) and acid detergent lignin (ADL) contents, describes the use of a refluxing apparatus and gives the FOSS Fibertec™ system as a suitable option.

Fibertec was used by 18 of 22 laboratories taking part in the collaborative study.

A globally accepted standard for amylase treated Neutral Detergent Fibre (aNDF) has already been issued – ISO 16472:2006 and AOAC 2002.04. This standard also states the Fibertec system as a valid option.

The advantage of the Fibertec apparatus compared to the manual (refluxing in beakers) can be summed up as follows:

- No transfer of sample improves accuracy and precision. The sample is weighed into the crucible and kept there throughout the analysis
- Integrated system for extraction and filtration of 6 samples at a time
- Quick vacuum filtration with anti-clog system
- Minimum contact with reagents as the system offers both heating up and addition of reagents and water
- Reagent addition system also offers simple wash down of sample stuck to the walls
- The built-in timer ensures that extraction conditions are kept from batch to batch
- Cold Extraction Unit for the ADL step

More information about Fibertec™ systems can be found on [www.foss.dk](http://www.foss.dk)

## FOSS application notes for Fibertec™ 2010 and Fibertec™ 1020 M6 :

AN 3437 Determination of Crude Fibre according to EN ISO 6865:2000 and 92/89/EEC

AN 3428 Determination of Crude Fibre according to AOAC 978.10, AACC 32-10 and AOCS 6-84

AN 3434 Determination of Neutral Detergent Fibre (NDF) according to EN ISO 16472:2006 and AOAC 2002:04

AN 3429 Determination of Acid Detergent Fibre (ADF) and Acid Detergent Lignin (ADL) according to EN ISO 13906:2008 and AOAC 973.18

## FOSS application notes for FiberCap 2021/2023:

AN 3801 Determination of Crude Fibre in agreement with AOAC 978.10, AACC 32-10 and AOCS 6-84

AN 3802 Determination of Crude Fibre in agreement with EN ISO 6865:2000 and 92/89/EEC

AN 3805 Determination of Neutral Detergent Fibre (NDF) in agreement with EN ISO 16472:2006 and AOAC 2002:04

AN 3804 Determination of Acid Detergent Fibre (ADF) and Acid Detergent Lignin (ADL) in agreement with EN ISO 13906:2008 and AOAC 973.18