

N/Protein Determination in Feed according to the Kjeldahl method

Reference: **AOAC 984.13** Protein (Crude) in Animal Feed and Pet Food

Tested with **VELP Scientifica DKL 20 Automatic Kjeldahl Digestion Unit** (Code S30100210) and **UDK 159 Automatic Kjeldahl Distillation&Titration System** (Code F30200150).



Introduction

Protein is a critical nutrient in cattle, pig, poultry, sheep, horse and pet foods.

Although protein supplements are high cost they are sometimes necessary in order to meet the animal's nutritional requirements. An adequate protein content in feed products is important for animal health and productivity, as well as for ranch profitability. The protein requirements of cattle vary with age, size, and expected performance.

During lactation, larger cattle typically require more pounds of crude protein per day than smaller animals, but as a lower percentage of their total dry matter intake. In other words, lighter cattle require higher quality feeds and forages at lower quantities compared with heavier cattle. Cattle requirements for crude protein rise with increasing lactation and rate of gain. Protein is required for milk production and reproductive tract reconditioning after calving.

Protein Determination in feed according to the Kjeldahl method

Kjeldahl is nowadays the most used method for determining nitrogen and protein contents in foods and feeds thanks to the high level of precision and reproducibility and to its simple application.

The modern Kjeldahl method consists in a procedure of catalytically supported mineralization of organic material in a boiling mixture of sulfuric acid and sulfate salt at digestion temperatures higher than 400 °C. During the process the organically bonded nitrogen is converted into ammonium sulfate. Alkalizing the digested solution liberates ammonia which is quantitatively steam distilled and determined by titration.

Samples:

Animal's feed products:	Indicative protein content:
1- Rapeseed cake	31.5 %
2- Rapeseed cake 2	30.0 %
3- Corn feed	6.5 %
4- Corn gluten	61.0 %
5- Soya cake	43.0 %
6- Potato's proteins	73.0 %

Sample Digestion

Grind the sample using a grinder (1 mm particles size).

Weigh 0.5 - 1 g of sample to an accuracy of 0.1 mg into a 250 ml test tube.

For each sample add in the test tube:

- 1 catalyst tablet VW (code A00000282; 4.875g K₂SO₄, 0.075 g CuSO₄ x 5H₂O, 0.050 g Se, 5 g)
- 13 ml concentrated sulphuric acid (96-98%)

Prepare some blanks with all chemicals and without sample.

Connect the Digestion Unit to a proper Aspiration Pump (JP code F30620198) and a Fume Neutralization System (SMS Scrubber code F307C0199) to neutralize the acid fumes created during digestion phase.

Digest the samples for 60 minutes at 420 °C according to the method "cereals and animal feed" (n° 7 on DKL 20).

Distillation and Titration

Let the test tubes to cool down to 50-60 °C.

Condition the UDK 159 unit by performing the Automatic Check up in Menu-System and a Wash down.

Distill the samples according to the following parameters (pre-defined method n° 7):

- H₂O (dilution water): 50 ml
- NaOH (32 %): 50 ml
- H₃BO₃ (4 % with indicators): 30 ml
- HCl (0.2N) as titrant solution
- Protein factor: 6.25

Distillation&Titration analysis time: from 4 minutes for one test.

Typical Results on Feed Products

The results are calculated as a percentage of nitrogen and percentage of protein.

Sample	Sample quantity (g)	Nitrogen %	Protein %
Rapeseed cake	0,510	5,084	31,774
	0,504	5,111	31,942
	0,501	5,058	31,615
	Average ± SD%	5.084 ± 0.027	31.777 ± 0.164
	RSD% *	0,521	0,515
Rapeseed cake 2	0,545	4,822	30,134
	0,508	4,836	30,225
	0,531	4,820	30,125
	Average ± SD%	4.826 ± 0.009	30.161 ± 0.055
	RSD% *	0,181	0,183
Corn feed	0,667	1,079	6,742
	0,663	1,076	6,727
	0,635	1,074	6,714
	Average ± SD%	1.076 ± 0.002	6.728 ± 0.014
	RSD% *	0,208	0,208
Corn gluten	0,504	9,859	61,618
	0,510	9,783	61,147
	0,501	9,799	61,244
	Average ± SD%	9.814 ± 0.040	61.336 ± 0.249
	RSD% *	0,418	0,415
Soya cake	0,500	7,005	43,781
	0,504	6,942	43,385
	0,505	6,966	43,535
	Average ± SD%	6.971 ± 0.032	43.567 ± 0.200
	RSD% *	0,456	0,459
Potato proteins	0,505	11,693	73,083
	0,500	11,728	73,300
	0,500	11,701	73,128
	Average ± SD%	11.707 ± 0.018	73.170 ± 0.115
	RSD% *	0,157	0,157

Protein Factor: 6.25

* RSD% = (Standard Deviation * 100) / Average

The complete procedure was verified by using 5 ml of glycine standard solution containing 28 mg of nitrogen, as reference substance.

The obtained recovery falls into the expected range: between 98% and 102%.

Conclusion

The obtained results are reliable and reproducible in accordance with the expected values, with a low relative standard deviation (RSD < 1%), that means high repeatability of the results.

Benefits of Kjeldahl method by using DKL 20 and UDK 159 are:

- High level of precision and reproducibility
- High productivity
- Worldwide official method
- Reliable and easy method
- Time saving
- Affordable equipment cost
- Moderate running costs