Nitrogen/Protein Determination by Combustion of Food and Animal Feed using Argon as Carrier Gas

Liliana Krotz, Francesco Leone and Guido Giazzi
Thermo Fisher Scientific, Strada Rivoltana, 20090 Rodano (MI), Italy

ABSTRACT

Protein determination is used for the evaluation of food and beverage product quality and potentially for the uncovering of potential mislabeling cases in food and animal feed. Elemental Analysis is a powerful analytical technique performing nitrogen determination for the determination of protein content and it is an alternative to the classical Kjeldahl Method. This poster shows the performance of the Thermo Scientific Flash*Smart* Elemental Analyzer for N/Protein determination , by using argon as carrier gas, in different nitrogen concentrations in food and animal feed reference materials, showing the advantages of elemental analysis over Kjeldahl Method.

INTRODUCTION

Protein is one of the most important nutrients of food and animal feed. The accurate determination of the amount of protein, through the determination of the nitrogen content, is fundamental for determining nutritional quality of animal feed and securing the safety of final food products intended for human consumption. Furthermore, protein content is the basis for trade of feed and official regulations establish protein content and labeling requirements.

Latest developments in analytical instrumentation have greatly improved the capabilities of the combustion method for the determination of nitrogen, making it faster, safer and more reliable than the traditional Kjeldahl method. The Dumas (combustion) method has been approved and adopted by industry associations (AOAC, AACC, AOCS, ASBC, IDF, IFFO, ISO and others).

The Thermo Scientific™ Flash*Smart*™ Elemental Analyzer (Figure 1), based on the dynamic combustion method (modified Dumas method), provides rapid and automated nitrogen determination without the use of hazardous chemicals and offers advantages in precision over traditional methods. The Flash*Smart* EA allows runs at both high and low nitrogen levels with no need to change configurations and without matrix effects.

The Flash*Smart* EA typically runs helium as carrier gas, which ensures high sensitivity. Considering the need for cost efficiencies and the likely increase in helium gas cost, an alternative gas, to be used as carrier gas, is needed. Argon can be used as alternative to helium in the Flash*Smart* EA. This poster shows the performance of the Flash*Smart* EA when running argon as carrier gas, and provides comparison of the results with data obtained when using helium as carrier gas.

Figure 1. Flash*Smart* EA Elemental Analyzer.

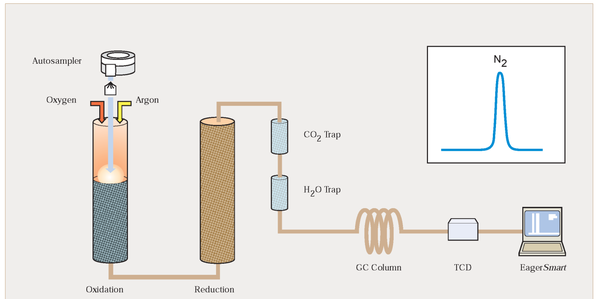
METHOD

The Elemental Analyzer operates according to the dynamic flash combustion of the sample. Samples are weighed in tin containers and introduced into the combustion reactor via the Thermo Scientific™ MAS Plus Autosampler together with the proper amount of oxygen.

After combustion, the produced gases are carried by an argon flow to a second reactor filled with copper, then swept through CO_2 and H_2O traps, a GC column and finally detected by a Thermal Conductivity Detector (TCD) (Figure 2).

A report is automatically generated by the Thermo Scientific™ Eager*Smart*™ Data Handling Software and displayed at the end of the analysis. The dedicated software converts automatically the nitrogen content in protein content using a specific protein factor.

Figure 2. Nitrogen/Protein configuration.



RESULTS

To evaluate the performance of the system several Flours and Animal Feed Reference Materials as the Thermo Scientific Pasta Reference Material, with different nitrogen content, were analyzed.

Instrument calibration was performed with about 50-70 mg of aspartic acid (10.52 N%) standard using K factor as calibration method. The protein factor 6.25 was used to calculate the protein content.

Table 1 shows the certified nitrogen values of the flours reference materials and Thermo Scientific Pasta Reference Material, the uncertainty declared by the supplier so as the accepted range according to the technical specification of the Flash*Smart* EA.

Table 2 shows the certified nitrogen values of the BIPEA (Bureau Inter-Professionnel d'Etudes Analytiques, France) animal feed reference materials.

Table 3 shows the experimental data obtained using argon as carrier gas. Samples were analyzed in triplicate.

Table 1. Certified N% of Pasta and Flours Reference Materials.

Reference Material	Specification	
	N%	Uncertainty (±)
Thermo Scientific Pasta	2.227	0.097
Wheat Flour	1.36	0.25
Rice Flour	1.38	0.05
Barley Flour	1.90	0.04
Oatmeal	1.90	0.10
Soy Bean Meal	7.50	0.05

Table 2. Certified values of BIPEA Animal Feed Reference Materials.

BIPEA Ref.Mat.	Moisture %	Fat %	Carbohydrate %	Kjeldahl Protein %		Combustion Protein %	
				Av.	±	Av.	±
Feed for Sow	9.8	2.8	48.7	16.0	0.6	16.2	0.6
Dehydrated Alfalfa	7.7	-	29.3	14.8	0.6	15.1	0.6
Hyperproteic Powder	-	0.8	-	85.4	3.4	86.4	3.5

Table 3. Experimental data of Reference Materials.

Reference Material	Weight (mg)	N%	RSD%	Protein %	RSD%
Thermo Scientific Pasta	150 - 160	2.22 2.21 2.22	0.26	13.87 13.81 13.87	0.26
Wheat Flour	140 - 150	1.37 1.36 1.35	0.74	8.57 8.51 8.41	0.95
Rice Flour	140 – 150	1.40 1.38 1.39	0.72	8.73 8.65 8.69	0.46
Barley Flour	140 – 150	1.90 1.92 1.92	0.60	11.90 11.97 12.01	0.47
Oatmeal	140 – 150	1.88 1.88 1.86	0.62	11.76 11.73 11.61	0.68
Soy Bean Meal	140 – 150	7.52 7.51 7.51	0.07	46.98 46.93 46.93	0.06
Feed for Sow	100 – 120	2.60 2.63 2.61	0.58	16.22 16.41 16.32	0.58
Dehydrated Alfalfa	100 - 140	2.37 2.38 2.37	0.24	14.81 14.87 14.79	0.28
Hyperproteic Powder	100 – 120	13.69 13.69 13.67	0.08	85.57 85.55 85.43	0.09

Table 4 shows the Nitrogen/Protein data of different samples using argon and helium carrier gas including the range of weight used for analysis. The protein factor used to calculate the protein content was 6.25 for animal feed while 6.38 for milk samples. No remarkable difference was observed.

Table 4. Reproducibility of N/Protein data using helium and argon as carrier gases.

Sample	Argon carrier gas					Helium carrier gas				
	Weight (mg)	N%	RSD%	Prot. %	RSD%	Weight (mg)	N%	RSD%	Prot.%	RSD%
Milk (liquid)	100-125	0.51 0.52 0.51	1.12	3.20 3.23 3.20	0.54	135-140	0.51 0.51 0.52	1.12	3.21 3.20 3.24	0.65
Milk powder 1	110-115	2.31 2.30 2.34	0.85	14.76 14.68 14.93	0.85	200-220	2.34 2.33 2.34	0.11	14.90 14.89 14.92	0.11
Milk powder 2	110-115	2.52 2.50 2.48	0.71	16.06 15.97 15.84	0.71	190-200	2.50 2.50 2.51	0.13	15.97 15.96 16.00	0.13
Biscuits	125-140	1.20 1.19 1.18	0.84	7.47 7.45 7.38	0.64	190-210	1.19 1.17 1.19	0.98	7.41 7.31 7.43	0.87
Mais	130-140	1.48 1.49 1.49	0.39	9.24 9.29 9.29	0.31	150-200	1.50 1.50 1.49	0.38	9.41 9.39 9.34	0.38
Sunflower	130-140	5.69 5.75 5.71	0.53	35.54 35.95 35.68	0.58	170-200	5.65 5.61 5.64	0.37	35.31 35.05 35.22	0.37
Soy	130 - 150	8.11 8.08 8.00	0.71	50.66 50.51 49.99	0.70	170-180	7.98 8.03 8.01	0.31	49.87 50.21 50.06	0.34
Gluten	130 - 140	9.31 9.27 9.28	0.22	58.20 57.92 58.02	0.24	170-180	9.34 9.33 9.31	0.16	58.36 58.33 58.21	0.14
Poultry feed	130 - 140	4.31 4.39 4.30	1.14	26.96 27.43 26.87	1.11	150-160	4.37 4.35 4.42	0.82	27.30 27.17 27.65	0.91

SUMMARY

For the N/Protein determination of all type of raw and final food and animal feed, the application showed that the Dumas Method meets manufacturers and laboratories requirements, including the compliance to official methods. The Thermo Scientific Flash*Smart* Elemental Analyzer based on the combustion method (Dumas) offers advantages over the Kjeldahl Method for the N/Protein determination in terms of automation, easy to use and cost per sample.
The Flash*Smart* Elemental Analyzer, using argon as carrier gas, enables to analyze nitrogen in a wide range from low to high content without matrix effect. The nitrogen data obtained fall within the tolerance declared in the certificates and the RSD% obtained was less than 2% of the performance requirements of the Official Methods. No memory effect was observed when changing the type of sample, indicating complete combustion and detection of the element.
The Dumas Combustion method has been approved and adopted by Official Organizations such as ASBC, AOAC, AACC, AOCS, ISO, IFFO and IDF.

TRADEMARKS

Poster presented at the AOAC Conference 2017, September 24-27, Atlanta, GA.
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