N/Protein Determination in Soybean
according to Dumas combustion method

Reference: **AOAC 992.23** Crude Protein in Cereal Grains and Oil seeds

Tested with **VELP Scientifica NDA 701 Dumas Nitrogen Analyzer** (Code F30800070)

Introduction

Soybean meal is the most important protein source used to feed farm animals. It represents two-thirds of the total world output of protein feedstuffs, including all other major oil meals and fish meal (Oil World, 2010). Indeed it is an important part of the diets of ruminants animals due to its high amount (more than 60%) of rumen-degradable protein, good amino acid balance and high cell-wall digestibility, besides being also very palatable to ruminants.

Soybean meal is the by-product of the extraction of soybean oil. In the solvent extraction process, the soybeans are cracked, heated, flaked and the oil is extracted by solvent. The extracted flakes are then dried to eliminate the solvent, toasted and ground. The soybeans may have been de-hulled prior to extraction, and the hulls may be added back at the end of the process.

Protein Determination in Soybean flour

The Dumas method starts with a combustion furnace (CF) to burn the sample, obtaining elemental compounds.

Water is removed by a first physical trap (WT1 - **DriStep™**), placed after the combustion, and a second chemical one (WT2). Between the two, the elemental substances passed through a reduction furnace (RF).

The auto-regenerative CO_2 adsorbers (CO_2) let pass only the elemental nitrogen that is detected by the **LoGas™** innovative Thermal Conductivity Detector (TCD) with no requirement for a reference gas.

The NDA 701 is controlled via PC through the intuitive **DUMASoft™**.
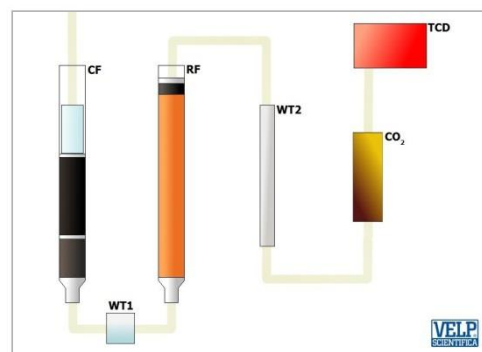
NDA 701 Preliminary Operations (daily)

Follow the operating manual to start the NDA 701 and check that the following parameters are set:

Temperature Combustion reactor (Code A00000158): 1030 °C
Temperature Reduction reactor (Code A00000226): 650 °C
Flow rate MFC1 He: 190 ml/min
Flow rate MFC2 He: 220 ml/min

Condition the system by testing 2 EDTA standard (Code A00000149) and 3 to 5 empty tin foils (Code A00000153) as Check up.

Verify the calibration curve with one or more tests as Standard by testing the same standard used for the curve creation.

Sample Preparation

Grind test samples to suitable fineness to obtain 2.0% relative standard deviation (RSD) for 10 successive nitrogen determinations.

Using a spatula, put moreless 50 mg of sample directly into the tin foil.

Close the tin foil, obtaining a capsule.

Load the capsule into the autosampler.

Analysis Procedure

Fill the following fields in the database: **Sample name, Weight, Method, Sample type, Calibration number**

The "SOYA BEAN" method shows the following parameters:
Protein factor: 5.70
O$_2$ flow rate: 400 ml/min
O$_2$ factor: 1.8 ml/mg

Press ⊙ to start the analysis.
Analysis time: from 3 minutes for one run.

Samples:

Soybean flour   Indicative protein content: 42.0 - 44.0 %

Typical Results on Soybean Flour

The table below shows the results obtained on the same sample of soy bean flour analyzed for 10 days running. The results are calculated as a percentage of nitrogen and percentage of protein on an average of 10 runs perfomed daily on the same sample.

Sample	Day n°	Average Nitrogen % ± SD	Average Protein % ± SD	RSD %
Soybean Flour	1	7.549 ± 0.064	43.030 ± 0.363	0.843
	2	7.504 ± 0.080	42.771 ± 0.457	1.067
	3	7.512 ± 0.063	42.817 ± 0.360	0.840
	4	7.540 ± 0.062	42.975 ± 0.357	0.827
	5	7.562 ± 0.030	43.104 ± 0.169	0.395
	6	7.555 ± 0.053	43.062 ± 0.303	0.700
	7	7.575 ± 0.051	43.175 ± 0.290	0.672
	8	7.566 ± 0.048	43.127 ± 0.275	0.638
	9	7.620 ± 0.060	43.333 ± 0.342	0.788
	10	7.578 ± 0.029	43.192 ± 0.165	0.383

Protein Factor: 5.70

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

Conclusion

The obtained results are reliable and in accordance with the expected value, showin a good ripetibility and an excellent reproducibility of the analysis.
The combustion method, relying on the Dumas principle, for the determination of total nitrogen in soybean flour, has been included as an official alternative to the Kjeldahl method.

Results have been obtained with the following calibration curve: in a range of 1.3 – 4.8 mg N with 5 measurements of EDTA (%N = 9.57) (Code A00000149).

Benefits of Dumas combustion method are:
- High productivity, non-stop performance
- Time saving, few minutes required
- Moderate running costs
- Totally unsupervised, fully automated
- Omission of harsh and toxic chemicals
- Eco-friendly, low amount of residues and wastes

Several organizations working with standardization and recommendation of chemical methods have approved combustion methods for the determination of nitrogen.

Thanks to development in sophisticated instrumentation, the Dumas principle, became a practical and popular alternative for the determination of nitrogen in organic matrices.