

N/Protein Determination in Milk Powder according to the Kjeldahl method

Reference: **AOAC 991.20** Nitrogen (Total) in Milk; **IDF 20-1**, **ISO 8968-1 Second Edition 2014-02-01** Milk and milk products - Determination of nitrogen content

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



Introduction

Powdered milk or dried milk is a manufactured dairy product made by removing almost all the water. This product has a far longer shelf life than liquid milk and does not need to be refrigerated, due to its low moisture content.

It can be reconstituted by adding water in order to obtain a ready-to-use liquid milk: on the water addition depends the fat content, so its high nutritional quality, combined with its versatile and multi-functional properties, makes it attractive for many food applications.

Dried milk is widely used as food ingredient because it is suitable for several applications: in confectionery, such as chocolate and caramel candy, manufacture of infant formula and recipes for bakery.

Protein Determination in Milk 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.

Sample

Milk Powder Type 1: protein expected value 12.4% Type 2: protein expected value 11.8%

Sample Digestion

Transfer the sample to a container. Mix the sample by repeatedly rotating and inverting the container.

Weight about 1 gram of sample in a nitrogen-free weighing boat (Code CM0486000) and transfer in a test tube

For each sample, add in the test tube:

- 2 catalyst tablet CM (code CT0006650; 3.5 g K₂SO₄, 0.1 g CuSO₄, 5H₂O Missouri)
- 2 Antifoam S code CT0006650
- 20 ml concentrated sulphuric acid (96-98%)
- 5 ml of hydrogen peroxide (~30%)

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 15 minutes at 150 °C, plus 15 minutes at 250 °C and 40 minutes at 420 °C according to the method "milk and derived products" (n° 1 on DKL 20).

Distillation and Titration

Let the test tubes 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°1):

- H₂O (dilution water): 50 ml
- NaOH (32 %): 70 ml
- H₃BO₃ (4 % with indicators): 30 ml
- H₂SO₄ (0.1 N) as titrant solution
- Protein factor: 6.38

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

Typical Results on different Milk Powders

The results are automatically calculated by UDK 159 as a percentage of nitrogen and percentage of proteins. This is "protein" on a total nitrogen basis.

Sample	Sample quantity (g)	Nitrogen %	Protein %
Milk Powder type 1	0.963	1.945	12.410
	1.046	1.940	12.380
	0.986	1.923	12.270
	0.957	1.956	12.480
	0.986	1.950	12.440
Average \pm SD%		1.943 \pm 0.012	12.396 \pm 0.080
RSD% *		0.642	0.642
Milk Powder type 2	1.012	1.840	11.740
	1.068	1.837	11.720
	1.047	1.840	11.740
	1.123	1.845	11.770
	1.085	1.840	11.740
Average \pm SD%		1.840 \pm 0.030	11.742 \pm 0.018
RSD% *		0.152	0.152
Protein Expected Value: Type1: 12.4%; Type 2: 11.8%			
Protein Factor: 6.38			

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

The complete procedure was verified by using 5 ml of glycine standard solution (3%) 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