Nitrogen Determinations in Breweries and Maltings

1. Introduction

Nitrogen, respectively protein determination is one of the most important analysis task to be done in the laboratories of breweries and maltings for routine incoming inspection and process control. Typical analysis samples are raw fruits, malt, wort and beer. Sometimes also coagulated and with $MgSO_4$ precipitable proteins in wort or beer have to be measured in connection with further special investigations. So far, the nitrogen determination technique was the Kjeldahl method with all known disadvantages. As an alternative, all nitrogen analyses in breweries and maltings, can also be done with the nitrogen/protein analyzer ***macro N*** in a fully automatic and environmental friendly way.

2. Analytical Performance

Reproducibility (Precision)

Raw fruit:	VC = approx. 0.5 % (rough grind), repectively 1 % (fine grind)
Malt:	VC = approx. 0.7 - 1 % (fine grind)
Wort:	VC = approx. 0.7 - 1.5 %
Beer:	VC = approx. 0.7 - 1.5 %

(VC = variation coefficient = relative standard deviation).

The reproducibility to be reached with the macro N is at least equal to the Kjeldahl method for all brewery relevant products. For reliable results double determinations are sufficient in most cases.

Examples (10 times Determinations)

Raw fruits and malts	% Protein	VC
barley raw fruit (fine grind)	10.29	0.4
wheat raw fruit (fine grind)	11.94	0.4
wheat raw fruit (rough grind)	11.93	1.0
barley malt (fine grind)	11.45	0.7
wheat malt (fine grind)	11.46	0.7

cont'd

Worts and Beers	mg/l N	VC
wort 1	762	0.8
wort 2	810	1.3
wort 3	850	0.7
wort 4	998	0.7
wort 5	1170	0.8
wort 6	1367	0.5
beer 1	797	1.3
beer 2	954	0.3
beer 3	1078	0.2
beer 4	1086	0.5

Comparability with Kjeldahl

It is well known that the recovery rates reached with the Kjeldahl method depend on a variety of factors as digestion time, catalysts, sample matrix etc. The MEBAK-Kjeldahl determination requires copper as a catalyst and as a consequence it is a relatively weak digestion.

With the much stronger digestion by using the combustion method of the macro N the found values are slightly higher compared to the MEBAK-Kjeldahl. If necessary, the macro N results can be recalculated toward Kjeldahl results. This makes it possible to compare with other laboratories which are working with the Kjeldahl method. Therefore, it is necessary to determine the conversion factors macro N → Kjeldahl for each particular sample matrix: out of a sufficient number of representative samples, the ratio Kjeldahl/macro N is calculated. The average of these ratios is representing the conversion factor for the particular sample matrix: % N (macro N) x factor = % N (Kjeldahl); refer to the below described examples. The determination of the conversion factors has to be done only once by the user with his/her particular sample material. The examples in 3.1.3 and 3.2.3 are not of general use !

3. Analysis Procedure

Raw Fruits and Malts

Sample Preparation and Sample Weight

Degree of grinding (refer to MEBAK Analyses Manual) for malts: fine grind, for raw fruits: rough or fine grind. Grind roll distance of the MEBAK mill: 0.2 mm (fine grind), respectively 1.0 mm (rough grind). Sample weight: approx. 1 g.

Measuring Parameter for Brewing Grain and Malt

Refer to Operation Manual, Chapter 20, Example No. 48 (identical with no. 46, respectively no. 48 Q, quick measurement 9 minutes).

Measuring Examples

Barley and Wheat Raw Fruit (rough grain, double determiantions)

	macro N			Kjeldahl			Kj. /	macro N
Sample	**% N**	**% N**	**Mean**	**% N**	**% N**	**Mean**	**macro N**	**corr.**
Barley								
1	1.70	1.74	1.72	1.59	1.61	1.60	0.930	1.62
2	1.45	1.50	1.48	1.40	1.42	1.41	0.956	1.40
3	1.39	1.39	1.39	1.29	1.31	1.30	0.935	1.31
4	1.55	1.52	1.53	1.48	1.50	1.49	0.974	1.44
5	1.80	1.79	1.79	1.68	1.70	1.69	0.944	1.69
6	1.80	1.76	1.78	1.68	1.69	1.69	0.949	1.68
7	1.80	1.83	1.81	1.71	1.73	1.72	0.949	1.71
8	1.61	161	1.61	1.46	1.45	1.46	0.907	1.52
					Mean value		**0.943**	
					Standard deviation		**0.02**	
Wheat								
1	1.54	1.54	1.54	1.48	1.47	1.48	0.958	1.47
2	1.90	1.89	1.90	1.84	1.86	1.85	0.974	1.82
3	1.82	1.82	1.82	1.75	1.77	1.76	0.967	1.74
4	1.50	1.51	1.50	1.46	1.48	1.47	0.979	1.44
5	1.74	1.68	1.71	1.65	1.65	1.65	0.967	1.64
6	1.43	1.43	1.43	1.36	1.35	1.36	0.948	1.37
7	1.46	1.43	1.44	1.38	1.37	1.38	0.955	1.38
8	1.90	1.90	1.90	1.79	1.80	1.80	0.945	1.82
9	1.57	1.56	1.57	1.47	1.48	1.48	0.939	1.50
10	1.60	1.61	1.60	1.50	1.51	1.51	0.939	1.53
					Mean value		**0.957**	**± 0.014**

Barley and Wheat Malt (fine grain, double determinations)

Sample	macro N % N	% N	Mean	Kjeldahl % N	% N	Mean	Kj. / macro N	macro N corr.
BarleyMalt								
1	1.81	1.81	1.81	1.68	1.70	1.69	0.934	1.70
2	1.87	1.87	1.87	1.73	1.73	1.73	0.925	1.76
3	1.82	1.84	1.83	1.70	1.72	1.71	0.936	1.72
4	1.81	1.82	1.82	1.70	1.71	1.71	0.936	1.72
5	1.76	1.77	1.76	1.62	1.64	1.63	0.924	1.66
6	1.80	1.80	1.80	1.65	1.65	1.65	0.917	1.69
7	1.73	1.73	1.73	1.68	1.70	1.69	0.975	1.63
8	1.83	1.84	1.83	1.70	1.70	1.70	0.927	1.72
9	1.75	1.74	1.75	1.71	1.73	1.72	0.983	1.65
10	1.70	1.71	1.70	1.61	1.63	1.62	0.951	1.60
					Mean value		0.941	
					Standard deviation		0.022	
Wheat Malt								
1	1.75	1.75	1.75	1.65	1.65	1.65	0.942	1.66
2	1.86	1.87	1.87	1.73	1.75	1.74	0.932	1.77
3	1.75	1.76	1.75	1.68	1.67	1.68	0.957	1.66
4	1.85	1.86	1.85	1.74	1.74	1.74	0.940	1.75
5	1.79	1.78	1.78	1.68	1.70	1.69	0.948	1.69
6	1.88	1.92	1.90	1.82	1.84	1.83	0.957	1.80
7	1.95	1.97	1.96	1.86	1.87	1.87	0.951	1.86
8	1.76	1.76	1.76	1.71	1.69	1.70	0.965	1.67
9	1.60	1.60	1.60	1.49	1.49	1.49	0.930	1.52
					Mean value		0.947	± 0.012

Wort and Beer

Sample Preparation and Sample Weight

Before pipetting, the beer has to be totally degased by means of an ultrasonic bath or longer lasting stirring (magnetic stirror). Pipet volume for wort and beer: 3 ml.

Measuring Parameter

Operation manual Chapter 20, Example 6a

Measuring Examples (Wort, Mean Values of Double Determinations)

Sample	macro N mg/l N	Kjeldahl mg/l N	Kjeldahl / macro N	macro N corr.
1	771	756	0.9805	750
2	772	756	0.9793	751
3	756	735	0.9722	734
4	800	767	0.9588	779
5	913	893	0.9781	889
6	852	823	0.9660	829
7	877	851	0.9704	854
8	873	851	0.9748	850
9	943	938	0.9947	920
10	832	798	0.9591	810
	Conversion factor = Mean value		**0.9734 ± 0.0108**	

Special Instrument Conditions

a. Sensitive macro N version required
b. Calibration and daily factor is THAM-solution, refer Application Note No. B001E (Calibration of the Lower Measuring Range with Aqueous Standards)
c. Reduction tube filled with copper wire (Merck no. 2701). Caution, faster saturation compared to Cu powder !
   If copper powder is used: VC = approx. 2-3 %
d. Absorber tube (filled with lead chromate) made of steel instead of quartz

Coagulated and MgSO4 precipitated (?) Nitrogen

The precipitation has to be carried out by following the MEBAK instructions. The precipitate is combusted in the macro N together with the filter paper. The filter papers have to be nitrogen-free. The sample volume to be entered in the macro N computer is the original volume of the wort or the beer before precipitation. The result will be given directly in mg/l or g/l N. For the measurement of the to be coagulated nitrogen, a complete filter paper with precipitate is folded and rolled to fit into a sample crucible. It has to be avoided that a part will get above the edge of the crucible. In case of MgSO4 precipitated nitrogen, the filter and residue contains a larger amount of MgSO4. During combustion this will lead to a tight layer of MgO at the surface which is preventing the sample from regular combusition. Therefore, the filter has to be divided in two parts which are analyzed in two crucibles. The two results will be added in order to get the total. Measuring parameter: Operation Manual, Chapter 20, Example no. 43.

4. Mixed Operation

For the analysis of solid samples (raw fruits, malts, etc.) and liquid samples (wort, beer) in one and the same automatic run, the following procedure is recommended:

Specification of the analyzer in accordance with Chapter 3.2.4. For solid samples copper powder can be used (copper wire is useful as well but does have a shorter lifetime). Liquid samples shall be analyzed with copper wire.

Important: for liquid and solid samples separate crucibles have to be used !

Sequence:

3 x 400 mg RUN IN (aspartic acid)

3 x 400 mg ASPARAGIN → daily factor for solid samples

- All solid samples - (reduction tube with copper powder)

- Now install reduction tube with copper wire -

3 x 3 ml RUN IN (THAM-solution)

3 x 3 ml THAM → daily factor for liquid samples

- All liquid samples -

When liquid samples come first or only liquid samples have to be analyzed, the instrument has to be preconditioned with 3 x 400 mg aspartic acid.

5. Literature

- Operation manual macro N
- Application Note No. B001E (Calibration of Low Concentration Range with Aqueous Standards)
- MEBAK, Brautechnische Untersuchungsmethoden, 2.Aufl., Selbstverlag der MEBAK, Weihenstephan

The measuring examples were provided by Staatliche Brautechnische Prüf- und Versuchsanstalt,
8050 Freising-Weihenstephan