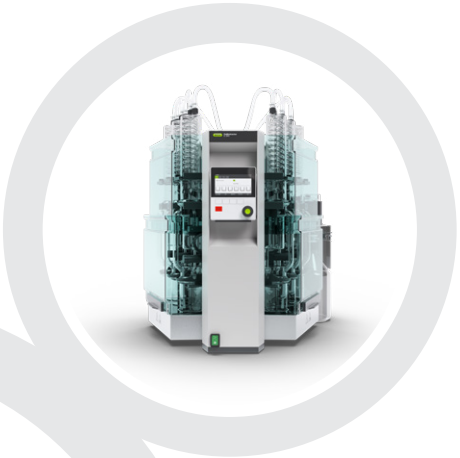
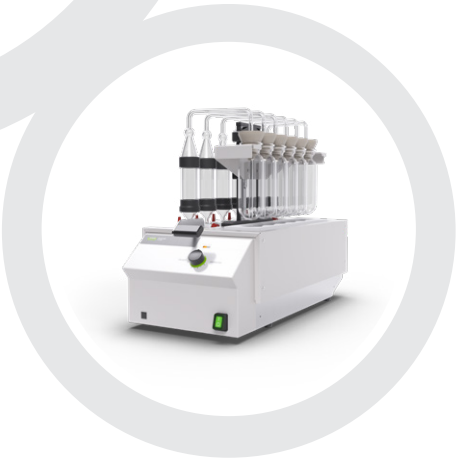
Application Note
No. 750/2021

Fat determination in ice cream

HydrolEx H-506, FatExtractor E-500:
Fat Determination in ice cream according to Weibull-Stoldt





1. Introduction

A simple and reliable procedure for fat determination of ice cream according to Weibull-Stoldt is introduced. The sample is hydrolyzed with the HydrolEx H-506. The Soxhlet extraction is performed with the FatExtractor E-500. Calculation of total fat content follows gravimetrically after the extract has been dried to a constant weight. This application follows official methods § 64 LFGB 01.00 20 2013 and DIN 10342 (1992).

2. Equipment

- HydrolEx H-506
- Suction set with vacuum pump, BUCHI (Order No. 11068473)
- FatExtractor E-500 Soxhlet, Standard Interface, with Analyte protection
- Analytical balance (accuracy ± 0.1 mg)
- Microwave oven
- Drying oven / Vacuum drying oven
- Weighing support for hydrolysis vessels, BUCHI (Order No. 11067040)

3. Chemicals and Materials

Chemicals:

- Quartz sand, particle size 0.3-0.9 mm, BUCHI (Order No. 037689)
- Celite® 545, BUCHI (Order No. 11068920)
- Hydrochloric acid 4 mol/L, 4 L HCl 32% (Hänseler, 20-2000-5) are filled up to 10 L with deionised water
- Petroleum ether, GPR RECTAPUR, boiling range 40-60 °C, 5 L, VWR (Order No. 23826.360)

For a safe handling please pay attention to all corresponding MSDS!

Samples:

- Fruit ice cream mixture with a certified fat content of 5.07 g/100 g (± 0.065 g/100 g), LVU No. 18-41
- Vanilla ice cream, labelled fat content 13%
- Chocolate covered ice cream stick, labelled fat content 18%
- Ice cream with chocolate pieces, labelled fat content 17%

The ice cream mixture was brought to ambient temperature and then homogenized by stirring for 2 minutes.

The vanilla ice cream was melted in the oven 40 °C, and then homogenized by stirring for 2 minutes.

Both ice creams samples containing chocolate were let to melt in a water bath at 90 °C, and then homogenized by stirring for 2 minutes.

4. Procedure

The fat determination according to Weibull-Stoldt includes the following steps:

- Sample homogenization
- Hydrolysis of the sample with 4 M hydrochloric acid to break up the matrix
- Filtration of the hydrolysis solution to separate the fat
- Drying of the filtered sample
- Soxhlet extraction of the fat
- Drying of the extract
- Weighing of the extract
- Calculation of fat content

4.1. Acid hydrolysis

4.1.1. Preparation of the glass sample tubes

1. Add approx. 20 g of quartz sand to the glass sample tube and compact the sand by gently tapping the glass sample tube onto the table
2. Add approx. 2 g Celite® 545 and spread it evenly using a spoon

The sand and the Celite® layer should not be mixed together. Otherwise the Celite® phase may breakthrough the frit and affect the results either by increasing the recovery or by blocking the frit.

4.1.2. Hydrolyzing the sample matrix

3. Place 2 g Celite® 545 in the hydrolysis vessel
4. Add up to 6 g of homogeneous sample[1] to the hydrolysis vessel and note the accurate weight of the sample.
5. Add 50 mL hydrochloric acid (4 M) and form a suspension by gently swirling the vessel
6. Add another 50 mL hydrochloric acid (4 M) making sure to rinse any remaining sample off the glass wall
7. Preheat the HydrolEx for 10 min
8. Insert the samples into the unit and lower the vessels
9. Connect the aspiration tubes and start the vacuum pump
10. Reduce the heat to level 2.5 when one position is boiling

Violent foaming can be prevented by adding 4 M hydrochloric acid dropwise. The degree of foaming depends on the sample and on the preheating time of the unit. Do not extend preheating excessively.

11. Hydrolyze the sample for 30 min after constant boiling is observed in each position
12. Add 50 mL of warm (50 °C) deionised water to each hydrolysis vessel at the end of the hydrolysis time
13. Switch off the heating and lift the hydrolysis vessels to the top position in order to filter the hydrolysate
14. Wash each of the vessels by gradually adding a total of at least 400 mL warm deionised water, until a neutral water pH (5-6) is reached
15. Check the pH with a pH paper on the bottom of the frit

For maximum efficiency, aspire aspirate all samples/rinsing water at the same time.

16. Stir the Celite® layers (without touching the sand layer) with a spatula to loosen the pulp
17. Carefully wipe off the spatula with a piece of tissue and add it on the top of the sample
18. Dry the glass sample tubes in a vacuum oven (2 h at 100 °C/200 mbar), in a drying oven (4 h at 100 °C) or in a microwave oven

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[1] The sample weight must be chosen according to the approximate fat content of the sample.
20-50 % 1.5-3.5 g 10-20 % 3.5-7 g <10 %: 7- 10 g
However, as ice cream samples can be foamy, it is recommended to weigh no more than 6 g.

Using a microwave oven accelerates the drying process. However, its control is more delicate. The sample can easily overheat (> 105 °C) if an inappropriate heating power is chosen. The following suggestion is valid for the drying of six hydrolyzed samples at the same time. First step: 15 min 640 W, second step: 9 min 480 W, power of microwave oven 800 W (the optimal parameters may depend on the model of microwave).

Faster drying at higher temperatures is not recommended because fat may decompose at temperatures above 105 °C. Oxidized fat can result in an excessive recovery.

19. Allow the glass sample tubes to cool down to room temperature in a desiccator
20. Add another layer of quartz sand (20 g). This prevents the Celite® from being re-suspended in the condensed solvent.

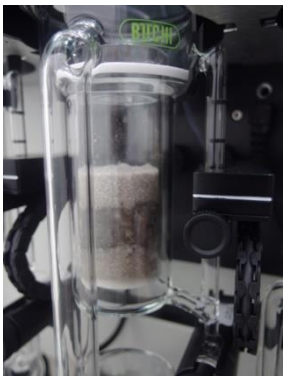
4.2. Fat extraction

4.2.1. Preparation of the beakers

Always use dry and clean beakers for the Soxhlet extraction. Dry them for at least 30 min at 102 °C. Let them cool down to ambient temperature in a desiccator for at least 1 h. Record the exact weight prior to extraction.

4.2.2. Soxhlet Extraction

Put the glass sample tubes into the extraction chamber and adjust the level sensor with the white line to the center of the upper sand layer. See Picture 1.



Picture 1: Adjusting the level sensor for the Soxhlet Extraction

Fill the solvent directly into the beakers and place them on their corresponding heating plate. Close the safety shield and lower the rack. Alternatively, fill in the solvent by the condensers after lowering the rack. Activate the occupied positions, open the cooling water or switch on the connected chiller and start the extraction according to the parameters listed in Table 1.

Table 1: Parameters for the Soxhlet Extraction with the FatExtractor E-500

Step	Time [min] / No. of cycles [-]	Heating level [-]
Solvent	Petroleum ether	
Extraction	20 cycles	6
Rinse	5 min	6
SmartDrying	on[2]	-
Solvent volume [mL]	100	

[2] Instead of using SmartDrying it is possible to set a drying time of 12 minutes and heating level 5 for petroleum ether.

4.2.3. Drying of the extract

Dry the beakers containing the extract in a drying oven at 102 °C until a constant weight is reached. Let the beakers cool down to ambient temperature for at least 1 h in a desiccator and record the weight.

Make sure that the cooling down time of the beakers in the dessicator is the same before and after extraction. Differences in beakers' temperature falsify the results.

4.3. Calculation

The results are calculated as percentage of the fat according to equation (1).

$$\%\,Fat = \frac{(m_{Total} - m_{Beaker})}{m_{Sample}} \bullet 100\% \qquad (1)$$

% Fat: Percentage of fat in the sample
m_{Total}: Beaker + extract [g]
m_{Beaker}: Empty beaker weight [g]
m_{Sample}: Sample weight [g]

5. Results

Determined fat content for the reference material corresponds well with the certified value. Also, the fat contents determined for the commercial samples are in line with the labelled values, with low relative standard deviations.

The complete findings are summarized in Tables 2-5.

Table 2: Fruit ice cream mixture, LVU No. 18-41 (Specification: 5.07 ± 0.065 g/100 g)

	m_{Sample} [g]	m_{beaker} [g]	m_{total}[g]	% Fat
Sample 1	6.0460	109.7221	110.0289	5.07
Sample 2	5.7703	110.8876	111.1786	5.04
Sample 3	5.8745	110.2489	110.5471	5.08
Average [%]				**5.06**
rsd [%]				0.37

Table 3: Vanilla ice cream, labelled fat content 13%

	m_{Sample} [g]	m_{beaker} [g]	m_{total}[g]	% Fat
Sample 1	4.8395	111.1267	111.7257	12.38
Sample 2	4.9432	110.9282	111.5381	12.34
Sample 3	4.8235	108.4376	109.0340	12.36
Average [%]				**12.36**
rsd [%]				0.16

Table 4: Ice cream with chocolate pieces, labelled fat content 17%

	m_{Sample} [g]	m_{beaker} [g]	m_{total}[g]	% Fat
Sample 1	4.0410	108.1300	108.7717	15.88
Sample 2	3.7537	111.1053	111.7054	15.99
Sample 3	3.8179	111.2585	111.8667	15.93
Average [%]				**15.93**
rsd [%]				0.34

Table 5: Chocolate covered ice cream, labelled fat content 18%

	m_{Sample} [g]	m_{beaker} [g]	m_{total}[g]	% Fat
Sample 1	3.6317	108.1185	108.7493	17.37
Sample 2	2.8807	110.9007	111.3984	17.28
Sample 3	3.7063	111.6421	112.2837	17.31
Average [%]				**17.32**
rsd [%]				0.27

6. Conclusion

The determination of fat in ice cream samples using the HydrolEx H-506 and the FatExtractor E-500 provides reliable and reproducible results. These results correspond well to the labelled values, with low relative standard deviations (rsd). With the FatExtractor E-500 Soxhlet, the time per cycles is reduced significantly. The programmed 20 cycles are accomplished in approximately 70 min.

7. References

[1] § 64 LFGB 01.00 20 (2013) Bestimmung des Fettgehaltes von Milch und Milchprodukten nach dem gravimetrischen Weibull-Berntrop-Verfahren

[2] DIN 10342 (1992) Bestimmung des Fettgehaltes von Milch und Milchprodukten nach dem gravimetrischen Weibull-Berntrop-Verfahren

Extraction Reports App

Operation Manual of HydrolEx H-506
Operation Manual of FatExtractor E-500