

# Saccharine in table-top Sweeteners

(according to EN 1376 and German Food and Feed Code §64 LFGB 57.22.99-2)

## Note

Pursuant to the valid copyright regulations this application note contains only a rough description of the content of the official method followed by a detailed description of the specific measurement procedure with the Spectroquant® Prove Spectrophotometers. A detailed description of the method specific handling steps can be found in the official method EN 1376 <sup>[1]</sup> and German Food and Feed Code §64 LFGB 57.22.99-2 <sup>[2]</sup>.

## Method

Sweeteners like saccharin are often used in calorie-reduced food and beverage products due to their low nutritional value whilst having at the same time a high sweetener power. Saccharine is the oldest and best-known sweetener with a sweetener power of approx. 200 – 400 times of sucrose. The chemical name for saccharine is Benzoic sulfimide <sup>[3]</sup>.

The Saccharin content is determined photometrically in the absorbance maximum at 270 nm. This method is based on the official method EN 1376 <sup>[1]</sup> and German Food and Feed Code §64 LFGB 57.22.99-2 <sup>[2]</sup> and describes the determination of Saccharine in table-top sweeteners.

## Measuring range

Description		
<b>Method 2536</b>	Saccharine EN 1376	0.0 – 1200.0 mg/g

## Sample material

- Table-top sweeteners



## Reagents and auxiliaries

Cat. No.	Description
<b>1.73027</b>	Spectroquant® UV/VIS Spectrophotometer Prove 300 plus
<b>1.73028</b>	Spectroquant® UV/VIS Spectrophotometer Prove 600 plus
<b>100784</b>	Rectangular cells quartz 10 mm
<b>814114</b>	Saccharin sodium salt dihydrate for synthesis (≥99.0%)
<b>109141</b>	Sodium hydroxide solution 0.1 mol/l (0.1N) Titripur®

Also first generation Prove instruments are compatible and preprogrammed with this method.

## Additional needs

- Mortar and pestle
- Analytical balance
- Drying cabinet (optional)
- Volumetric flasks, 50-mL, 100-mL, 250-mL, 500-mL
- Standard laboratory glassware (e. g. glass beakers) and pipettes
- Analytical balance.

## Sample preparation

- Pulverize sample finely in a mortar

## Procedure

### Saccharine determination

#### Reagent blank

Sodium hydroxide solution 0.1 mol/l (0.1 N).

#### Sample

Weigh pulverized sample to a volumetric flask and dissolve it according to EN 1376 <sup>[1]</sup> resp. German Food and Feed Code §64 LFGB 57.22.99-2 <sup>[2]</sup>.

#### Note

*The sample weight should approx. be equivalent to an amount of 30 mg Saccharin sodium salt resp. 35 mg Saccharin sodium salt dihydrate.*

### Measurement

#### Note

*It is advisable to measure the reagent blank and the sample using the same cell as the one used for the zero adjustment or else a cell with identical optical characteristics and an identical absorption (matched pair).*

- Open the methods list (<Methods>) and select Method No. 2536 "Saccharine EN 1376".
- The instrument automatically prompts a "Zero adjustment".
- For the zero adjustment fill a clean and dry 10-mm rectangular cell with distilled water.
- After prompting, insert the filled rectangular cell into the cell compartment. The zero adjustment is performed automatically.
- Confirm the performance of the zero-adjustment procedure by clicking on <OK>
- A window with an input field to enter the sample weight pops up.
- Enter the weight of the sample in milligram (mg), accurate to 0.1 milligram (mg), confirm with <OK> and click on <START> to switch to the measurement procedure.

#### Note

*It is possible to enter a sample weight in a range of 1.0 to 3000.0 mg.*

- Fill the prepared reagent blank into a clean and dry 10-mm rectangular cell. Insert the cell into the cell compartment. The measurement is performed automatically. A (✓) symbol appears behind the cue "Insert Reagent Blank".
- Confirm the measurement by clicking on <OK>.
- Finally fill the prepared sample solution into a clean and dry 10-mm rectangular cell. Insert the cell into the cell compartment. The measurement is

performed automatically. A (✓) appears behind the cue "Insert Sample".

- Confirm the measurement by clicking on <OK>.
- Read off the result in mg/g, the absorption for the reagent blank ( $A_{RB}$ ) and the sample ( $A_{Sample}$ ) and the ratios  $A_{255}/A_{270}$  and  $A_{285}/A_{270}$  for the sample measurement from the display.
- Tap the <START> button to start the measurement procedure for the next sample.

#### Note

*The absorption ratios  $A_{255}/A_{270}$  and  $A_{285}/A_{270}$  for the sample measurement are useful to check the plausibility of the measurement. With these ratios it is possible to identify if the measurement is influenced by interfering substances.*

*Typical values for pure saccharin sodium salt in Sodium hydroxide solution 0.1 mol/l are*

$$\text{Ratio } A_{255}/A_{270} = 0.83$$

$$\text{Ratio } A_{285}/A_{270} = 1.55$$

*For further details see also EN 1376 <sup>[1]</sup> resp. German Food and Feed Code §64 LFGB 57.22.99-2 <sup>[2]</sup>.*

### Evaluation

Statement of the results:

- Saccharine-Na [mg/g]
- Absorption of reagent blank  $A_{RB}$
- Absorption of sample  $A_{Sample}$
- Ratio  $A_{255}/A_{270}$
- Ratio  $A_{285}/A_{270}$

#### Note

- *To recalculate the result to mg/g Saccharine sodium salt use recalculation factor 1.175.*
- *To recalculate the result to mg/g Saccharine use recalculation factor 0.893.*

### Method control

- The method can be checked using **Cat. No. 814114** Saccharin sodium salt dihydrate for synthesis (≥99.0%) as standard substance
- Prepare a stock solution with 1000 mg/l **Saccharin sodium salt** by dissolving 280.0 mg **Saccharin sodium salt dihydrate** in approx. 220 mL Sodium hydroxide solution 0.1 mol/l. Transfer the solution completely to a 250-mL volumetric flask and fill up to the mark with Sodium hydroxide solution 0.1 mol/l.
- Dilute the stock solution to 120 mg/l **Saccharin sodium salt** with Sodium hydroxide solution 0.1 mol/l (30 mL stock solution ad 250 mL in a 250-mL volumetric flask).

- Analyze the prepared standard as described in section "Measurement".
- Hereby enter a sample weight of 30.0 mg.
- The target value is 1000 mg/g

## Adjustment

- In case of significant deviations in the method control procedure the preprogrammed factor or the current factor used in the calculation of the displayed results can be adjusted by the user.
- The corrected factor must be recalculated as follows:  
**Factor corrected = Current factor x (target value standard / measured value standard)**

- To edit the preprogrammed factor, select method 2536 from <Methods>.
- Close the window for the "Zero adjustment" by clicking on <X>.
- Close the input field for the sample weight by clicking on <X>
- Click <Settings> and select the list "FACTORS".
- Tip on the input field "Factor", enter the corrected factor and confirm by clicking on <OK>.
- Close the window for the "Zero adjustment" by clicking on <X>.
- For the next measurement restart the method by selecting the method anew from <Methods>.

## Note

- To find the used factor, select Method 2536 from <Methods>.
- Close the window for the "Zero adjustment" by clicking on <X>.
- Close the input field for the sample weight by clicking on <X>.
- Click <Settings> and select the list "FACTORS".

For more information visit,  
**[SigmaAldrich.com/photometry](http://SigmaAldrich.com/photometry)**

## Literature

1. Foodstuffs - Determination of saccharin in table top sweetener preparations - Spectrometric method; EN 1376:1996.
2. German Food and Feed Code §64 LFGB 57.22.99-2:1998 Bestimmung von Saccharin in Tafelsüßen
3. Matissek R., Steiner G., Fischer M.; (2010): Lebensmittelanalytik, 4.Auflage, Springer-Verlag, Berlin Heidelberg.

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