Fat and Oil

Formol Nitrogen in Hydrolyzed Protein

Acid-base titration by
Automatic Potentiometric Titrator

Abstract

Formol nitrogen in hydrolyzed protein (HAP) is measured by titrating with 0.1mol/L sodium hydroxide up to pH 8.5 by adding neutral formaldehyde after the first titration with 0.1mol/L sodium hydroxide to pH 8.5.

The amount of formol nitrogen is calculated from the difference in titration volume of 0.1mol/L sodium hydroxide between the first and second endpoint.

Reference

1) Test method prescribed by National Tax Administration Agency

Cautions in measurement

1) Neutralize formaldehyde before use in measurement.
2) Take due care when you handle chemicals.
4. Post-measurement care

Clean the electrode with pure water, and keep it dipped in water for use in next measurement.

5. Test equipment

Main unit: Automatic potentiometric titrator (Standard preamplifier: STD−)
Electrode: standard Combination glass electrode
standard Temperature compensation electrode

6. Reagent

Titrant: 0.1mol/L sodium hydroxide solution (f=1.004)
Reagent: Pure water, neutral formaldehyde solution

7. Measurement procedure

—Pretreatment—
1) To make neutral formaldehyde, add 0.1mol/L sodium hydroxide to pH=8.5.
2) Add pure water to 5.0mL sample making it 250mL in total.

—Measurement—
1) Transfer 25.0mL sample liquid to a 50mL tall beaker.
2) Titrate with 0.1mol/L sodium hydroxide up to pH=8.5.
3) Add 20.0mL of neutral formaldehyde solution.
4) Titrate with 0.1mol/L sodium hydroxide up to pH=8.5 to obtain formol nitrogen.

8. Formula

Formol nitrogen (W/V%) = (EP2 − EP1) × TF × Cl × K1 / (SIZE × C2)
EP1: First endpoint titration volume (mL)
EP2: Second endpoint titration volume (mL)
TF: Factor of titrant (1.004)
Cl: Concentration conversion coefficient (0.0014mg/mL)
K1: Unit conversion coefficient (100)
SIZE: Sample size (mL)
C2: Dilution ratio (0.1)
### 9. Example of measurement

--- Ambient condition ---

| Room temperature: 22.0 °C | Humidity: 24 % | Weather: Fair |

### Titration parameter

<table>
<thead>
<tr>
<th>Model: AT-510</th>
<th>Method No.: 50 (Combine)</th>
<th>Number of Method: 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>No.1 Method No.: 20</td>
<td>No.2 Method No.: 21</td>
<td></td>
</tr>
</tbody>
</table>

Method No.: 20
**<Auto Titration>**

- **<Titration>**
  - Form: Level Stop
  - APB No.: 1
  - Unit No.: 1
  - Detector No.: 1
  - Unit: pH
  - Max.Volume: 20.0mL
  - Wait Time: 0s
  - Direction: Auto

- **<Control>**
  - End Point No.: 1
  - 1st Level: 8.5pH
  - Over Titr.Vol.: 0mL
  - Gain: 1
  - Data samp.Pot.: 4.0mV
  - Data samp.Vol.: 0.5mL
  - Control Speed: Medium

- **Calculation**
  - Conc.2: Set
  - CO2=(EP2-EP1)*TF*C1*K1/SIZE*C2

Method No.: 21
**<Auto Titration>**

- **<Titration>**
  - Form: Level Stop
  - APB No.: 1
  - Unit No.: 1
  - Detector No.: 1
  - Unit: pH
  - Max.Volume: 20.0mL
  - Wait Time: 60s
  - Direction: Auto

- **<Control>**
  - End Point No.: 1
  - 1st Level: 8.5pH
  - Over Titr.Vol.: 0mL
  - Gain: 1
  - Data samp.Pot.: 4.0mV
  - Data samp.Vol.: 0.5mL
  - Control Speed: Medium

### Calculation

- **Calc.Type:** Sample
- **Conc.1:** EP1
- **Unit:** ml
- **EP No.:** 1

#### Temp.Comp.: Off

- **C1(mg/mL):** 0.0014
- **C2(mg/mL):** 0.1
- **K1:** 100

#### Result: calculation parameter

- **Calc.Type:** Sample
- **Conc.1:** EP1
- **Unit:** ml
- **EP No.:** 1

- **Temp.Comp.:** Off

- **C1(mg/mL):** 0.0014
- **C2(mg/mL):** 0.1
- **K1:** 100

(The above printout data were obtained from titration by AT-510)
### Measurement results

<table>
<thead>
<tr>
<th>n</th>
<th>Sample (mL)</th>
<th>Titration (mL) (EP2-EP1)</th>
<th>Formol nitrogen (W/V%)</th>
<th>Formol nitrogen batch processed</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>5.0</td>
<td>3.6149</td>
<td>1.0162</td>
<td>Mean 1.0138 W/V%</td>
</tr>
<tr>
<td>2</td>
<td>5.0</td>
<td>3.5888</td>
<td>1.0089</td>
<td>SD 0.0043 W/V%</td>
</tr>
<tr>
<td>3</td>
<td>5.0</td>
<td>3.6155</td>
<td>1.0164</td>
<td>RSD 0.4215 %</td>
</tr>
</tbody>
</table>

*The above results were obtained by 3 tests of the same sample.

* Red underline shows the data from page 3/4.

### 10. Summary

Hydrolyzed protein is the amino acid combination used for material of “tasty quality” additive in cooking. This is not food additive pertinent to food hygiene law but ought to indicate the name required by JAS.

Formol nitrogen is a family member of amino acids.

The sample measurement shows a good repeatability with 0.4% relative standard deviation. Precise and reliable measurement is assured by the automated potentiometry.

The analysis of hydrolyzed protein can be perfectly made by any of the following titration systems manufactured by Kyoto Electronics (KEM).

**[AT-610]**
- Awarded Product of Supreme Technology from Kyoto City
- Easy key entry by touch panel of large color LCD (8-inch wide)
- Simultaneous titration in parallel
- Both potentiometric and Karl Fischer moisture titration (coulometric • volumetric) can be performed at a time.

**[AT-510]**
- Compact and cost performance model
- PC card expands data memory for convenience and versatility.

**[AT-500N-1]**
- Low cost and high performance
- Easy view with back light LCD
- GLP/GMP conformed model

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