



Titratable Acidity and Formol Number in Fruit Juices

Introduction

The formol number, or formol titration, measures the total amino acid (NH₂-R-COOH) concentration. Using 2 successive end point titrations, it is possible to determine total titratable acidity and formol number.

Principle

The determination of formol number takes place in three steps:

- 1) Neutralisation of titratable acidity by means of an end point titration at pH 8.2 with NaOH 0.1 eq/l
- 2) Addition of an excess of formol (HCHO) to the solution. This operation locks the NH₂ groups of amino acids due to the decrease in pH and allows titration of the COOH groups of amino acids with an end point titration at pH 8.2
- 3) Second endpoint titration at pH 8.2 to determine total amino acid content. The result is then expressed as milliequivalents/100 ml or milliequivalent/l

Electrode and Reagents

pHC2401-8 Combined pH Electrode (part no. E16M400)

NaOH 0.1 eq/l solution in distilled water (see separate application note)

Distilled water

Check the pH of the distilled water. Using the titration manager as a pH meter, adjust the distilled water to pH 8.2, by means of few drops of base or acid solution, with NaOH 0.1 as titrant

Formol (HCHO) solution at 37% adjusted to pH 8.2

IUPAC Series pH standards

pH 4.005 (part no. S11M002) or pH 7.000 (part no. S11M004) and pH 10.012 (part no. S11M007)

End Point titration settings

Two linked methods

First method (can be used as titratable acidity)

Burette volume:	10 ml
Maximum volume:	30 ml
Stirring speed:	400 rpm
Working mode:	pH
Number of end points:	1
End point:	8.20 pH
Stirring delay:	30 seconds

Minimum speed:	0.2 ml/min
Maximum speed:	10 ml/min
Proportional band:	3.00 pH
End point delay:	10 seconds
Sample unit:	ml
Sample amount:	10
Result expression:	g/l (of tartaric acid or citric acid)
Titration:	Increasing pH

Second method (Formol number determination)

Burette volume:	10 ml
Maximum volume:	10 ml
Stirring speed:	400 rpm
Working mode:	pH
Number of end points:	1
End point:	8.20 pH
Stirring delay:	60 seconds
Minimum speed:	0.2 ml/min
Maximum speed:	5 ml/min
Proportional band:	2.00 pH
End point delay:	10 seconds
Sample unit:	ml
Sample amount:	10
Result expression:	meq/100 ml
Titration:	Increasing pH

Procedure

Connect the electrode to the electrode input.

Calibrate the electrode using the two IUPAC standards above.

Stir and pipette 10 ml of sample.

Always dilute the sample with the same volume of distilled water (25 ml for example).

Dip electrode and delivery tip in the solution

Start titration by pressing the RUN key.

At the end of the first method manually add 5 ml of HCHO solution for 10 ml of sample to the sample solution.

Results

For titratable acidity expressed as g/l of tartaric acid (HOOC-(CHOH)₂-COOH)

Result is normally expressed as g/l of tartaric acid (MW= 150.09 g/mol and 2 acid functions).

As 2 molecules of NaOH react with 1 molecule of tartaric acid:

$$R = V(\text{titr}) * C(\text{titr}) * 150.09 / V(\text{smp}) * 2$$

-V(titr) = total volume of titrant to reach the end point in ml

-C(titr) = Titrant concentration in eq/l (currently 0.1)

-V(smp) = sample volume 150.09 = molecular weight of tartaric acid

2 = Ratio between titrant and sample

For titratable acidity in g/l of tartaric acid

Enter in the first method

The actual sample amount in the SAMPLE screen in ml

The titrant concentration in the TITRANT screen in mol/l or eq/l

2 Titrants and 1 Sample in the COEFFICIENTS display
150.09 as molecular weight

The Titration Manager gives a result according the above formula.

For titratable acidity in g/l of citric acid (that has 3 acid functions with a MW of 192.4)

Enter in the first method

The actual sample amount in the SAMPLE screen in ml

The titrant concentration in the TITRANT screen in mol/l or eq/l

3 Titrants and 1 Sample in the COEFFICIENTS display
192.4 as molecular weight

The Titration Manager gives a result according the above formula.

For Formol number expressed as meq/l

$$\text{Formol no.} = V(\text{titr}) * C(\text{Titr}) * 1000 / V(\text{smp})$$

-V(titr) = total volume of titrant to reach the end point in ml

-C(titr) = Titrant concentration in eq/l or mol/l (currently 0.1)

-V(sample) = sample volume in ml

For formol number in meq/100ml

Enter in the second method

The actual sample amount in the SAMPLE screen in ml

The titrant concentration in the TITRANT screen in mol/l or eq/l

1 Titrant and 1 Sample in the COEFFICIENTS display (if necessary)

Enter in the RESULT screen

Result: 1
Unit: eq/l
Equation: 1
Formula: R1/10
Name: meq/100 ml

The Titration Manager gives 2 results:

Result expressed in meq/l

Result from equation formula, expressed in meq/100 ml

3 determinations on grapefruit juice

Acidity

Mean: 15.28 g/l tartaric acid

Standard deviation: 0.02 g/l tartaric acid

Rel. standard deviation: 0.13%

Formol number

Mean: 2.19 meq/100 ml

Standard deviation: 0.038 meq/100 ml

Rel. standard deviation: 1.7%

Working Range

Related to the calculation formula, using 10 ml for sample volume and 0.1 eq/l titrant concentration. For acidity determination 1 ml of titrant corresponds to 0.75 g/l of tartaric acid.

For formol number determination:

$$\text{Formol no. (meq/100 ml)} = V(\text{titr}) \text{ in ml}$$

For a 10 ml burette, formol number range can be estimated between 0.2 (experimental low limit corresponding to 0.2 ml) and 10 (nominal value of cylinder capacity):

$$\text{Formol no. (meq/l)} = \text{Formol no. (meq/100 ml)} * 10$$

Notes

Depending on local procedures, the first end point can vary between pH 8.0 and pH 8.4.

The most commonly used value is pH 8.2. We use this value in this application.

It is also possible to have different values for the pH end points (for example, pH 8.1 for the first and pH 8.4 for the second).

Adjust the pH of the formol solution to the same value as that used in the second end point titration (formol number determination).