

Food

## Amino Acid of Sake Wine

Acid-base titration by  
Automatic Potentiometric Titrator

Standard

Test method by National  
Tax Administration

### 1. Abstract

Japanese sake contains amino acids, of which known variety amounts to approximately 20 kinds. To quantify total amino acids, here we demonstrate Formol titration based on Test method for Total acid, Amino acid of Sake and Synthetic wine prescribed by National Tax Administration Agency.

Formol titration starts with 0.1mol/L sodium hydroxide for reaching pH 8.2. Then, add neutral formaldehyde. Again titration goes with 0.1mol/L sodium hydroxide to pH 8.2.

Titration volume after neutral formalin added is converted to glycine in order to obtain amino acids.



### 2. Reference

- 1) Test method for Total acid, Amino acid of Sake and Synthetic wine prescribed by National Tax Administration Agency

### 3. Cautions in measurement

- 1) Use a combination glass electrode for micro titration since 10mL sample is titrated without dilution.
- 2) It is recommended to use not only electrode but titration cell unit for micro titration in order to avoid the effect of carbon dioxide gas in the air.
- 3) Adjust pH of neutral formalin each time used in titration.
- 4) Obtain factor of titrant 0.1mol/L sodium hydroxide using sulfuric amide according to JIS K8001 General Rule of Test Method and ISO 6531-1.
- 5) Store titrant 0.1mol/L sodium hydroxide in zeolite to prevent ambient carbon dioxide from permeation.
- 6) Pure water used for dilution must be degassed of carbon dioxide.
- 7) Filter the sample to remove fruits particles in order to obtain good repeatability.

## 4. Post-measurement care

After the electrode is rinsed with pure water, keep its tip dipped in a beaker filled with pure water in order to avoid it from drying up.

## 5. Test equipment

Main unit : Automatic potentiometric titrator (Standard preamplifier: STD—)  
Electrode :  Option Combination glass electrode (for micro titration)  
 Standard Temperature compensation electrode  
Option : Micro titration cell unit

## 6. Reagent

Titrant : 0.1mol/L sodium hydroxide (f=1.00)  
Additive : Neutral formalin  
After 50mL formalin is adjusted with 0.1mol/L sodium hydroxide to pH8.2, add pure water to make it 100mL in total.

## 7. Measurement procedure

—Measurement—

- 1) Sample 10mL in micro titration cell.
- 2) Titrate with 0.1mol/L sodium hydroxide to pH8.2.  
This titration is not accounted for target calculation.
- 3) Add 5mL of neutral formalin.
- 4) Again titrate with 0.1mol/L sodium hydroxide to obtain amino acids.  
The same method can be used for step 2) and 4). Only 4) is significant for quantification.

## 8. Formula

Amino acid ( g/100mL ) = EP1 × TF × C1 × K1

EP1 : Titration volume ( mL )

TF : Factor of titrant ( 1.00 )

C1 : Concentration conversion coefficient (7.5 mg/mL)  
(Glycin (mg) equivalent to 1mL of 0.1mol/L NaOH )

K1 : Unit conversion coefficient ( 0.01 )

## 9. Example of measurement

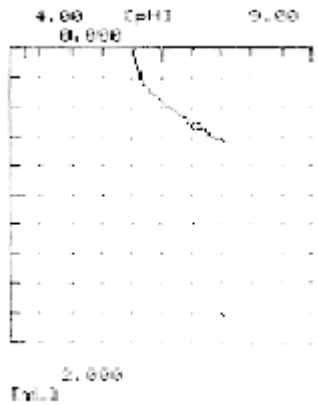
— Ambient condition —

Room temperature : 23.0 °C	Humidity : 47 %	Weather : Cloudy
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### -Titration parameter-

Model : AT-510	
Method No. : 32	
<Auto Intermit>	
<Titration>	<Calculation>
Form : Level Stop	Calc.Type : Sample
APB No. : 1	Conc.1 : Set
Unit No. : 1	CO1=EP1*TF*C1*K1
Detector No. : 1	
Unit : pH	Unit : g/100mL
Max. Volume : 20.0mL	EP No. : 1
Wait Time : 0s	Temp.Comp. : Off
Direction : Positive	
	<Constant>
	C1(mg/mL) : 7.5
	K1 : 0.01
<Control>	[Titr. constant]
End Point No. : 1	Factor : 1.00
1st Level : 8.2pH	
Over Titr.Vol. : 0mL	
Gain : 1	
Data samp.Pot. : 4.0mV	
Data samp.Vol. : 0.5mL	
Stability : 0.5mV/s	
Delay Time : 1s	
Limit Time : 30s	

### -Titration curve-

*** Result ***	
Sample No. : 69-01	
Date : 1999/08/16 16:37	
Sample ID :	
Method No. : 32	
<Auto Intermit>	
Method Name :	
Titr. Time : 00:02:30	
Size : <u>10.0mL</u>	
Conc-1 : <u>0.0448 g/100mL</u>	
End point-1	
Volume : <u>0.5977mL</u>	
Potential : 8.20pH	
	

(The above parameters and titration curve are printed out by AT-510)

#### «Titration parameter»

Form: of titration / APB No. the burette used in titration / Unit No.: APB Unit File number used in titration  
 Detector No.: the detector used in titration / Max Volume. of titration / Wait Time: before titration starts  
 Direction.: of titration

#### «Control parameter»

End Point No. number of EPs detected / 1st Level: potential of the first EP / Over Titr.Vol. over-titration volume  
 Gain: sensitivity of detection signal / Data samp.Pot.: potential changes of sampling signal / Data samp.Vol.: titration volume of sampling signal / Control Speed: of titration

#### «Result parameter»

Calc.Type: of formula / Conc.x formula 1 / Unit of result / EP No. for calculation  
 Temp.Comp. temperature compensation of titration liquid/ C1(mg/mL) concentration conversion coefficient  
 K1: unit conversion coefficient / Factor: of reagent / Blank 1: blank level 1

–Measurement results–

n	Sample (mL)	Titration (mL)	Amino acid (g/100mL)	Batched processed amino acid	
1	<u>10.0</u>	<u>0.5977</u>	<u>0.0448</u>	Mean	0.0443 g/100mL
2	10.0	0.5889	0.0442	SD	0.0005 g/100mL
3	10.0	0.5842	0.0438	RSD	0.8493 %

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

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

## 10. Summary

Japanese sake is made from fermented cooked rice, which is one of her traditional alcoholic beverages.

In the broad sense of meaning particularly in chemistry, Amino acid is the organic compound of functional group having both amino group and carboxyl group.

Quality control and evaluation of sake depends on total acids including amino acid of which measurement is of extreme importance.

The sample measurement shows a good repeatability with less than 0.9% relative standard deviation. Precise and reliable measurement is assured by the automated potentiometry.

The analysis of alcoholic beverage like sake 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

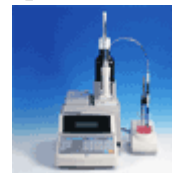
- Easy key entry by touch panel of large color LCD (8-inch wide)
- Simultaneous titration in parallel
- Both potentiometric and Karl Fischer moisture titration (coulometric·volumetric) can be performed at a time.

### 【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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