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Incorporated Administrative Agency

Food and Agricultural Materials Inspection Center

Tentative Translation

Japanese Agricultural Standards for Soy Sauce (*Shoyu*)

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(Scope)

Article 1: This standard applies to *Koikuchi Shoyu*, *Usukuchi Shoyu*, *Tamari Shoyu*, *Saishikomi Shoyu* and *Shiro Shoyu* in the type of soy sauce.

(Terms and Definitions)

Article 2: In this standard, definition of the terms listed in the left column of the following table is provided in the right column of the table.

Terms	Definition
Soy sauce (<i>Shoyu</i>)	<p>Following products (including products which is made with auxiliary use of ingredients such as sugars (including molasses, but excluding sugar alcohol; the same applies hereinafter), alcohol, etc.):</p> <ol style="list-style-type: none">clear liquid seasoning made from <i>Moromi</i> (mash) by fermentation and aging processes. <i>Moromi</i> (mash) is prepared by adding brine or <i>Kiage</i> (raw soy sauce) to <i>Shoyu Koji</i>, <i>Shoyu Koji</i> with steamed or puffed rice or <i>Shoyu Koji</i> with steamed rice saccharified by <i>Koji</i> mold (hereinafter referred to as the definition of <i>Moromi</i>). <i>Shoyu Koji</i> is cultured mold on prepared soybeans (including defatted soybeans; the same applies hereinafter) or prepared soybeans with grains (including a thing which is added wheat gluten to) such as wheat, rice by steaming and/ or other method (hereinafter referred to as the definition of <i>Shoyu Koji</i>). (including products which made with auxiliary use of enzyme such as cellulose (for proteolytic enzyme, limited to the one which is used during the heating process of <i>Kiage</i> in order to prevent cloudiness by the substance mostly consist of the protein of <i>Shiro Shoyu</i>) in the manufacturing process; hereinafter referred to as “Soy sauce made by the regular fermenting method”);clear liquid seasoning made from fermented and aged <i>Moromi</i> which added the hydrolyzed vegetable protein (vegetable protein of soybeans, etc. hydrolyzed by acid; the same applies hereinafter), the enzymatic hydrolyzed vegetable protein (vegetable protein soybeans, etc. hydrolyzed by proteolytic enzyme; the same applies hereinafter) or the fermentative hydrolyzed vegetable protein (wheat gluten hydrolyzed by fermentation; the same applies hereinafter) to (hereinafter referred to as “Soy sauce made by the mixing-fermenting method”);the mixture which added the hydrolyzed vegetable protein, the enzymatic hydrolyzed vegetable protein or the fermentative hydrolyzed vegetable protein, or a combination of above more than two into the preceding clause 1, 2 or <i>Kiage</i>, or a combination of the above more than two, (hereinafter referred to as “Soy sauce made by the mixing method”).

<i>Koikuchi Shoyu</i> (Common soy sauce)	Soy sauce, those using soybeans with wheat of almost the same quantity as soybeans or these mixture with grains such as rice, as the ingredient of <i>Shoyu Koji</i>
<i>Usukuchi Shoyu</i> (Light color soy sauce)	Soy sauce, those using soybeans with wheat of almost the same quantity as soybeans or these mixture with grains such as rice or with wheat gluten, as the ingredient of <i>Shoyu Koji</i> , and those using the <i>Moromi</i> prepared with or without steamed rice, puffed rice or these rice saccharified by <i>Koji</i> mold, and being controlled in the manufacturing process so that the color is not deepened
<i>Tamari Shoyu</i>	Soy sauce, those using soybeans or soybeans with a small quantity of wheat or these mixture with grains such as rice, as the ingredient of <i>Shoyu Koji</i>
<i>Saishikomi Shoyu</i> (Refermented soy sauce)	Soy sauce, those using soybeans with wheat of almost the same quantity as soybeans or these mixture with grains such as rice, as the row material of <i>Shoyu Koji</i> , and using the <i>Moromi</i> prepared with <i>Kiage</i> instead of brine
<i>Shiro Shoyu</i> (Extra light color soy sauce)	Soy sauce, those using small quantity of soybeans with wheat or these mixture with wheat gluten, as the ingredient of <i>Shoyu Koji</i> and being extremely controlled in the manufacturing process so that color is not deepened
<i>Kiage</i> (Raw soy sauce)	Raw liquid pressed from the fermented and aged <i>Moromi</i>
A use ratio of hydrolyzed vegetable protein, etc.	A ratio of the total amount of nitrogen in hydrolyzed vegetable protein, enzymatic hydrolyzed vegetable protein and fermentative hydrolyzed vegetable protein using as the ingredients for the total amount of nitrogen in final product

(The Standard for *Koikuchi Shoyu*)

Article 3: The criteria of *Koikuchi Shoyu* are as follows:

Classification	Criteria		
	Special grade	Superior grade	Normal grade
Manufacturing method	Shall be made by the regular fermenting method.	—	—
Property	Having a transparent bright madder red color, special flavor and matured taste with mellowed saltiness and umami, which are peculiar to the fermented <i>Koikuchi Shoyu</i> that enough fermentation and aging are performed, its all aspects are excellent, and being free from objectionable taste and odor and mold.	Having a transparent bright madder red color, special flavor and matured taste with mellowed saltiness and umami, which are peculiar to the fermented <i>Koikuchi Shoyu</i> that enough fermentation and aging are performed, its all aspects are good, and being free from objectionable taste and odor and mold.	Having a transparent bright madder red color, special flavor and matured taste with mellowed saltiness and umami, which are peculiar to <i>Koikuchi Shoyu</i> , its all aspects are good, and being free from objectionable taste and odor and mold.
Degree of color	Less than No.18 of the standard color for soy sauce (by the color indicating method of JIS Z8781-4 (2013), where : L=30.0, a=46.1, b=51.6 ; the same applies hereinafter);	Same as left	Less than No.18 of the standard color for soy sauce

	however, for those with sterilization process similar to heating, instead of heating, less than No.22 of the standard color for soy sauce (by the color indicating method of JIS Z8781-4, where : L=36.7, a=45.6, b=62.9 ; the same applies hereinafter)		
Total nitrogen	Not less than 1.50% (weight/volume)	Not less than 1.35% (weight/volume)	Not less than 1.20% (weight/volume)
Soluble solids excluding salt	Not less than 16% (weight/volume)	Not less than 14% (weight/volume)	—
Ingredients	<p>Not using a thing except following mentioned ingredients:</p> <ol style="list-style-type: none"> 1. soybean; 2. wheat, barley and naked barley; 3. rice; 4. adlay; 5. wheat gluten; 6. salt; 7. hydrolyzed vegetable protein, enzymatic hydrolyzed vegetable protein and fermentative hydrolyzed vegetable protein; 8. sugars; 9. alcohol, <i>Shochu</i> (rice wine spirit) and <i>Sake</i> (rice wine); 10. rice fermentative seasoning, vinegar, <i>Mirin</i> and <i>Mirin</i> type seasoning. 		
Additives	<ol style="list-style-type: none"> 1. Additives shall comply with the provisions of 3.2 of the General Standard for Food Additives (CODEX STAN 192-1995, Rev. 7-2006), which was stipulated by the Joint Food Standard Committee of the United Nations Food and Agriculture Organization and the World Health Organization. In addition, the conditions for use of such additives shall comply with the provisions of 3.3 of CODEX STAN 192. 2. The amounts of respective additives used shall be accurately recorded and the record shall be kept. 3. Information regarding the compliance of the additives with the provision of 1 shall be provided to general consumers by one of the following methods: <ol style="list-style-type: none"> (1) use of Internet for the public to view; (2) publications such as brochures and leaflets to easily attract general consumers' attention; (3) display in the store to easily attract general consumers' attention; (4) notification to the general consumers upon request from them, clearly indicating the contact address on the products. <p>In case of the products for business use, the above may be omitted.</p> 		
Net contents	The net contents shall be consistent with the one indicated on the label.		

(The Standard for *Usukuchi Shoyu*)

Article 4: The criteria of *Usukuchi Shoyu* are as follows:

Classification	Criteria		
	Special grade	Superior grade	Normal grade
Manufacturing method	Same as specified in the manufacturing method in the preceding Article.	—	—
Property	Having a light madder red color including yellow, special flavor and matured taste with mellowed saltiness and umami, which are peculiar to the fermented <i>Usukuchi Shoyu</i> that enough fermentation and aging are performed, its all aspects are excellent, and being free from objectionable taste and odor and mold.	Having a light madder red color including yellow, special flavor and matured taste with mellowed saltiness and umami, which are peculiar to the fermented <i>Usukuchi Shoyu</i> that enough fermentation and aging are performed, its all aspects are good, and being free from objectionable taste and odor and mold.	Having a light madder red color including yellow, special flavor and matured taste with mellowed saltiness and umami, which are peculiar to <i>Usukuchi Shoyu</i> , its all aspects are excellent, and being free from objectionable taste and odor and mold.
Degree of color	Not less than No.22 of the standard color for soy sauce	Same as left	Not less than No.18 of the standard color for soy sauce
Total nitrogen	Not less than 1.15% (weight/volume)	Not less than 1.05% (weight/volume)	Not less than 0.95% (weight/volume)
Soluble solids excluding salt	Not less than 14% (weight/volume)	Not less than 12% (weight/volume)	—
Ingredients	Same as specified in ingredients in the preceding Article.		
Additives	Same as specified in additives in the preceding Article.		
Net contents	Same as specified in the net contents in the preceding Article.		

(The Standard for *Tamari Shoyu*)

Article 5: The criteria of *Tamari Shoyu* are as follows:

Classification	Criteria		
	Special grade	Superior grade	Normal grade
Manufacturing method	Same as specified in the manufacturing method in Article 3.	—	—
Property	Having a dark reddish-brown color, special flavor and matured taste with mellowed saltiness and umami, which are peculiar to the fermented <i>Tamari Shoyu</i> that enough fermentation and aging are performed, its all aspects are excellent, and being free from objectionable taste and odor and mold.	Having a dark reddish-brown color, special flavor and matured taste with mellowed saltiness and umami, which are peculiar to the fermented <i>Tamari Shoyu</i> that enough fermentation and aging are performed, its all aspects are good, and being free from objectionable taste and odor and mold.	Having a dark reddish-brown color, special flavor and matured taste with mellowed saltiness and umami, which are peculiar to the fermented <i>Tamari Shoyu</i> , its all aspects are good, and being free from objectionable taste and odor and mold.
Degree of color	Less than No.18 of the standard color for soy sauce		
Total nitrogen	Not less than 1.60% (weight/volume)	Not less than 1.40% (weight/volume)	Not less than 1.20% (weight/volume)
Soluble solids excluding salt	Not less than 16% (weight/volume)	Not less than 13% (weight/volume)	—
Ingredients	Same as specified in ingredients in Article 3.		
Additives	Same as specified in additives in Article 3.		
Net contents	Same as specified in the net contents in Article 3.		

(The Standard for *Saishikomi Shoyu*)

Article 6: The criteria of *Saishikomi Shoyu* are as follows:

Classification	Criteria		
	Special grade	Superior grade	Normal grade
Manufacturing method	Shall be made by the regular fermenting method or mixing-fermenting method.	—	—
Property	Having a dark reddish-brown color, special flavor and matured taste with mellowed saltiness and umami, which are peculiar to the fermented <i>Saishikomi Shoyu</i> that enough fermentation and aging are performed, its all aspects are excellent, and being free from objectionable taste and odor and mold.	Having a dark reddish-brown color, special flavor and matured taste with mellowed saltiness and umami, which are peculiar to the fermented <i>Saishikomi Shoyu</i> that enough fermentation and aging are performed, its all aspects are good, and being free from objectionable taste and odor and mold.	Having a dark reddish-brown color, special flavor and matured taste with mellowed saltiness and umami, which are peculiar to the fermented <i>Saishikomi Shoyu</i> , its all aspects are good, and being free from objectionable taste and odor and mold.
Degree of color	Less than No.18 of the standard color for soy sauce		
Total nitrogen	Not less than 1.65% (weight/volume); however, for those made by the mixing-fermenting method, not less than 2.00% (weight/volume)	Not less than 1.50% (weight/volume)	Not less than 1.40% (weight/volume)
A use ratio of hydrolyzed vegetable protein, etc. (Only as for mixing-fermenting method)	Not more than 20%	—	—
Soluble solids excluding salt	Not less than 21% (weight/volume)	Not less than 18% (weight/volume)	—
Ingredients	Same as specified in ingredients in Article 3.		
Additives	Same as specified in additives in Article 3.		
Net contents	Same as specified in the net contents in Article 3.		

(The Standard for *Shiro Shoyu*)

Article 7 The criteria of *Shiro Shoyu* are as follows:

Classification	Criteria		
	Special grade	Superior grade	Normal grade
Manufacturing method	Same as specified in the manufacturing method in Article 3.	—	—
Property	Having a light amber color, special flavor and matured taste with mellowed saltiness and umami, which are peculiar to the fermented <i>Shiro Shoyu</i> that enough fermentation and aging are performed, its all aspects are excellent, and being free from objectionable taste and odor and mold.	Having a light amber color, special flavor and matured taste with mellowed saltiness and umami, which are peculiar to the fermented <i>Shiro Shoyu</i> that enough fermentation and aging are performed, its all aspects are good, and being free from objectionable taste and odor and mold.	Having a light amber color, special flavor and matured taste with mellowed saltiness and umami, which are peculiar to the fermented <i>Shiro Shoyu</i> , its all aspects are good, and being free from objectionable taste and odor and mold.
Degree of color	Not less than No.46 of the standard color for soy sauce (by the color indicating method of JIS Z 8781-4, where : L=76.7, a=12.5, b=81.9)		
Total nitrogen	Not less than 0.40% (weight/volume) Less than 0.80% (weight/volume)	Not less than 0.40% (weight/volume) Less than 0.90% (weight/volume)	Same as left
Soluble solids excluding salt	Not less than 16% (weight/volume) in the condition that exclude added sugars	Not less than 13% (weight/volume)	Not less than 10% (weight/volume)
Directing reducing sugar	Not less than 12% (weight/volume)	Not less than 9% (weight/volume)	Not less than 6% (weight/volume)
Ingredients	Not using a thing except following mentioned ingredients: 1. soybean; 2. wheat, barley and naked barley; 3. wheat gluten; 4. salt; 5. hydrolyzed vegetable protein, enzymatic hydrolyzed vegetable protein and fermentative hydrolyzed vegetable protein; 6. sugars; 7. alcohol, <i>Shochu</i> (rice wine spirit) and <i>Sake</i> (rice wine); 8. rice fermentative seasoning, vinegar, <i>Mirin</i> and <i>Mirin</i> type seasoning.		
Additives	Same as specified in additives in Article 3.		
Net contents	Same as specified in the net contents in Article 3.		

(Method of measurement)

Article 8 The method of measurement for degree color, total nitrogen and soluble solids excluding salt in the standards from Article 3 through the preceding article, and for the direct reducing sugar in the standard of the preceding article shall be as follows;

Items	Method of measurement
Degree of color	Pour the sample into a test tube of 10 mm in diameter and compare the color with the standard color for soy sauce.
Total nitrogen	<p>Total nitrogen is measured by Kjeldahl method or combustion method.</p> <ol style="list-style-type: none"> 1. Kjeldahl method <ol style="list-style-type: none"> (1) Procedure <ol style="list-style-type: none"> (a) Digestion of sample <ol style="list-style-type: none"> (i) In case of using power variable heating device (capable of heating Kjeldahl flask , and boiling 50 mL of water in Kjeldahl flask with 3 - 5 boiling stones within 5 minutes when the flask is placed on the heat source of the apparatus which has been keeping maximum output for 10 minutes; the same applies hereinafter) <p>Take 1 mL of sample into 50 - 300 mL Kjeldahl flask, add 5 g of Kjeldahl catalysts (mixture of potassium sulfate and $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ in the ratio of 9 to 1 ; the same applies hereinafter) and approximately 8 mL of sulfuric acid. Heat gently with power variable heating device until bubbling becomes calm. Keep boiling for approximately 90 minutes more after the digestion solution becomes transparent. Boil for not less than 2 hours totally. After digestion stops, cool to room temperature, add 20 - 50 mL of water depending on distillation apparatus, dissolve the sample with low heating as needed. For the blank test, operate without a sample in the same procedure</p> (ii) In case of using Block Digester (capable of boiling 50 mL of water in digestion tube at 420°C within 2 minutes 30 seconds; the same applies hereinafter) <p>Take 1 mL of sample into 250 - 300 mL digestion tube, add 5 g of Kjeldahl catalysts and approximately 8 mL of sulfuric acid. Heat gently with Block Digester set beforehand at 200°C until bubbling become calm, and then set at 420°C. Keep boiling for approximately 90 minutes more after the digestion solution becomes transparent. Boil for not less than 2 hours totally. After digestion stops, cool to room temperature, add 20 - 50 mL of water depending on distillation apparatus, dissolve the sample with low heating as needed. Operate without a sample in the same procedure for the blank test.</p> (b) Distillation <ol style="list-style-type: none"> (i) In case of using Shioiri / Okuda style steam distillation apparatus <p>Pour 25 - 30 mL of boric acid solution (prepared by dissolving 10 - 40 g of boric acid in 1,000 mL of water with heating; the same applies hereinafter) into receiver for distillate (hereinafter referred to as “the receiver”), add a few drops of methyl red / bromocresol green indicator (prepared by dissolving 0.15 g of bromocresol green and 0.10 g of methyl red in 200 mL of 95% ethanol with heating; the same applies hereinafter), and place it so that distillate outflow port is immersed in the solution of the receiver. Connect the Kjeldahl flask containing digestion solution to distillation apparatus, add appropriate volume of 25 – 45% sodium hydrate solution to contain 16 g or more of sodium hydrate in order to form an alkaline solution and distill until 100 mL or more of distillate is obtained. Remove the outflow port from the solution and wash the port end with small amount of water.</p> (ii) In case of distilling all digestion solution with Parnas / Wagner type steam distillation apparatus <p>Pour 25 - 30 mL of boric acid solution into receiver, add a few drops of methyl red / bromocresol green indicator and place it so that distillate outflow port is immersed in the solution of the receiver. Wash the digestion solution into the distillation tube with water 3 times. The volume of water for wash is 30 mL or more depending on the capacity of the distillation tube.</p>

Add appropriate volume of 25 – 45% sodium hydrate solution to contain 16 g or more of sodium hydrate in order to form an alkaline solution and distill until 100 mL or more of distillate is obtained. Remove the outflow port from the solution and wash tip of the port with small amount of water.

- (iii) In case of distilling a part of digestion solution with Parnas / Wagner type steam distillation apparatus

Wash the digestion solution into a volumetric flask with water and add water until the whole volume becomes 100 mL. Use this as the sample solution.

Pour 25 - 30 mL of boric acid solution into the receiver, add a few drops of methyl red / bromocresol green indicator and place it so that distillate outflow port is immersed in the solution of the receiver. Pour 25 mL of the sample solution into distillation tube with a volumetric pipette, add appropriate volume of 25 – 45% sodium hydrate solution to contain 4 g or more of sodium hydrate in order to form an alkaline solution and distill until 100 mL or more of distillate is obtained. Remove the outflow port from the solution and wash tip of the port with small amount of water.

- (iv) In case of using automatic distillation apparatus (conducting rapidly automatically steam distillation of Kjeldahl method and including apparatus combined automatic distillation device and automatic titration device (automatically judging the end point of titration and having a burette capacity of 20 mL or more; the same applies hereinafter) ; the same applies hereinafter)

Pour 25 - 30 mL of boric acid solution into the receiver, add a few drops of methyl red / bromocresol green indicator and place it so that distillate outflow port is immersed in the solution of the receiver. Add 30 mL of water and appropriate volume of 25 - 45% sodium hydrate solution to contain 16 g or more of sodium hydrate in order to form an alkaline solution and distill until 100 mL or more of distillate is obtained according to the operation procedure of automatic distillation apparatus. Remove the outflow port from the solution and wash the port end with small amount of water. In case of using apparatus combined automatic distillation device and automatic titration device, distill and titrate in the procedure suitable for apparatus.

(c) Titration

- (i) Manual titration (method by judging the end point of titration by looking at discoloration of indicator)

In case of distilling a part of digestion solution with Parnas / Wagner type steam distillation apparatus, titrate with 0.025 mol / L of sulfuric acid using 25 mL burette. In case of distilling all digestion solution with Parnas / Wagner type steam distillation apparatus, titrate with 0.05 mol / L of sulfuric acid using 25 mL burette. The end point is where the solution turns pale reddish gray through green and dirty colorless. Report as the titration value to the nearest 0.01 mL. For the blank test, titrate in the same procedure.

- (ii) Automatic titration (method by judging the end point of titration automatically)

Titrate according to the operation procedure of automatic titrator. For the blank test, titrate in the same procedure.

(2) Calculations

- (a) In case of distillation and titration for all digestion solution

$$\text{Total nitrogen (w / v \%)} = (T - B) \times F \times M \times A \times 2 / (1\,000 \times V) \times 100$$

- (b) In case of distillation and titration for a part of digestion solution

$$\text{Total nitrogen (w / v \%)} = (T - B) \times F \times M \times A \times 2 / (1\,000 \times V) \times (100 / 25) \times 100$$

where

T is the volume of H₂SO₄ titrant (mL)

B is the volume of H₂SO₄ titrant for blank (mL)

F is the factor of 0.05 mol/L H₂SO₄

M is Atomic weight of nitrogen, 14.007

A is concentration of H₂SO₄ titrant (mol / L)

V is sampling volume (mL)

Note 1: The grade of water used for the test shall be A2 specified in Japanese Industrial Standards K 0557 (1998) (hereinafter referred to as “JIS K 0557”), equivalent grade or higher quality.

Note 2: The grade of volumeter used for sampling shall be air displacement type or positive-displacement type with piston and its random error shall be within $\pm 1.0\%$ and its systematic error shall be 0.3% or less calculated according to Japanese Industrial Standards K 0970 (2013) (hereinafter referred to as “JIS K 0970”) when set volume is 1 mL. If such volumeter isn't available, calculate sampling volume (mL) by measuring the density of sample according to Japanese Industrial Standards K 0061 (2001) (hereinafter referred to as “JIS K 0061”) and weighing accurately, to the nearest 0.1 mg, approximately 1.0 - 1.3 g of sample.

In case of using air displacement volumeter for sampling, use the reverse-pipetting (method of sampling set volume when using pipette discharging liquid in 2 steps by aspirating sample solution with pushing the push button to the second stop, discharging by pressing the button to the first stop and leaving the residue of the sample solution in the tip)

Note 3: The grade of the reagent used for the test shall be special grade or appropriate grade specified in JIS.

Note 4: Block Digestor shall have exhaust manifold etc. for emitting sulfuric acid mist. In case of ensuring safe analysis condition, the digestion can start from 420°C by adding 1 mL of 30% hydrogen peroxide after pouring sulfuric acid. In case of being in danger of leaking out digestion solution by bubbling etc., stop heating, cool until bubbling blows over and then restart heating.

Note 5: The grade of the glass volumeter used for the test shall be class A specified in Japanese Industrial Standards R 3505 (1994) (hereinafter referred to as “JIS R 3505”), equivalent grade or higher grade.

Note 6: When one drop clearly shows a color exceeding the end point in the blank test, the titration value shall be 0 mL.

2 Combustion method

(1) Apparatus for measuring total nitrogen shall be with capability as follows:

- (a) A furnace able to maintain a given temperature greater than or equal to 870 °C in oxygen (with a purity of 99.9% or higher), to pyrolyze the sample. When using dedicated soy sauce apparatus, a furnace able to maintain a given temperature greater than or equal to 680 °C in oxygen-helium mixed gas.
- (b) A structure capable of separating free nitrogen (N₂) from other combustion products, to measure of nitrogen (N₂) by a thermal conductivity detector .
- (c) A mechanism for converting nitrogen oxides (NO_x) to nitrogen (N₂).
- (d) Having the average value of the nitrogen content within the theoretical value $\pm 0.15\%$ and having the relative standard deviation within 1.3% (in case of using lysine hydrochloride (with a purity of 99% or higher)), or within 0.98% (in case of using nicotinic acid (with a purity of 99% or higher)).
- (e) Taking measures so that samples with a salt concentration of approximately 20% can be measured.

(2) Procedure

- (a) According to the operation procedure for each apparatus, weigh accurately, to the nearest 0.1mg or less, a standard product for preparing calibration curves (ethylenediaminetetraacetic acid (EDTA) (with a purity of 99% or higher and described a nitrogen ratio), DL-aspartic acid (with a purity of 99% or higher and described a nitrogen ratio) or other standard product of the same purity (excluding those used in paragraph 2(1)(d))) . Then measure in the procedure suitable for each apparatus and prepare a calibration curve.
- (b) According to the operation procedure for each apparatus, when taking 100 mg or more of the sample, weigh accurately to the nearest 0.1 mg. When taking 40 mg or more but less than 100 mg of the sample, weigh accurately to the nearest 0.01mg. Then measure in the procedure suitable for each apparatus.
- (c) Measure the density of the sample according to JIS K 0061.

(3) Calculation

Calculate the total nitrogen content of the sample (w/w %) by the calibration curve and obtain the total nitrogen content (w/v %) by using the following equation.

	<p>Total nitrogen content (w/v %) = Total nitrogen content of the sample (w/w %) × Density (g/mL)</p> <p>Note: The sampling method can be replaced with a method using a volumeter that can accurately measure the set capacity (with the piston, and air-displacement type or with the air layer capacity being 25% or less of the set capacity or positive displacement type, the systematic error is within ± 1.0% and the random error is 0.3% or less in the set capacity according to JIS K 0970). In this case, the total nitrogen content (w/v %) is obtained by dividing the amount of nitrogen obtained from the calibration curve by the set capacity.</p>
Soluble solids excluding salt	<ol style="list-style-type: none"> 1. Measurement of soluble solids Read the degree of the sample with a refractometer for sugar keeping sample and refractometer at 20 ° C, and indicate value by %. 2. Measurement of salt content It is measured by the potentiometric titration method or the Mole method. <ol style="list-style-type: none"> (1) Preparation of sample solution Pour 5 mL of sample into a volumetric flask using a volume pipette and add water until the whole volume becomes 250mL. Use this as the sample solution. (2) Measurement <ol style="list-style-type: none"> (a) Potentiometric titration method Take 10 mL of the sample solution into a 100-200 mL beaker with a volume pipette. Add water to the level where the electrode is immersed. Add 1 mL of nitric acid (1 + 1) (equal volume of nitric acid and water) and 1 mL of Tween 20 solution (1 to 2 g of Tween 20 and 100 mL of water with a measuring cylinder; the same applies hereinafter). Mount the beaker to a potentiometric titration apparatus. While stirring, titrate with 0.05 mol / L or 0.1 mol / L silver nitrate solution, and determine the end point according to the operation of the titration apparatus. For the blank test, titrate in the same procedure using water in place of the sample solution. In this case, when no end point is detected or the volume of the silver nitrate solution required for titration is less than 0.01 mL, the titration value shall be 0 mL. (b) Mole method Take 5 mL of sample solution into a porcelain evaporating dish or Erlenmeyer flask using a volume pipette, add 1 mL of 2% potassium chromate solution as an indicator and titrate with 0.02 mol / L silver nitrate solution using an amberburette. When the color of the liquid turns slightly orange, define it as the end point. For the blank test, titrate in the same procedure using 5 mL of water instead of the sample solution. In this case, when one drop clearly shows a color exceeding the end point, the titration value shall be 0 mL. (3) Calculation <ol style="list-style-type: none"> (a) Potentiometric titration method Salt content (%) = $[(T - B) / 1000] \times A \times F \times M \times (250 / 10) \times (1 / V) \times 100$ (b) Mole method Salt content (%) = $[(T - B) / 1000] \times A \times F \times M \times (250 / 5) \times (1 / V) \times 100 \times C$ where T is the volume of the silver nitrate solution required for titration of the sample solution (mL) B is the volume of the silver nitrate solution required for titration in the blank test (mL) A is the concentration of the silver nitrate solution used for the titration (mol / L) F is factor of silver nitrate solution M is 58.44 (Formula weight of sodium chloride) V is sampling volume (mL) C is correction factor (<i>Shiro Shoyu</i> : 1.00, <i>Usukuchi Shoyu</i> : 0.99, <i>Koikuchi Shoyu</i> : 0.98, <i>Tamari Shoyu</i> and <i>Saishikomi Shoyu</i> : 0.97)

	<p>Note1: The grade of water used for the test shall be A2 specified in JIS K 0557, equivalent grade or higher quality.</p> <p>Note2: The grade of reagents used for the test shall be special grade or appropriate grades specified in the Japanese Industrial Standards.</p> <p>Note3: The grade of the glass volumeter used for the test shall be class A specified in JIS R 3505, equivalent grade or higher grade.</p> <p>Note4: The sample solution can be replaced with the sample using a device capable of measuring a constant volume and diluted with water. In this case, eliminate "$\times (250/10)$" or "$\times (250/5)$" in the calculation equation in (3).</p> <p>Note5: The potentiometric titrator shall have a burette capacity of 20 mL or more. In this case, for the electrode, an indicator electrode (silver electrode etc.) and a reference electrode suitable for chloride measurement, or a composite electrode of them shall be used.</p> <p>Note6: In the case where it is possible to accurately determine the end point of the potentiometric titrator, the concentration of the silver nitrate solution can be changed.</p> <p>Note7: Instead of the Tween 20 solution, a solution containing an anionic surfactant suitable for a potentiometric titrator can be used.</p> <p>Note8: The amber burette shall have a burette capacity of 25 mL or more.</p> <p>3. Calculation of soluble solid excluding salt Soluble solids excluding salt (%) = Soluble solids (%) - Salt content (%)</p>
Direct reducing sugar	<p>Dilute the sample so that the whole volume increases by 20 to 25 times. Take 5 to 20 mL of the diluted solution, add 20 mL of Fehling's solution and water so that the whole volume become 59 mL. Boil accurately 2 minutes and rapidly cool, add 10 mL of 25 % sulfuric acid and 3 g of potassium iodide. Titrate with 0.1 mol/L sodium thiosulfate solution using 1% starch solution as an indicator. Separately carry out the blank test without adding the sample. The difference between the values of the both titrations is determined as the direct reducing sugar.</p>