Japan Customs Analysis Methods

No. 403

Quantitative Analysis Methods for Starch Derivatives (Issued in June 1999) (Updated in June 2003)

1. Scope

This analysis method is applied for identifying starch derivatives (for example, esterified starch, cationic starches, etc.) covered by the term of subheading 3505.10 of Customs Tariff low (Appendix Table - Customs Tariff Schedule).

2. Outline of Test Method

This method determines "esterified starches and other starch derivatives", "other modified starches" and "unmodified starches", according to the following procedures:

(1) General tests
(2) Sample purification
(3) Analyses specific for the kinds of starch derivatives

3. Apparatus

(1) For analysis of alkyl etherified starches

A gas chromatograph (GC), equipped with a flame ionization detector.

(2) For Brabender viscography

A Brabender viscograph

4. Reagents

All chemicals must be JIS special reagent grade or equivalent, unless otherwise specified. All solutions for titration can be a commercial standard solution for volumetric analysis as long as it is JA.5 solution for titration of JIS K 8001 or standardized on the Japanese Pharmacopoeia for its strength (factor).

(a) 0.1 mol/L iodine solution

Dissolve 1.3 g of iodine and 4 g of potassium iodide in water and add a drop of dilute hydrochloric acid. Dilute to 100 mL volume with water (stored in the dark).

(b) Fehling's solution

Refer to the Japan Customs Analysis Method No. 102 "Analysis Methods for Honey."

(c) Glucoamylase solution

Dissolve glucoamylase (1, 4 - α - Glucan glucohydrolase EC 3.2.1.3) in 0.2 M acetic acid buffer solution (pH 4.8) so that the potency equals to 7 unit/ml.(1)

(d) Reagent for the Hanes method

Refer to the Japan Customs Analysis Method No. 108 "Quantitative Analysis of Sucrose in Confectionary".

(e) 1% Eosin Y solution

Dissolve 1 g of Eosin Y in water and dilute to 100 mL volume with water.

(f) 0.1% Eosin Y solution

Dilute 10 mL of 1% Eosin Y solution to 100 mL volume with water.

(g) 0.1 mol/L sodium hydroxide solution

Dissolve 4.5 g of sodium hydroxide in water and dilute to 1000 mL volume with water.

(h) 0.45 mol/L sodium hydroxide solution

Dissolve 20 g of sodium hydroxide in water and dilute to 1000 mL volume with water.

(i) 2 mol/L sodium hydroxide solution

Dissolve 8 g of sodium hydroxide in water and dilute to 100 ml volume with water.

(j) 0.2 mol/L hydrochloric acid

Add water to 18 mL of hydrochloric acid to make 1,000 mL volume in total.

(k) 0.1 mol/L acetic acid

Weigh 6.0 g of acetic acid and add water to make it to 1,000 mL volume in total.

(l) F-kit (test kit for the determination of acetic acid)

Use an existing test kit for the determination of acetic acid in foodstuffs and other material, produced by Roche Diagnostics Co.

(m) Amidol solution

Dissolve 0.5 g of amidol and 10 g of $NaHSO_4$ in water and dilute to 50 mL volume with water.

(n) Ammonium molybdate solution

Dissolve 8.3 g of ammonium molybdate in water and dilute to 100 mL volume with water.

(o) Phosphorus standard stock solution

Dissolve 4.339 g of KH_2PO_4 (containing 1.0 g of phosphorus) in water and dilute to 1000 mL volume with water.

(p) Phosphorus standard solution

Dilute 10 mL of the phosphorus standard stock solution with water to 1,000 mL in total.

Note 1) 1 unit means the amount of enzyme which produces 10 mg of glucose when incubated at pH4.5, 40°C for 30 min with soluble starch as the substrate.

5. General Analysis

5.1. Iodine Color Reaction

Take 0.5 g of sample in a test tube, disperse with 5 mL of water and observe the dissolving behavior; in water, carboxymethylated starch (CMS) dissolves and phosphorylated starch swells or dissolves. Then, add a few drops of the iodine solution and observe the color reaction after mixing well.

Results of the iodine color reaction regarding various types of starches and their derivatives are exemplified below (the colors in parentheses will appear when 5 to 10 min is elapsed after the addition of the iodine solution).

Table Iodine color reactions of starches (and their derivatives)

Tapioca	Purplish blue	
Wheat	Purple	[Brown]
Soluble starch	Purplish blue	
Dextrin	Purplish red	
Pregelatinized-starch	Blue	
Corn	Purple	
Corn (High amylose type)	Purplish red	
Corn (Waxy corn)	Brown	
Corn, cross-linked with phosphate	Purple	
Corn, phosphomonoesterified	Purplish blue	[Purple]
Corn, acetate	Purple	[Brown]
Corn, hydroxypropyl-etherified	Purple	[Brown]
Potato	Purplish blue	[Blue]
	(more blue-colored than that of tapioca starch)	
Potato, cationic	Purplish blue	[Blue]
Potato, acetate	Purplish blue	[Blue]
Potato, hydroxypropyl-etherified	Purplish blue	[Blue]
Potato, cyanoethylated	Purplish blue	[Blue]

5.2. Reducing Ability against Fehling's Solution

Weigh 1 g of sample in a test tube and disperse in 5 mL water. Add 2 to 3 mL of the Fehling's solution, mix thoroughly and then heat the tube gradually using a gas burner. When the sample extremely swells and forms a gel, making even heating difficult, stir the liquid with a glass stick and heat with keen attention to prevent explosive boil.

After heating for about 3 min, confirm the production of reduced copper (reddish cuprous oxide precipitates).

5.3. Moisture Content

Depending on the characteristics of sample and the presence of any additives, determine the moisture content in the sample appropriately. For example, the following methods can be applied:

(1) Accurately weigh 2 to 3 g of the sample and dry it at 105 to 110°C to a constant weight.

(2) Accurately weigh 2 to 3 g of the sample and dry it at 135°C for an hour.

(3) Accurately weigh 2 to 3 g of the sample and dry it at 100°C under low pressure to a constant weight.

5.4. Ash Content

Accurately weigh approximately 5 g of sample in a porcelain crucible which has previously been dried to a constant weight. After carbonizing the sample using a gas burner, incinerate it in an electric furnace at 550 to 600 °C.

If incineration does not occur completely due to residual carbon (the sample remains black), cool the crucible and add a few drops of water or nitric acid (1 + 1) to wet the ash, then heat it again.

5.5. Microscopic Observation of Starch Particles

Take a small amount of sample on a glass slide, followed by offering one drop of water, and place a cover glass over the wet sample. Set the glass slide on an optical microscope and observe starch particles. Standard starches should also be observed as reference.

In the observation, it is required to confirm whether starch particles have polarization crosses when observed under polarized light. The polarization is an important indicator to know to what extent the starch sample has been processed.

5.6. Infrared Absorption Spectroscopy

Measure the infrared absorption (IR) spectrum of sample using the KBr tablet method, etc. and confirm the presence of starch and any additive(s) by comparing the obtained spectrum with the standard ones.

Esterified starches may show an absorption peak near 1730 cm^{-1}.

5.7. Extraction with Organic Solvents

Perform extractions from a starch sample with ethyl ether, ethanol and 80% methanol in order. Quantify the respective extracts and measure their IR spectra for qualitative analysis purpose.

Extracts and their quantities might provide important information for identifying what kind of starch derivatives the sample is.

5.8. Brabender Viscography

Use a Brabender viscograph to measure a viscogram of sample. For the measurement, pour a dried sample together with 450 mL of water into a measuring vessel. Set the measuring vessel to the apparatus and wait 5 min after the liquid temperature reaches a constant temperature of 30°C. Raise the temperature at the rate of 1.5°C/min until at 92.0°C (hold 15 minutes). Compare the obtained viscogram with that of raw material starch (or unmodified starch of the same kind).

As for the measuring conditions such as the rotating speed and the sample quantity, refer to 9. "Brabender viscograms".

5.9. Decomposition Behavior under Enzymatic Hydrolysis

Weigh 0.3 g of a purified sample (purification methods are described later) in an Erlenmeyer flask. Add 50 mL of water to the flask and heat in a boiling water bath for 30 min for gelatinization of the sample, and dilute to 100 mL volume. Transfer 30 mL of the solution to an Erlenmeyer flask with the addition of 1 mL of the gulcoamylase solution. Keep it in a thermostat bath at 37°C for 2.5 hours for enzymatic reaction. Using 1 mL of the reaction solution, determine the amount of reducing sugar produced by glucoamylase using the Hanes method. Conduct the same procedure

for purified raw material starch (or unmodified starch of the same kind).

The consumption of 0.01 mol/L sodium thiosulfate solution for a starch derivative may be smaller than that of its raw material starch (or unmodified starch of the same kind).

[Example] Phosphorylated starch

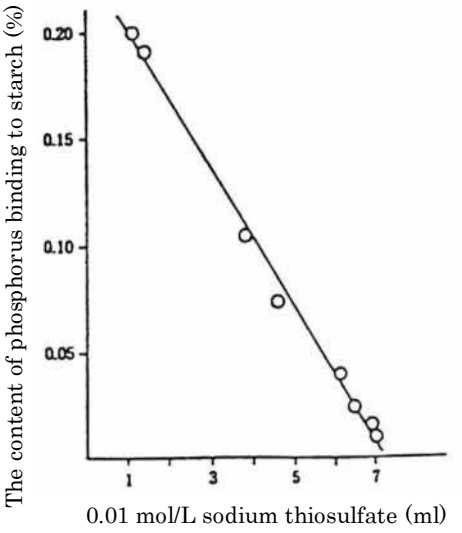


Fig. Relationship between the content of phosphorus binding to starch and the consumed volume of 0.01 mol/L sodium thiosulfate solution; Correlation Coefficient: r = 0.997

6. Sample Purification

6.1. Using semipermeable membrane

Put a sample, processed by the extractions of 5.7., in a semipermeable membrane tube (to be cut to a length suitable for the sample quantity), and dialyze under running water for 3 days. After the dialysis, take out the inside material on a filter paper. After water is absorbed and removed to some extent, dry the material, together with the filter paper, at room temperature for 24 hours. (Determine the moisture content in the purified and dried sample.)

6.2. Using a Large Amount of Water

Take a sample into a glass-stoppered Erlenmeyer flask, and add water more than 50 times as much as the volume of the sample. Wash the sample for 60 min by shaking the flask from time to time. Using a Büchner funnel, filter the sample liquid under vacuum. If necessary to dry up quickly, add a large amount of methanol into the funnel in low vacuum and stir the liquid gently. Then, aspirate the funnel again (in high vacuum). Repeat this step twice to decrease the water content in the washed sample. Take out the washed sample onto a filter paper. After water is absorbed and removed to some extent, dry the washed sample, together with the filter paper at room temperature for 24 hours. (Determine the moisture content in the washed sample after dried.)

Since smaller particle types of starch, such as wheat starch, become more prone to clogging of a filter paper, it may be difficult to conduct filtration. In this case, separate starch particles, for example, by centrifugation.

7. Analysis Methods specific for Modified Starch-types

7.1. Cross-linked Starch [Sedimentation test]

Prepare a solution by mixing 10 g of zinc chloride, 26 g of ammonium chloride and 64 mL of water. Transfer 15 mL of the solution into a test tube with a 30 mm-inner diameter. Add 150 mg of purified sample (on a dry basis) to the test tube, heat for 10 min in a boiling water bath and cool under running water immediately. Transfer 10 mL of the liquid in the test tube into a 10 mL measuring cylinder and allow it to stand for 12 hours. If the liquid is divided into two layers (a clear supernatant and a semitransparent layer), it is considered that the sample has characteristics on sedimentation. (Read, as the sedimentation volume, the volume of the semitransparent layer at the time.)

Conduct the same procedures for purified raw material starch (or untreated starch of the same kind) as a reference.

Although cross-linked starch generally shows characteristics on sedimentation, it should be noted that potato starch, even if cross-linked, may not show any characteristics of sedimentation due to its particles floating in the solution.

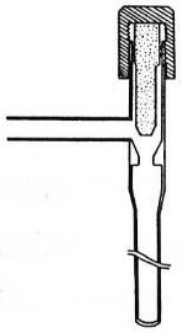
7.2. Alkyl Etherified Starch [decomposition with hydriodic acid]

Transfer approximately 100 mg of purified sample into a decomposition tube like the example below. Add 2 mL of hydriodic acid to the tube cooling with a freezing

mixture (acetone-ice type). Close the tube stopper (plug) tightly after reducing the inner pressure with a vacuum pump, and dissolve the test material by shaking. Heat the decomposition tube in an oil bath or the like at a temperature of 140°C for 20 min.

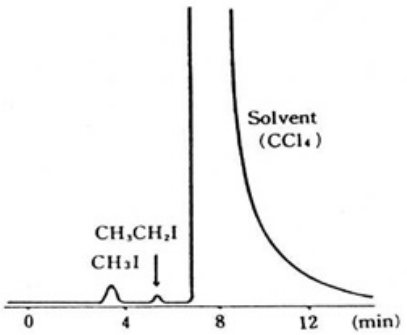
Cool the decomposition tube at room temperature. Then, put 1.5 mL of carbon tetrachloride into the suction port of the composition tube (for connecting with vacuum pump tube) while cooling with a freezing mixture, and loosen the plug so that the carbon tetrachloride is loaded into the composition tube. Close the plug tightly and shake the tube to ensure absorbing decomposition products in carbon tetrachloride. Inject the carbon tetrachloride solution into a gas chromatograph.

Decomposition tube

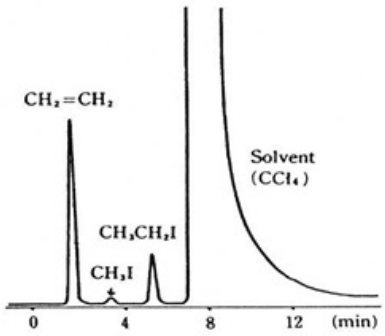


[Examples of Gas chromatography conditions and Results]

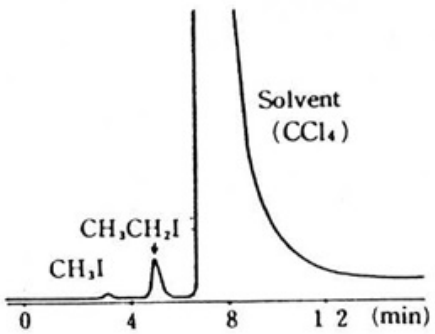
Separation column	(1) - (4)	: 5% silicone OV-101 / Chromosolve DMC (80 - 100 meshes) packed in a glass column (3 mm I.D. × 4 m)
	(5) - (6)	: DB-WAX 30 m × 0.25 mm I.D. × 0.25 µm film thickness
Oven Temp.		: 50°C (constant)
Injection Port Temp.		: 200°C
Detection Temp.		: 200°C (FID)
Carrier gas/flow rate	(1) - (4)	: He, 20 mL/min (constant flow)
	(5) - (6)	: He,1 mL/min (constant flow)



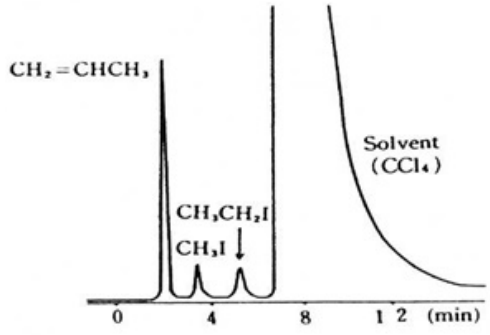
(1) Unmodified starch



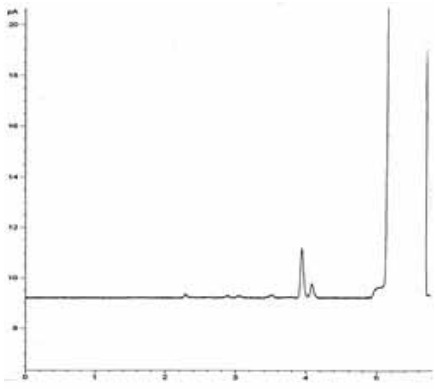
(2) Hydroxyethylated starch



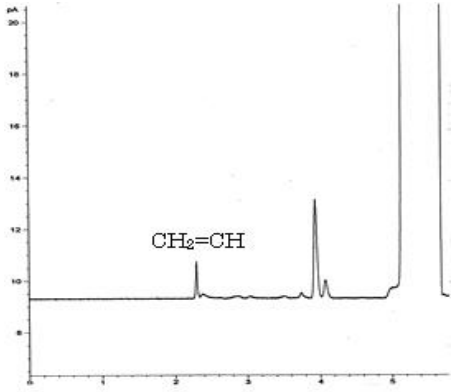
(3) Carboxymethylated starch



(4) Hydroxypropylated starch



(5) Unmodified starch



(6) Hydroxypropylated starch

In case of hydroxyprpylated starch, the following method can be applied.

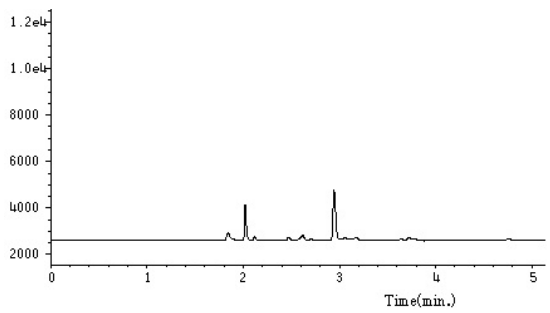
Weigh approximately 60 mg of sample in a decomposition bottle.(2) Add approximately 80 mg of adipic acid and 2.0 mL of hydriodic acid in order, close the bottle plug tightly, and seal with tape. Shake the bottle for 30 seconds. Then heat it using a heating device(3) at 150°C for 60 min, shaking the bottle every 5 min. After the heating, cool the bottle under running water to room temperature. Inject the upper layer of the solution into a gas chromatograph.

Note 2) a 5 mL volume of heat and pressure resistant glass bottle with screw cap. The inner side of the bottom should be a conical shape. The outer diameter is 20 mm, the height to the neck is 50 mm and the volume from the bottom to 30 mm height is 2 ml. The cap should be made of a heat resistant resin and the inner cap and seal should be made of a fluorocarbon resin.

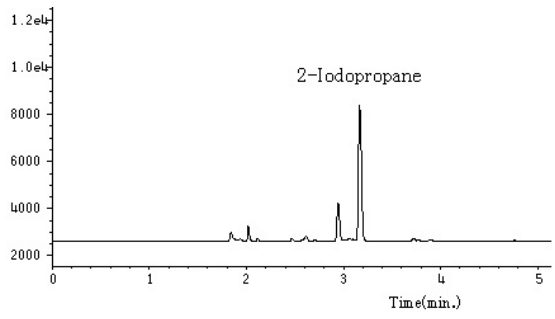
Note 3) A heating device with a square-shaped aluminum block, 60 - 80 mm thickness, having holes (32 mm depth) in diameter of 20.6 mm, which can control the inside temperature of the heat block within ±1°C. When a heater with stirring function is used, put a stir bar into the bottle after the addition of adipic acid..

[Examples of Gas chromatography conditions and Results]

Separation Column : HP-5, 30 m × 0.32 mm I.D. × 0.25 µm film thickness
Oven Temp. : 40°C (8 min) to 320°C (5 min), ramp rate of 30°C/min
Injection Port Temp. : 320°C
Detector Temp. : 320°C (FID)
Carrier gas/flow rate : He, 1 mL/min (constant flow)



Unmodified starch



Hydroxypropylated starch

7.3. Cationic Starch

(1) Eosin Y (anionic dye) Adsorption Test

Take approximately 0.5 mg of purified sample in a test tube, add 2 - 3 mL of 1% Eosin Y solution and disperse thoroughly. Then, drop a small amount of the dispersion liquid on a filter paper and observe the absorption behavior. In case of cationic starch, the dye is adsorbed (by the starch particles).

Alternatively, take approximately 0.5 mg of purified sample into a test tube, add 2 - 3 mL of 1% Eosin Y solution and disperse thoroughly. Leave the test material to sediment, and observe the coloration in the sediment (starch) layer. The observation of the coloration will become easier by previously washing the dyed starch particles with water.

Carry out the same procedure for purified raw material starch (or unmodified starch of the same kind) as a reference.

(2) Measurement of Conductivity

Cationic starch can also be identified through conductivity measurement.

Take 1 g of purified sample in a beaker, add 100 mL of water,[4] and disperse. Heat the dispersed liquid in a boiling water bath for gelatinization. After cooling to room temperature, measure the conductivity of the gelatinized liquid.

Carry out the same procedure for purified raw material starch (or unmodified starch of the same kind) as a reference.

Note 4) The conductivity of water to be used should be around 1.6 μS/cm.

Table Conductivities of 1% gelatinized starch solutions

Starch		Conductivity (μS/cm, at 25°C)
Unmodified starch	(potato)	6.5
	(wheat)	5.0
	(corn)	6.7
Cationic starch		34.0
		20.3
		27.5
		60.7
Cyanoethylated starch		70.7
Carbamic acid phosphate starch		237
Acetylated starch		7.3
Hydroxyethylated starch		5.2
Hydroxypropylated starch		14.9
		10.8
Carboxymethylated starch		1888
Phosphorylated starch		295
Soluble starch		65.2

7.4. Acetylated Starch

(1) Measurement of IR spectra

Measure an IR spectrum of purified sample to examine whether acetylated. Acetylated starch generally shows an absorption peak near 1730 cm^{-1} (See 8. "Infrared absorption spectra of acetylated starches"). However, it is important to note that the absorption peak may hardly be recognized, even it is acetylated.

In addition, IR spectra of methanol extracts or water extracts from unpurified samples generally show absorptions derived from sodium acetate.

(2) Determination of the degree of acetylation (Titration method)

Accurately weigh approximately 5 g of purified sample and put it into a 500 mL glass stoppered-Erlenmeyer flask. Add 100 mL water to the flask and disperse by shaking. By using phenolphthalein as an indicator, drop 0.1 mol/L sodium hydroxide solution into the sample liquid until the color changes slightly red. Then, add 25 mL of 0.45 mol/L sodium hydroxide solution to the flask. Plug the flask, and shake for 30 min at room temperature. Back titrate surplus alkali with 0.2 mol/L hydrochloric acid until the color of phenolphthalein disappears.

Conduct the same procedures for purified raw material starch (or raw starch of the same kind) and use the result as blank data. However, the sampling amount should be very close to that (on a dry basis) of the test material (modified starch).

$$A, \% = \frac{(V_0 - V) \times F \times N \times 0.043 \times 100}{S \times (100 - M)/100}$$

Where —

A : content of acetyl groups, dry base (%)

V_0 : titration volume for blank (mL)

V : titration volume for sample (mL)

F : factor of hydrochloric acid

N : concentration of hydrochloric acid (mol/L)

S : amount of purified sample (g)

M : moisture of purified sample (%)

$$\text{Degree of substitution (ds)} = \frac{162A}{4300 - 42A}$$

Note 5) In case of using 0.2 mol/L hydrochloric acid as titrant, the titration volume for blank solution will exceed 50 mL and therefore even a 50 mL-burette will require additional titrant. However, when 0.25 mol/L hydrochloric acid is used as titrant, the titration volume is reduced within 50 mL.

(3) Determination of the degree of acetylation (Enzymatic method)

(i) Preparation of standard solution for calibration curve

Using a whole pipette, add 10.0 mL of 0.1 mol/L acetic acid solution to a 100 mL-volumetric flask. Dilute to volume with water. Using whole pipettes, add 5.0 mL, 10.0 mL, and 15.0 mL of the solution into separate 100 mL-volumetric flasks. Dilute to volume with water (the concentrations of the standard solutions are 0.03, 0.06, and 0.09 g/L, respectively). Use these solutions as standard solutions for constructing a calibration curve.

(ii) Preparation of test solution for determination of total acetic acid

Accurately weigh approximately 1.2 g of purified sample and put it in a 100 mL-Erlenmeyer flask. Add 50 mL water to the flask and disperse by shaking. Add 5 mL of 2 mol/L-sodium hydroxide to the flask and stir thoroughly. Attach a tube (acting as an air cooler) to the flask and heat it in a boiling water bath for 30 min, while stirring occasionally. After cooling, using phenolphthalein as an indicator, drop 1 mol/L-sulfric acid into the sample liquid until the red color disappears. Transfer this solution to a 100 mL-volumetric flask and dilute to volume with water. Use this liquid as a test solution.

(iii) Preparation of test solution for determination of free acetic acid

Accurately weigh approximately 1.2 g of purified sample and put it in a 100 mL-Erlenmeyer flask. Add 50 mL water to the flask and disperse by shaking. Transfer this liquid to a 100 mL-volumetric flask. Dilute to volume with water and filtrate. Use the filtrate as a test solution.

(iv) Construction of calibration curve

According to the manufacturer's instructions for using an F-kit for determination of acetic acid, measure the absorbance of the standard solutions prepared in 1. above at 340 nm. Construct a calibration curve of acetic acid by plotting the

concentrations of the standard solutions versus their absorbance.

(v) Determination of acetic acid in test solution

According to the manufacturer's instructions for using an F-kit for determination of acetic acid, measure absorbance of the test solutions prepared in 2. and 3. above at 340 nm. Calculate the amount of acetic acid (g/L) in the respective test solutions using the calibration curve prepared in 4. above. Note that the measurement of absorbance in 4. and 5.above should be performed at the same time.

(vi) Calculation of DS value

Calculate the DS value of the sample from the following formula:

$$As,\% = Xs \times 0.7169 \times \frac{100}{1000} \times \frac{1}{(100-M)/100 \times Ws} \times 100$$

$$Ab,\% = Xb \times 0.7169 \times \frac{100}{1000} \times \frac{1}{(100-M)/100 \times Wb} \times 100$$

Where —

As : content of total acetyl groups, dry base (%)

Ab : content of free acetyl group, dry base (%)

Xs : amount of total acetic acid (g/L)

Xb : amount of free acetic acid (g/L)

M : moisture in purified sample (%)

Ws : amount of purified sample for determination of total acetic acid (g)

Wb : amount of purified sample for determination of free acetic acid (g)

$$0.7169 = \frac{CH_3CO}{CH_3COOH}$$

$$\text{Degree of substitution (ds)} = \frac{162 \times (As - Ab)}{4300 - 42 \times (As - Ab)}$$

7.5. Starch Phosphates [determination of phosphorus content by Allen's method]

If the sample is expected to be a starch phosphate, it is required to determine the phosphorus content, according to the following procedures.

Conduct the same procedures for raw material starch (or raw starch of the same kind) purified with a semipermeable membrane, as a reference.

(1) Accurately weigh 1 to 2 g of sample purified with a semipermeable membrane and put it in a decomposition tube.

(2) Add 10 mL of perchloric acid into the tube and heat at 250 °C.

(3) Add 3 drops of nitric acid into the tube when decomposition begins.

(4) Continue heating until the solution becomes colorless and transparent.

(5) After cooling, transfer the solution to a 25 mL-volumetric flask and dilute to volume with water.

(6) Using a whole pipette, transfer 2 mL of the solution to another 25 mL volumetric flask.

(7) Add 2 mL of perchloric acid to the flask.

(8) Add 2 mL of amidol solution to the flask.

(9) Add 1 mL of ammonium molybdate solution to the flask.

(10) Dilute to volume with water

(11) Initiate color reaction by leaving to stand for 20 min

(12) Measure its absorbance at 530 nm.

(13) Conduct the same procedures for 2 mL water as a blank test.

(14) Using 1 to 10 mL of the standard phosphorus solutions, construct a calibration curve by plotting their data.

$$P,\% = \frac{Pm}{S \times (100-M)/100 \times 1000 \times 2/25} \times 100$$

Where —

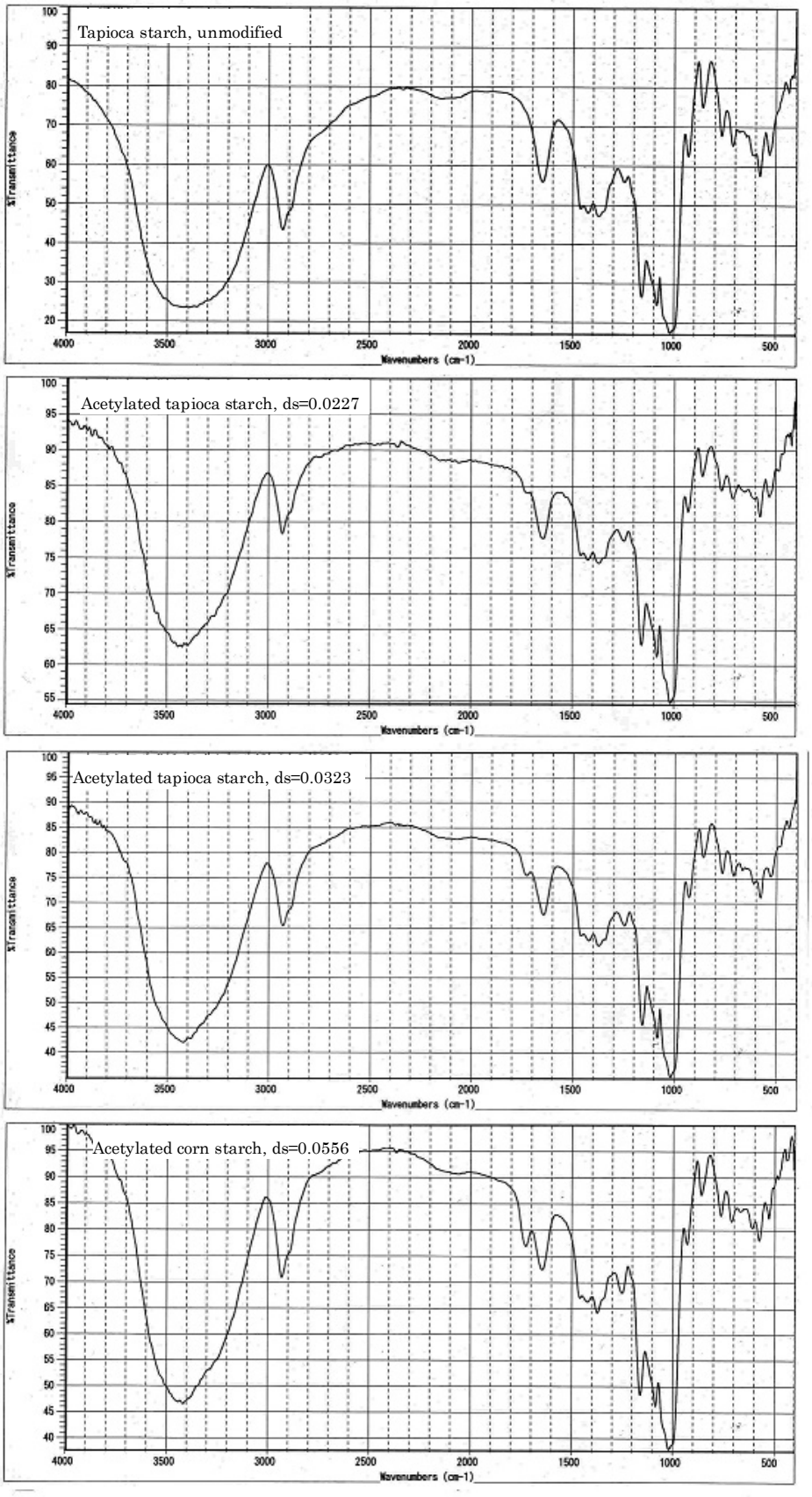
P : phosphorus content (%)

Pm: weight of phosphorous in test solution, calculated from calibration curve (mg)

S : amount of purified sample (g)

M : moisture in purified sample (%)

8. Infrared absorption spectra of acetylated starches



9. Brabender viscogram

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Measurement conditions for viscograms (1) to (23)

Collect a dried test sample to a beaker, add deionized water and make muddy by stirring. (Deionized water used should be 450 mL in total.) Transfer the muddy liquid to a measuring vessel and place it in a brabender viscograph. The viscograms (1) to (23) were recorded under the following conditions:

Rotational speed : 150 rpm (constant)
Temp. Prog. : initial, 30 °C (hold 5 min); program rate, 1.5° C/min; final temp., 92.5 °C (hold 15 min)

Corn starch, acetylated (Australia)	33
Wheat starch (Reagent: Wako Pure Chemical Industries, Ltd.)	34
Wheat starch, acetylated (Australia)	35
Wheat starch, cationic (Australia)	36

Measurement conditions for viscograms (24) to (36)

Collect a dried test sample in a beaker, add deionized water and make muddy by stirring. (Deionized water used should be 450 mL in total.) Transfer the muddy liquid to a measuring vessel and place it in a brabender viscograph. The viscograms (24) to (36) were recorded under the following conditions:

Rotational speed : 75 rpm (constant)
Temp. Prog. : initial, 30°C (hold 5 min); program rate, 1.5° C/min; final temp., 92.0°C (hold 15 min)

Potato starch, containing titanium oxide	37
Potato starch, containing calcium chloride	38
Potato starch, containing calcium hydroxide	39
Potato starch, containing titanium oxide, calcium chloride and calcium hydroxide	40
Tapioca starch, containing titanium oxide, calcium chloride and calcium hydroxide	41
Corn starch, containing titanium oxide, calcium chloride and calcium hydroxide	42
Tragacanth gum, karaya gum, ghatti gum, gum arabic	43
Locust bean gum, guar gum, tamarind gum	44
Agar-agar, carrageenan, sodium alginate	45
Microorganism mucilage, C.M.C., pectin	46
Guar gum derivatives	47
Guar gum, containing titanium oxide, calcium chloride and calcium hydroxide	48

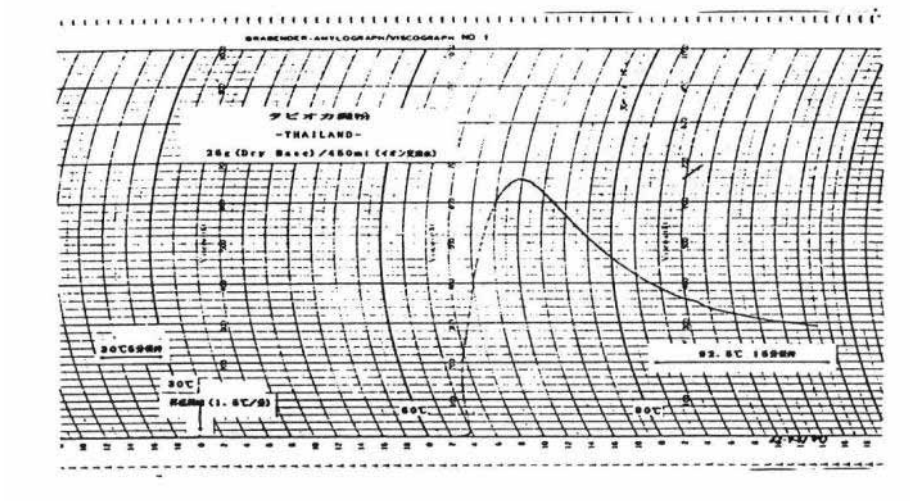
Measurement conditions for viscograms (37) to (48)

Collect a dried test sample in a beaker, add deionized water and make muddy by stirring. (Deionized water used should be 450 mL in total.) Transfer the muddy liquid to a measuring vessel and place it in a brabender viscograph. The viscograms (37) to (48) were recorded under the following conditions:

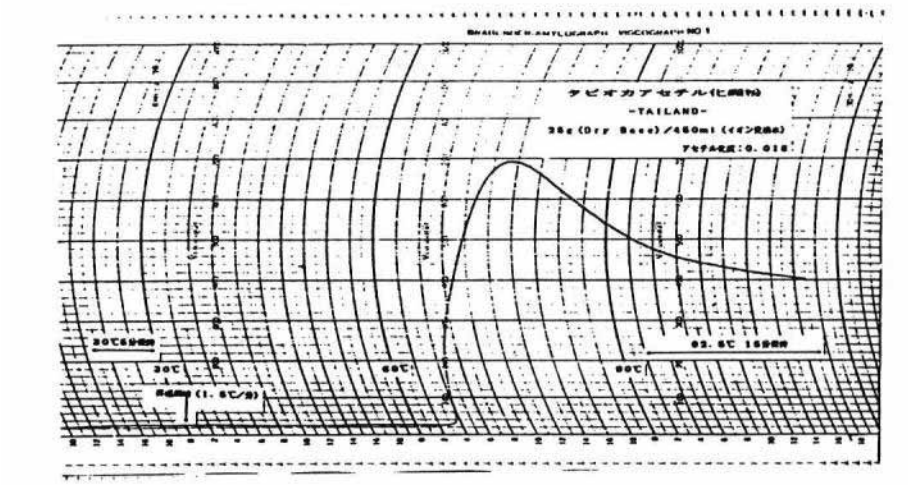
Rotational speed : 75 rpm (constant)
Temp. Prog. : initial, 25°C; program rate, 1.5° C/min; temp., 92.5°C (hold 10 min); program rate, -1.5° C/min; final temp., 80.0°C.

As for the details, refer to Report of the Central Customs Laboratory, Vol. 17, 51-57 (1977), "*Viscosity Curves of Polysaccharides by Viscograph*".

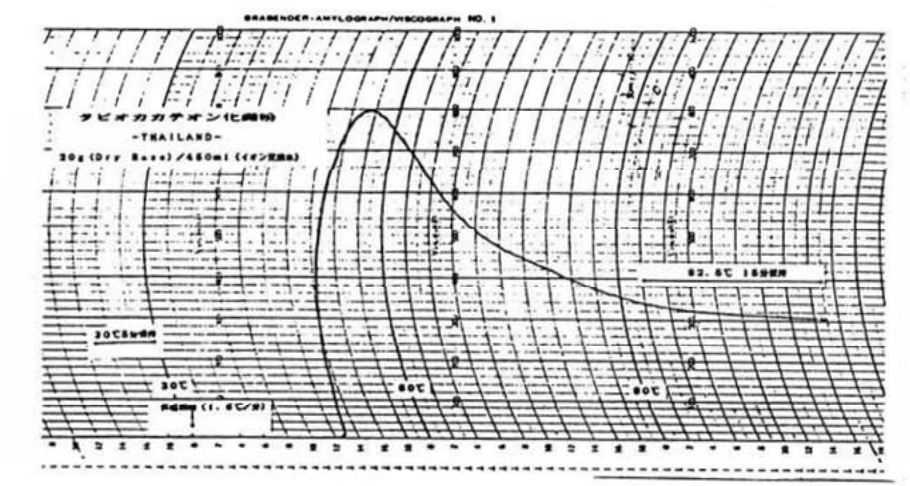
(1) Tapioca starch (THAILAND)



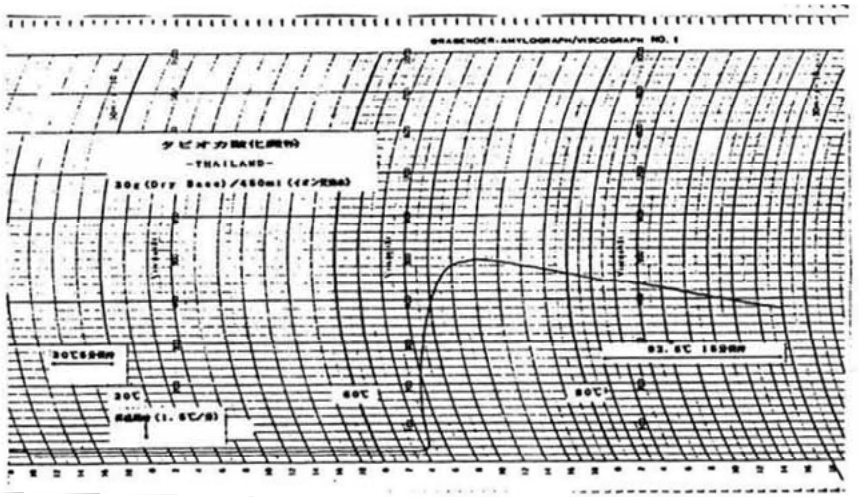
(2) Acetylated tapioca starch (THAILAND)



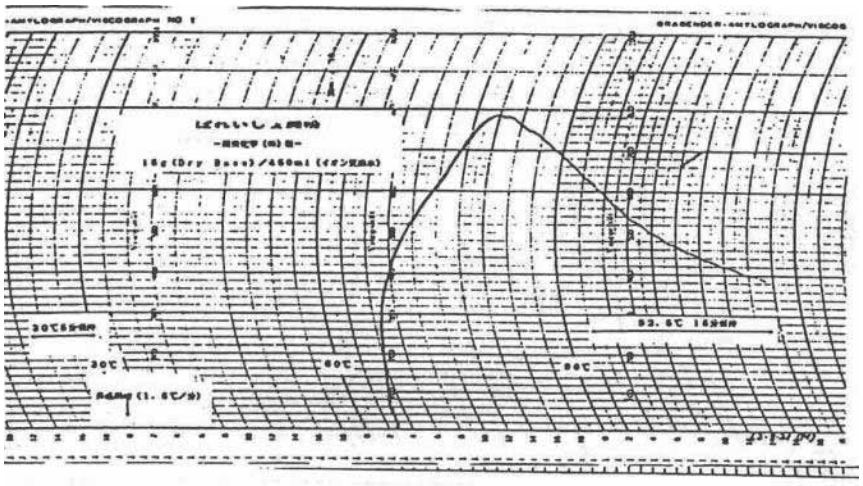
(3) Cationic tapioca starch (THAILAND)



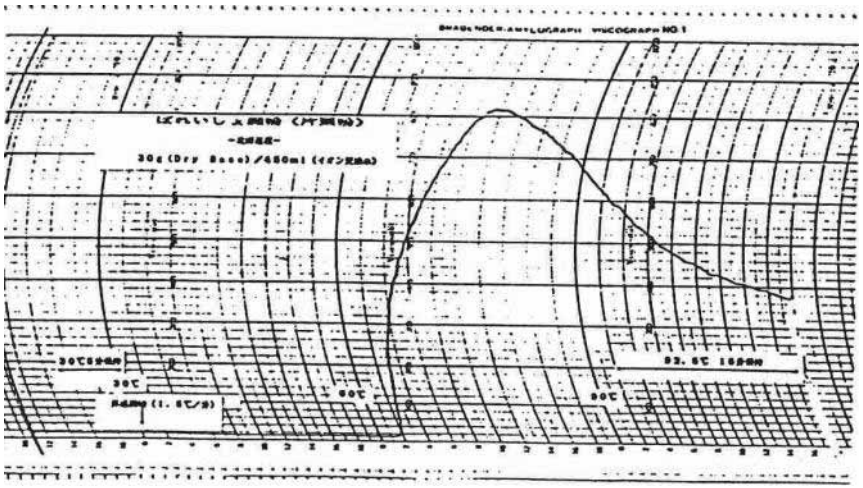
(4) Oxidized tapioca starch (THAILAND)



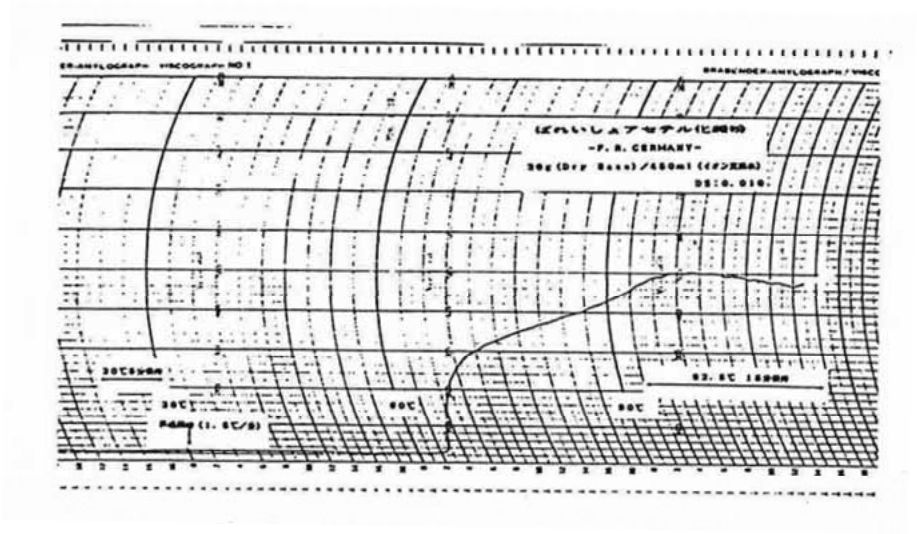
(5) Potato starch (Reagent: Kanto Chemical Co., Inc.)



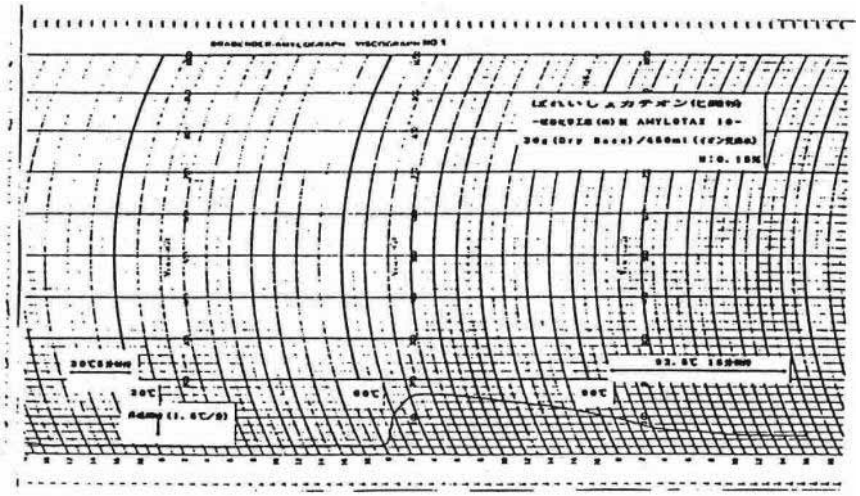
(6) Potato starch (Hokkaido, Japan)



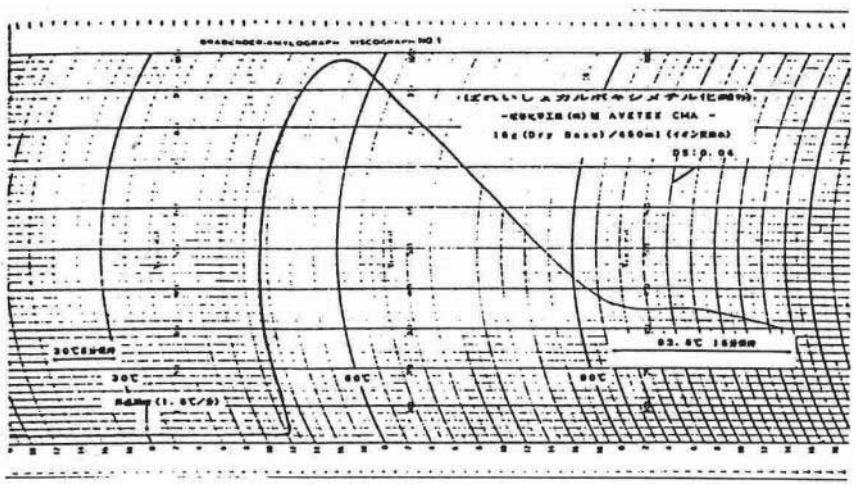
(7) Potato starch, acetylated (Germany)



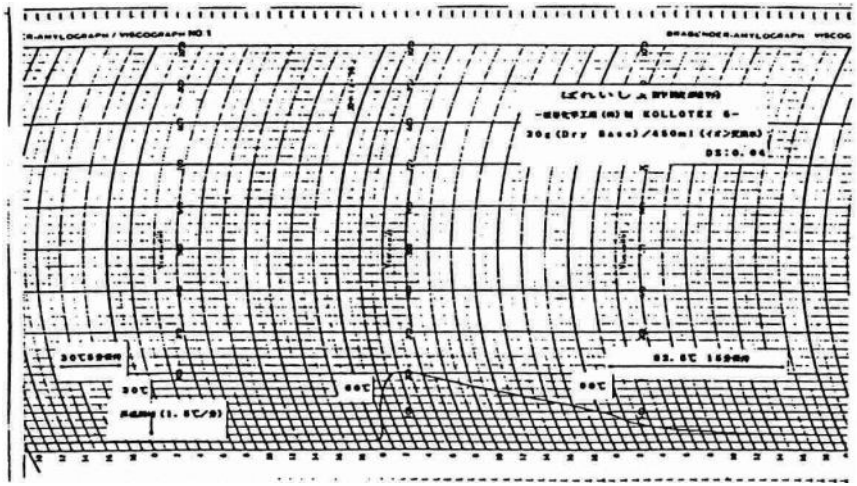
(8) Potato starch, cationic (Matsutani Chemical industry Co, Ltd.)



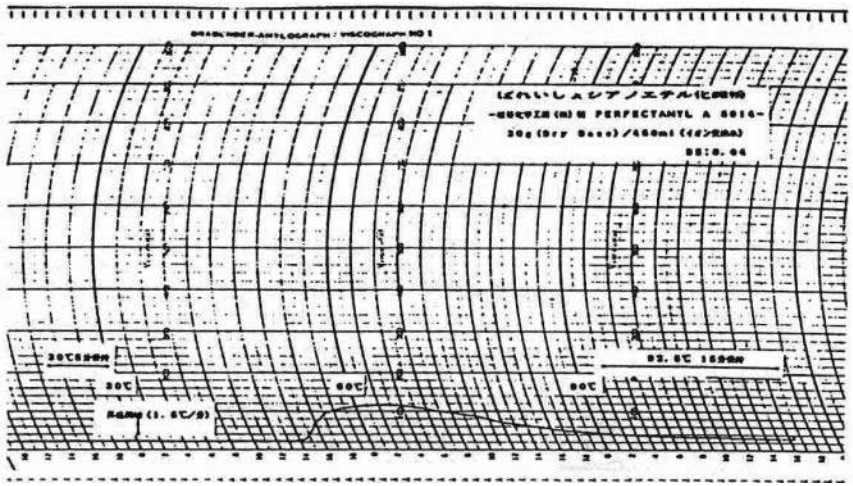
(9) Potato starch, carboxymethylated (Matsutani Chemical industry Co, Ltd.)



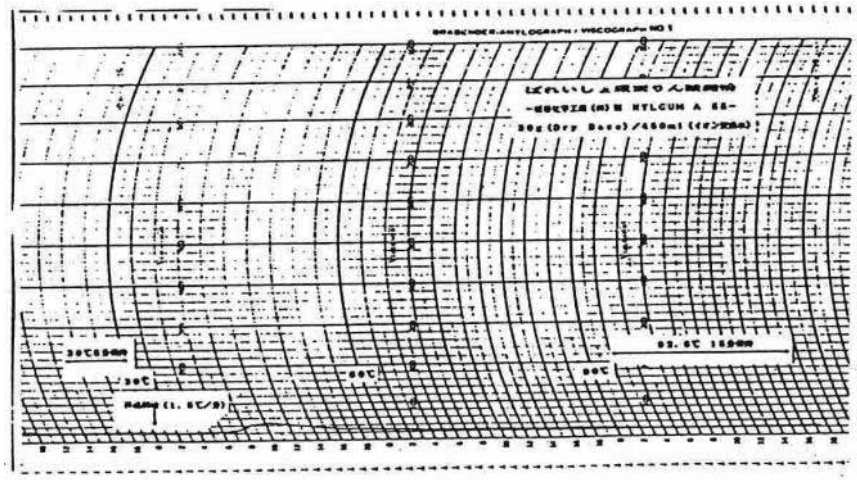
(10) Potato starch, acetylated (Matsutani Chemical industry Co, Ltd.)



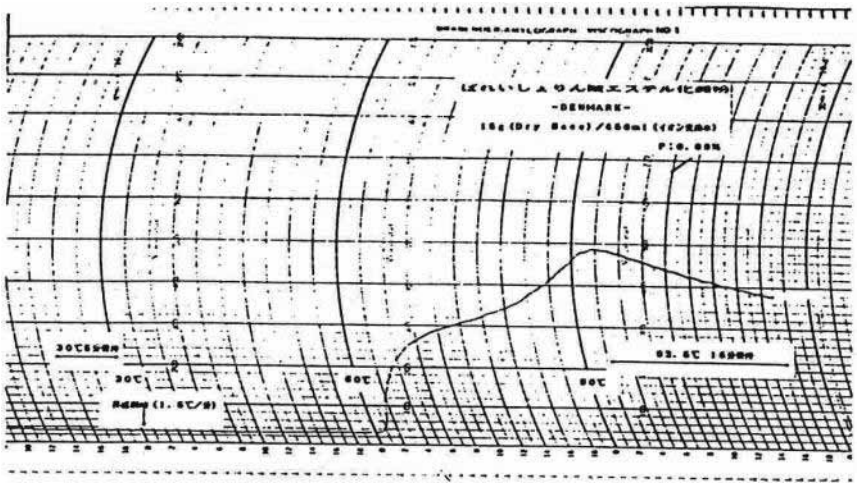
(11) Potato starch, cyano-ethylated (Matsutani Chemical industry Co, Ltd.)



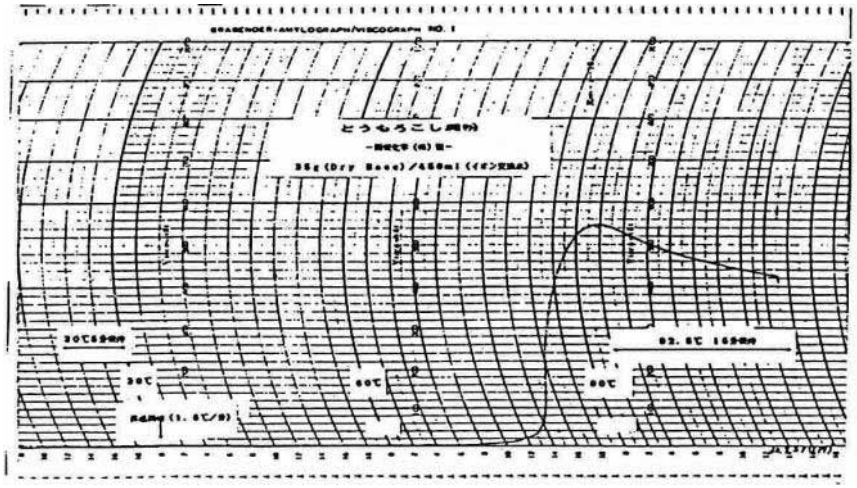
(12) Potato starch, urea-phosphate (Matsutani Chemical industry Co, Ltd.)



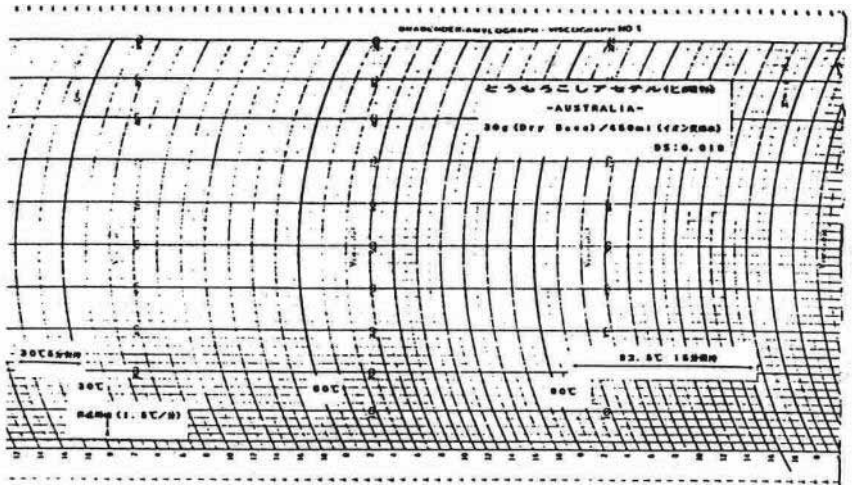
(13) Potato starch phosphate (Denmark)



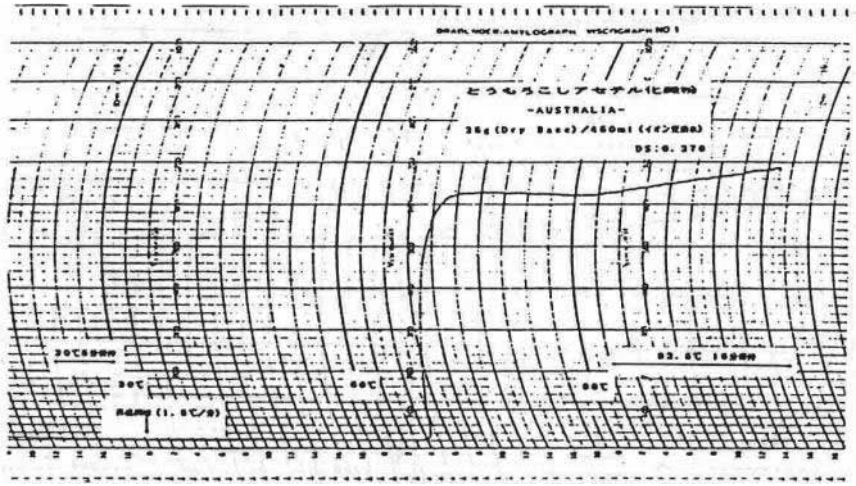
(14) Corn starch (Reagent: Kanto Chemical Co., Inc.)



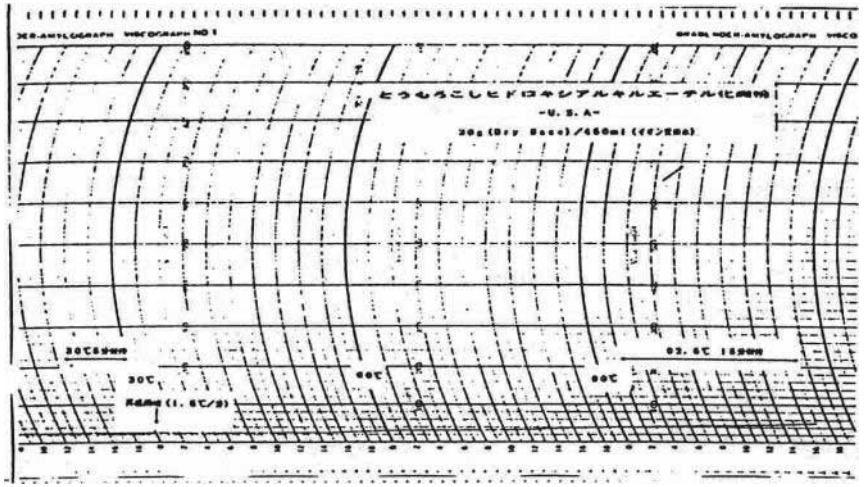
(15) Corn starch, acetylated (Australia)



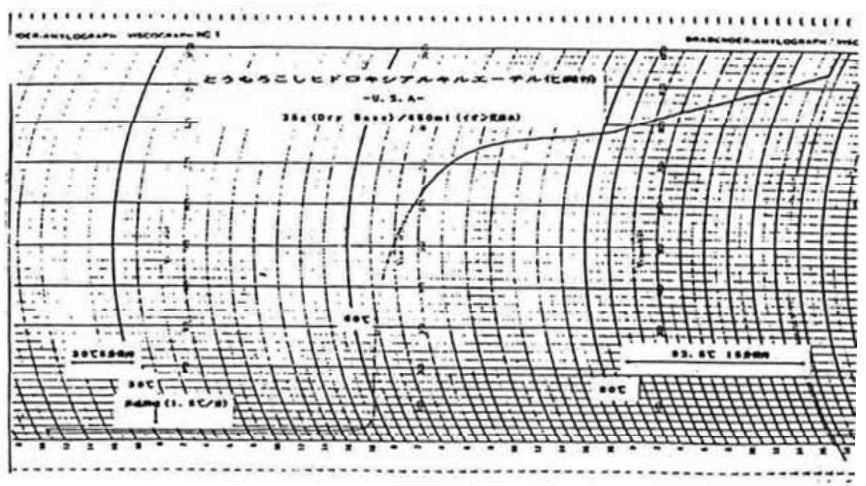
(16) Corn starch, acetylated (Australia)



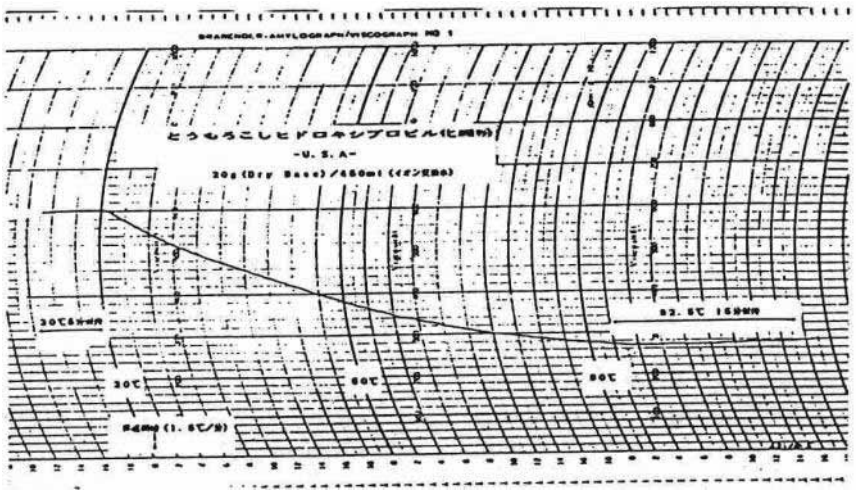
(17) Corn starch, hydroxylalkyletherified (U.S.A.)



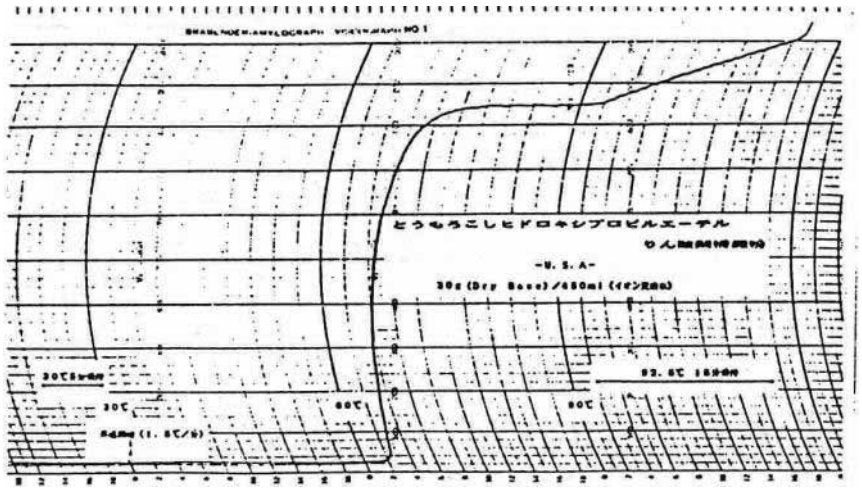
(18) Corn starch, hydroxylalkyletherified (U.S.A.)



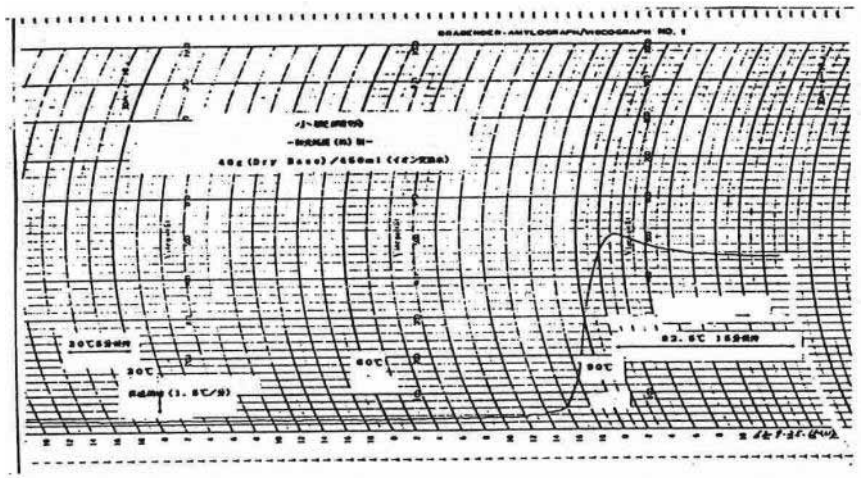
(19) Corn starch, hydroxypropylated (U.S.A.)



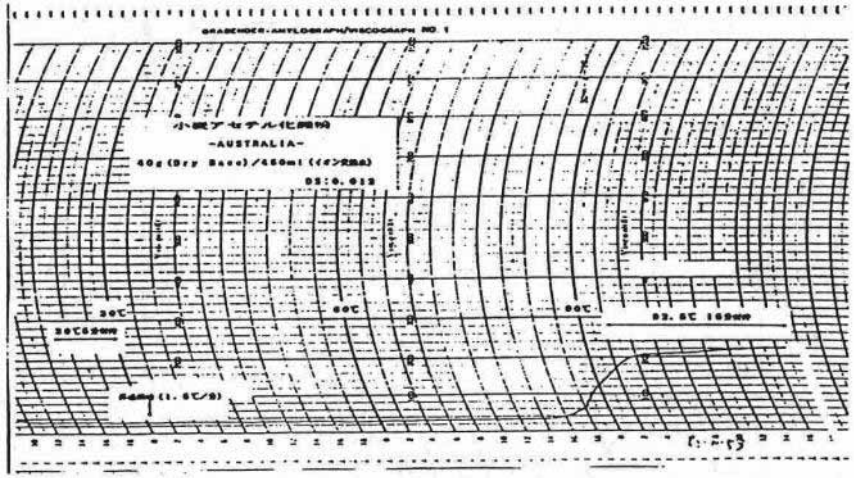
(20) Corn starch, hydroxypropylated, phosphate (cross-linked) (U.S.A.)



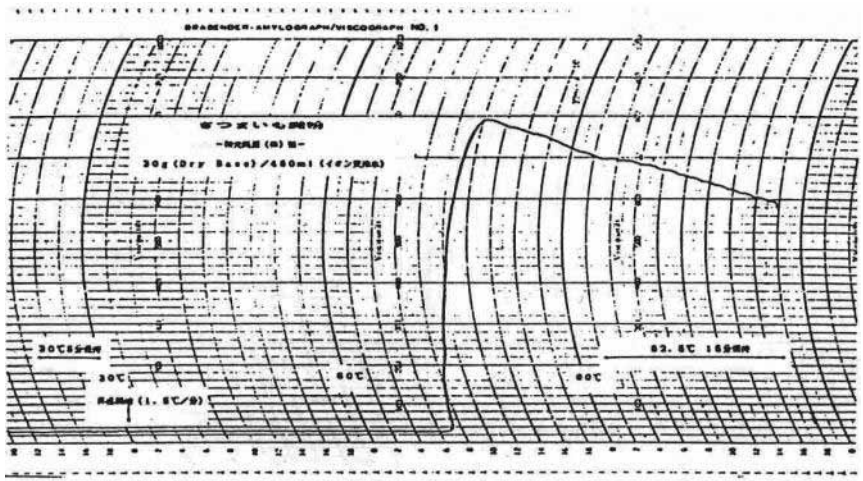
(21) Wheat starch (Reagent: Wako Pure Chemical Industries, Ltd.)



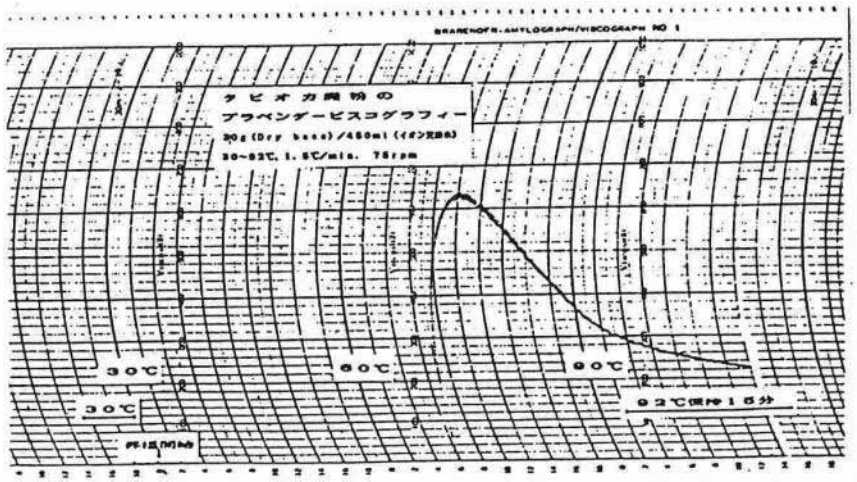
(22) Wheat starch, acetylated (Australia)



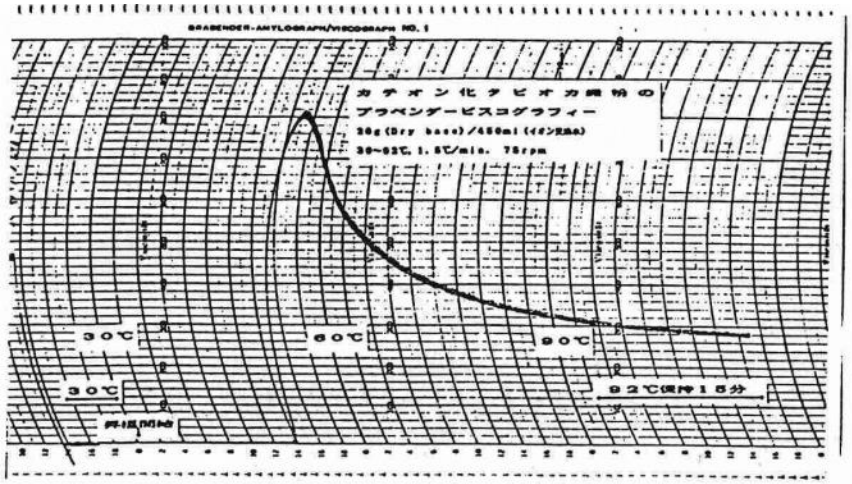
(23) Sweet potato starch (Reagent: Wako Pure Chemical Industries, Ltd.)



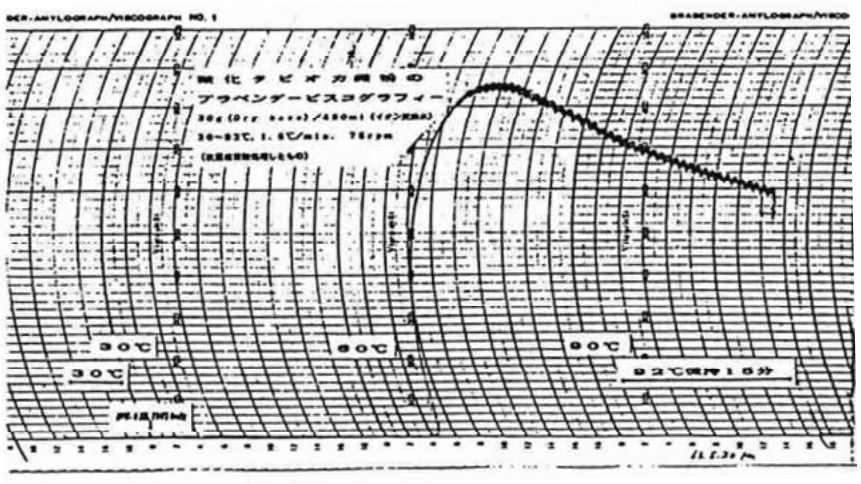
(24) Tapioca starch (Thailand)



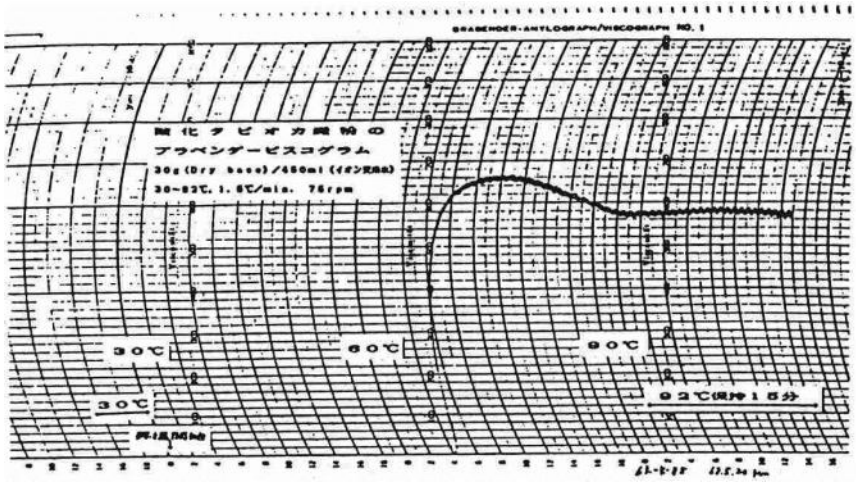
(25) Tapioca starch, cationic (Thailand)



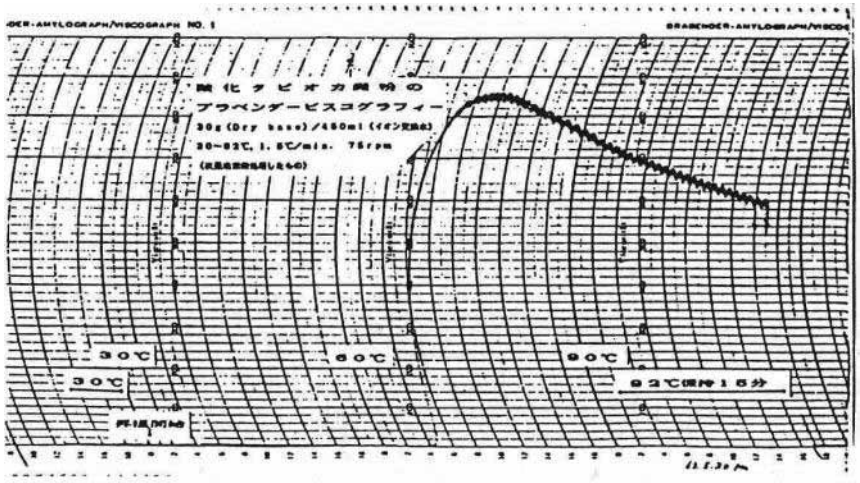
(26) Tapioca starch, oxidized (Thailand)



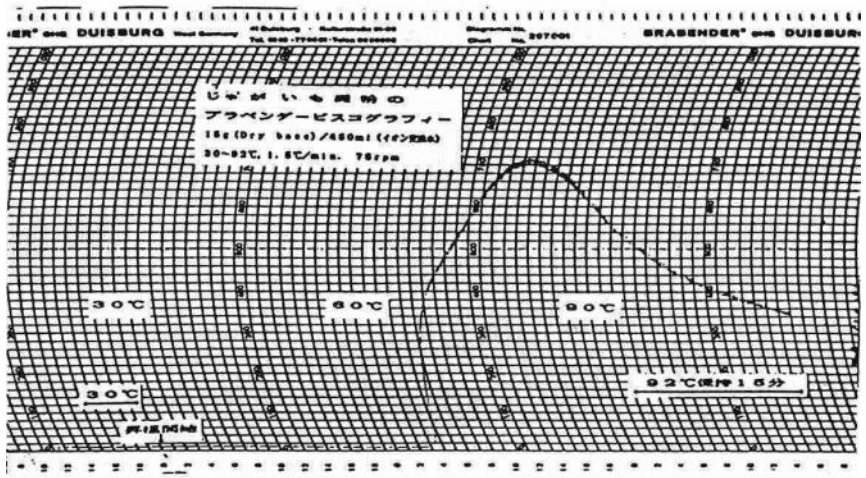
(27) Tapioca starch, oxidized (Thailand)



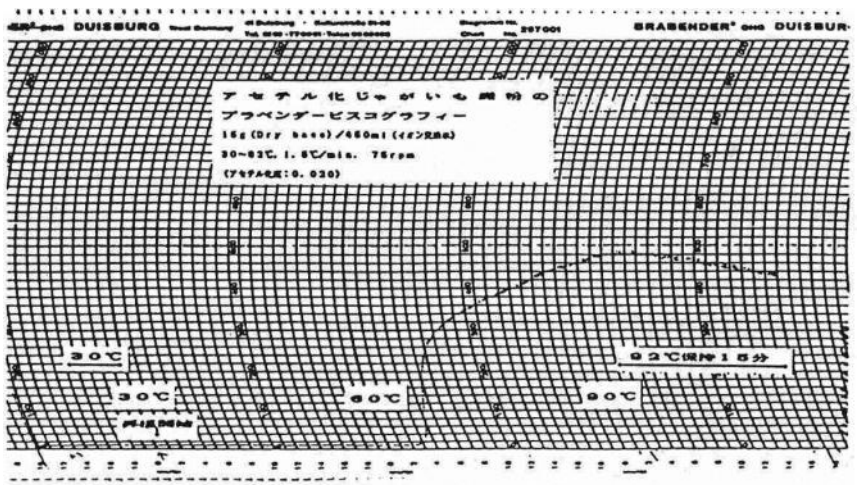
(28) Tapioca starch, oxidized (treated with hypochlorous acid; Thailand)



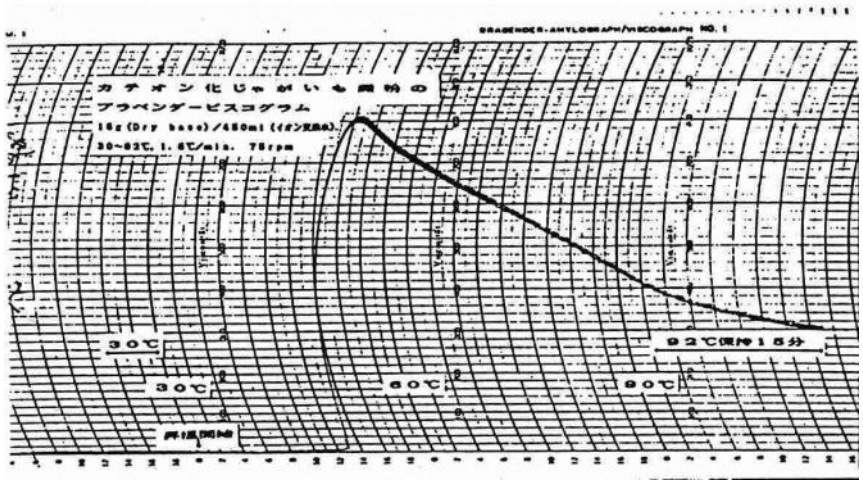
(29) Potato starch (Reagent: Kanto Chemical Co., Inc.)



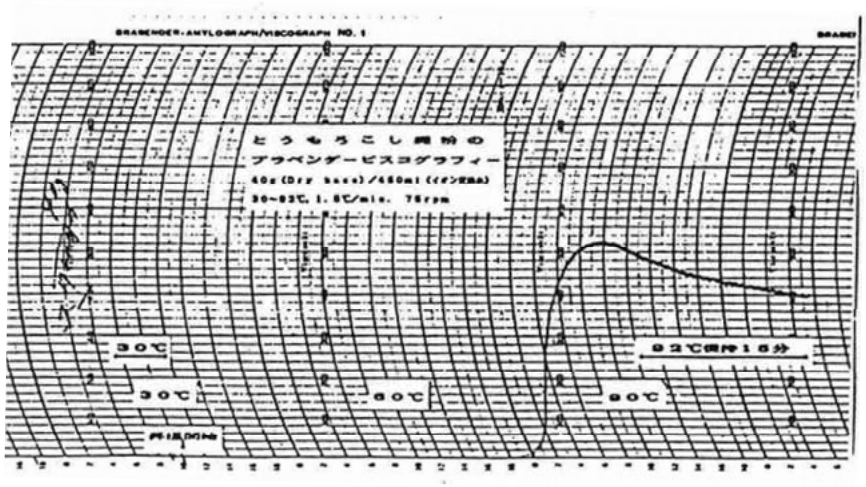
(30) Potato starch, acetylated (Germany)



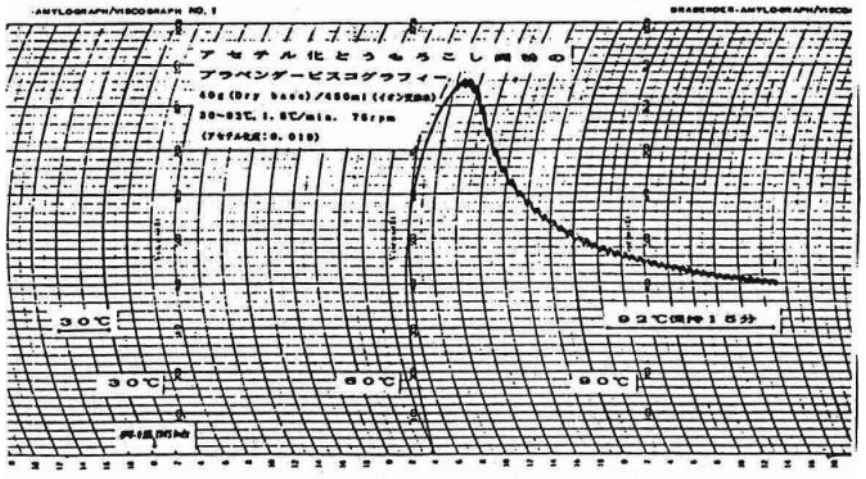
(31) Potato starch, cationic (Germany)



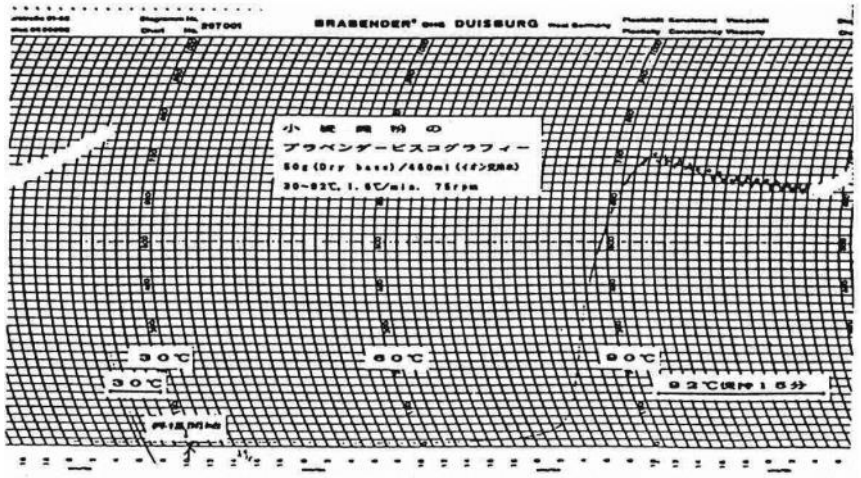
(32) Corn starch (Reagent: Kanto Chemical Co., Inc.)



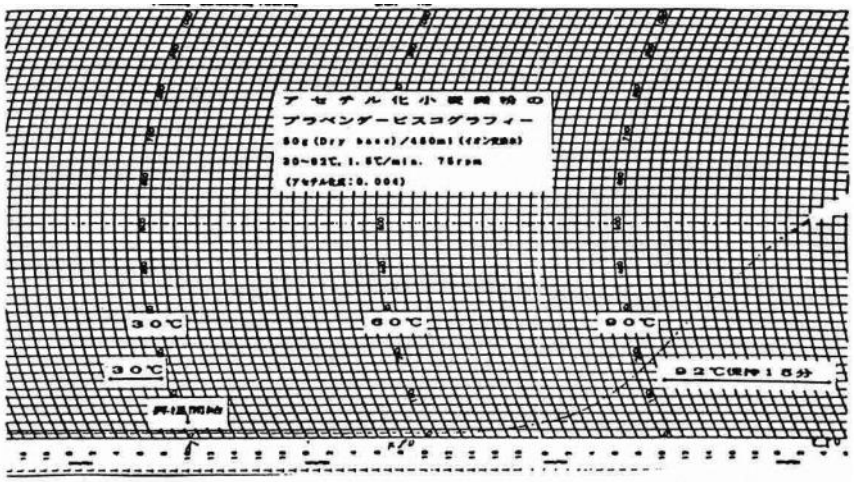
(33) Corn starch, acetylated (Australia)



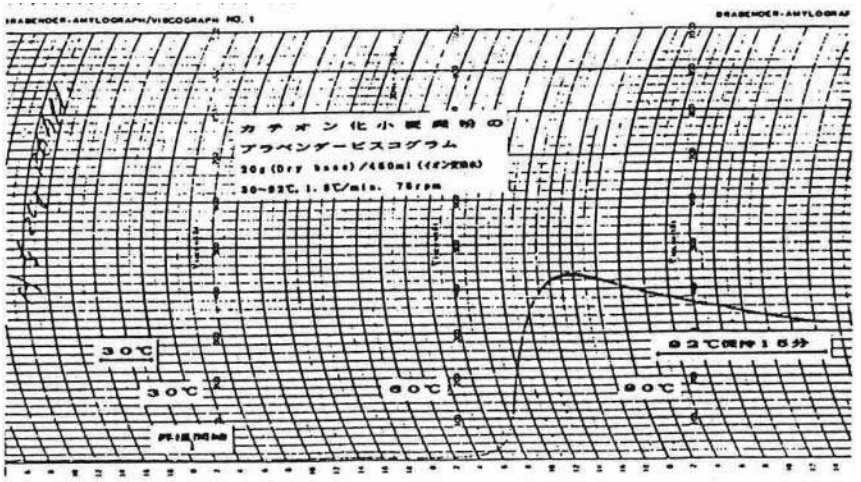
(34) Wheat starch (Reagent: Wako Pure Chemical Industries, Ltd.)



(35) Wheat starch, acetylated (Australia)

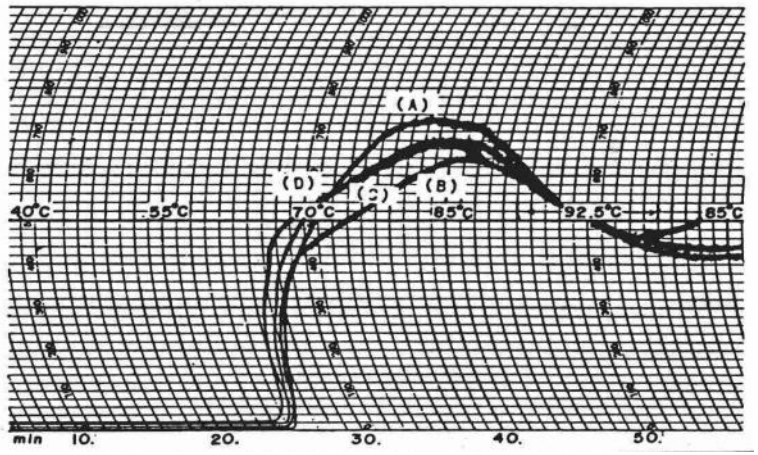


(36) Wheat starch, cationic (Australia)



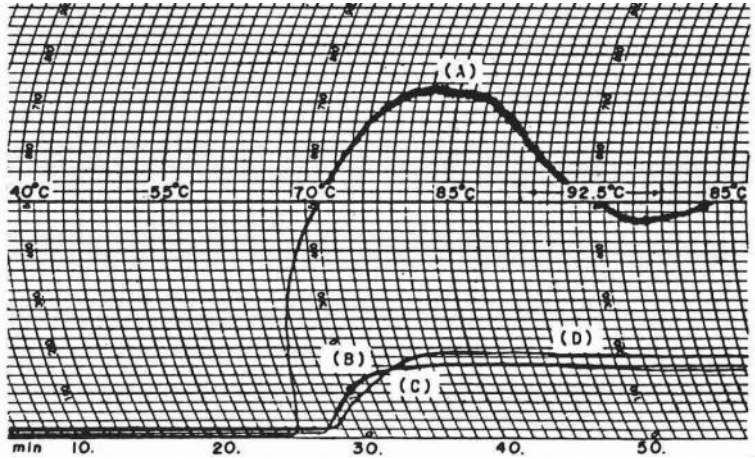
(37) Potato starch, containing titanium oxide

(A) Potato starch 20 g; (B) Potato starch 20 g + TiO_2 1g; (C) Potato starch 20 g + TiO_2 2.5 g; (D) Potato starch 20 g + TiO_2 5g



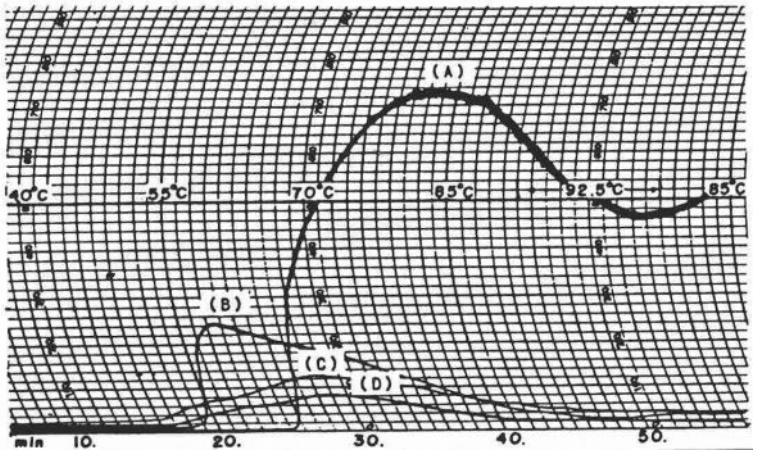
(38) Potato starch, containing calcium chloride

(A) Potato starch 20 g; (B) Potato starch 20 g + $CaCl_2$ 1g; (C) Potato starch 20 g + $CaCl_2$ 2.5 g; (D) Potato starch 20 g + $CaCl_2$ 5 g



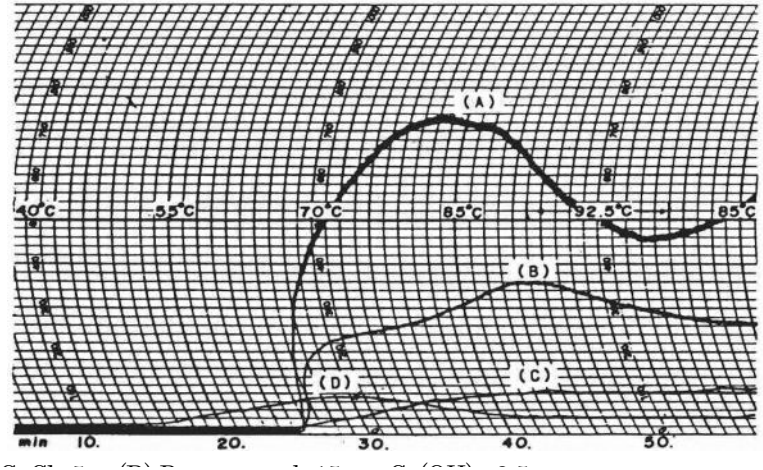
(39) Potato starch, containing calcium hydroxide

(A) Potato starch 20 g; (B) Potato starch 20 g + $Ca(OH)_2$ 1 g; (C) Potato starch 20 g + $Ca(OH)_2$ 2.5 g; (D) Potato starch 20 g + $Ca(OH)_2$ 5g



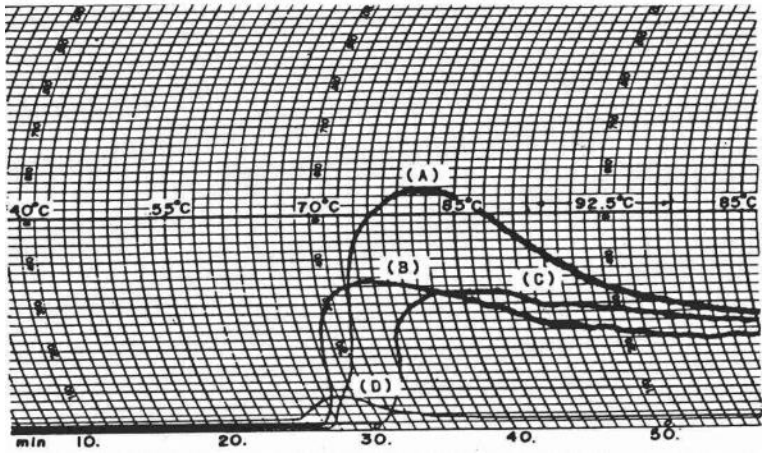
(40) Potato starch, containing titanium oxide, calcium chloride and calcium hydroxide

(A) Potato starch 20 g; (B) Potato starch 15 g + TiO_2 15 g; (C) Potato starch 15



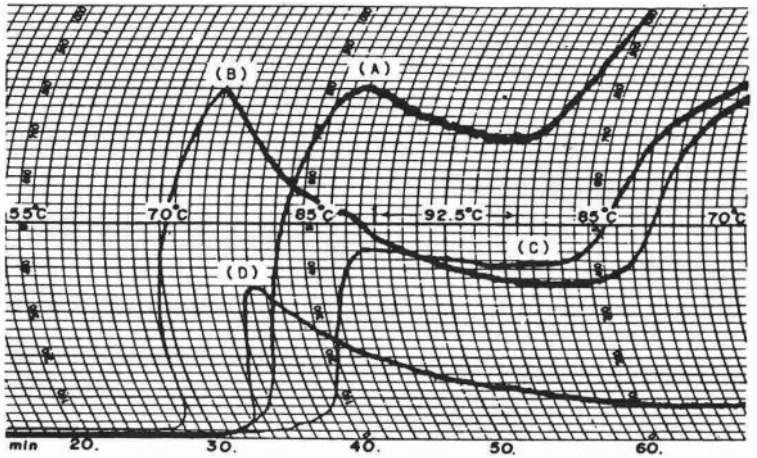
(41) Tapioca starch, containing titanium oxide, calcium chloride and calcium hydroxide

(A) Tapioca starch 30 g; (B) Tapioca starch 25 g + TiO_2 5 g; (C) Tapioca starch 25 g + $CaCl_2$ 5 g; (D) Tapioca starch 25 g + $Ca(OH)_2$ 5 g



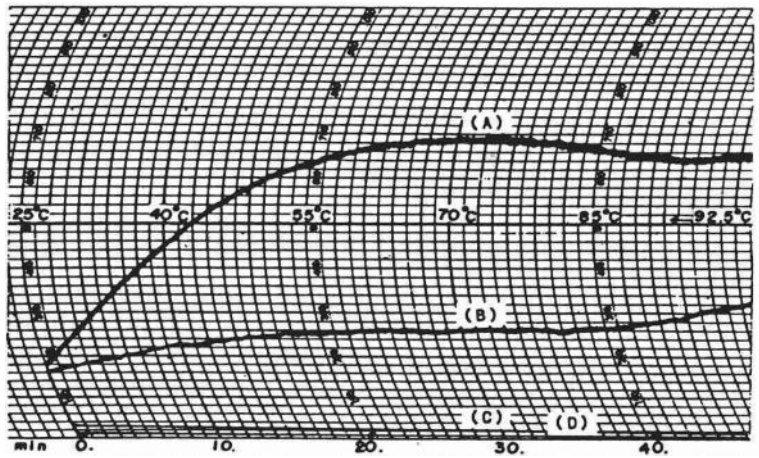
(42) Corn starch, containing titanium oxide, calcium chloride and calcium hydroxide

(A) Corn starch 50 g; (B) Corn starch 40 g + TiO_2 10 g; (C) Corn starch 40 g + $CaCl_2$ 10 g; (D) Corn starch 40 g + $Ca(OH)_2$ 10 g



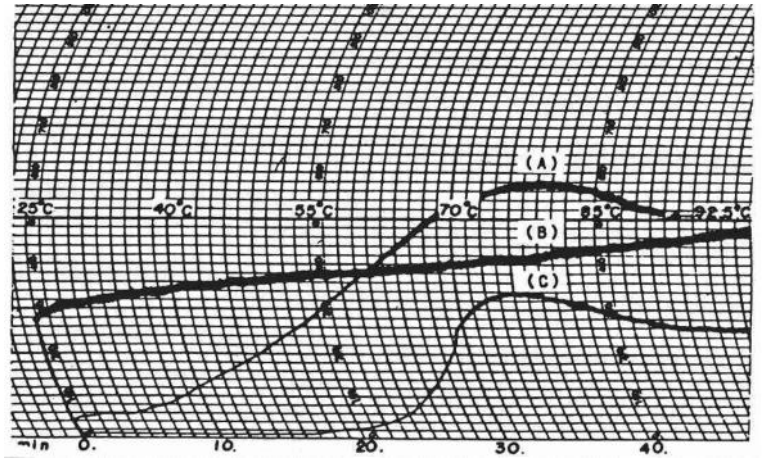
(43) Tragacanth gum, karaya gum, ghatti gum, gum Arabic

(A) Tragacanth 15 g; (B) Karaya 15 g; (C) Ghatti 20 g; (D) Arabic 40 g



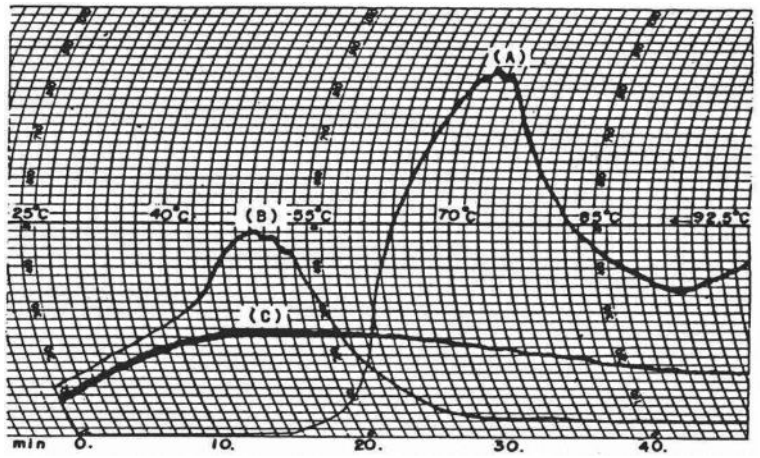
(44) Locust bean gum, guar gum, tamarind gum

(A) Locust bean 7 g; (B) Guar 10g; (C) Tamarind 20 g



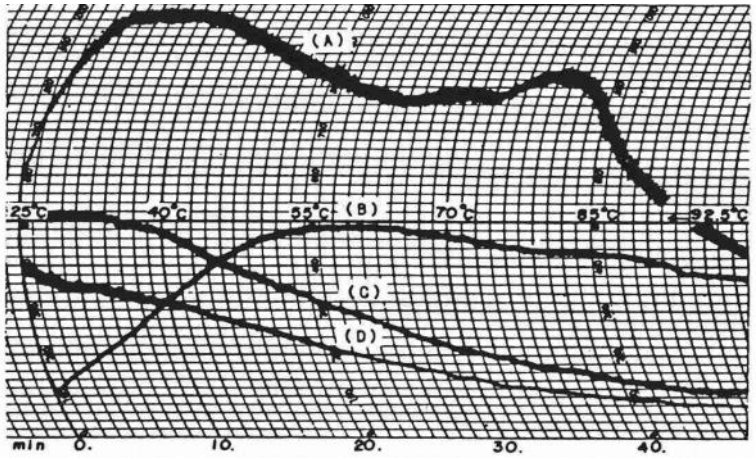
(45) Agar-agar, carrageenan, sodium alginate

(A) Agar-agar 20 g; (B) Carrageenan 7 g; (C)Sodium alginate 10 g



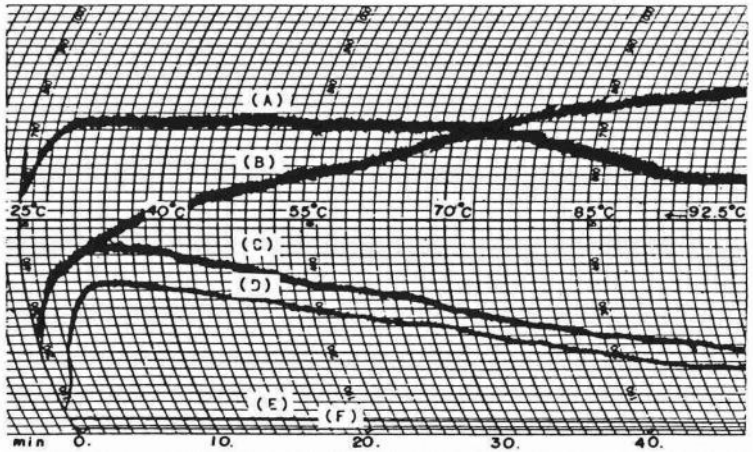
(46) Microorganism mucilage, C.M.C., pectin

(A) Xanthan 7 g; (B) Kelzane 10 g; (C) C.M.C. 10 g; (D) Pectin 30g



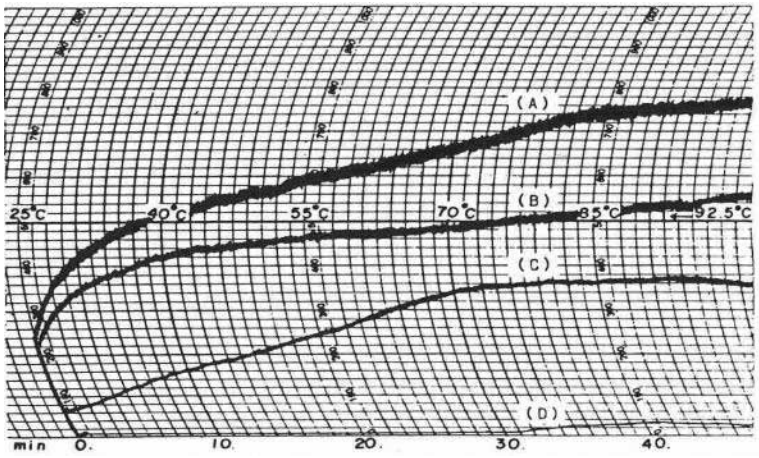
(47) Guar gum derivatives

(A) Cationic 10 g; (B) Guar 10 g; (C) Carboxy methyl (I) 10 g; (D) Carboxy methyl (II) 10 g; (E) High carboxy methyl 10 g; (F) Hydroxy propyl 10 g



(48) Guar gum, containing titanium oxide, calcium chloride and calcium hydroxide

(A) Guar 10 g; (B) Guar 7 g + TiO_2 3 g; (C) Guar 7 g + $CaCl_2$ 3 g; (D) Guar 7 g + $Ca(OH)_2$ 3 g



10. References

(1) Sekikawa Y., Shimada M. (1987), Reports of the Central Customs Laboratory **27**: 1 (in Japanese).

(2) Ujihara S., Sekikawa Y., Shimada M. (1987), Reports of the Central Customs Laboratory **27**: 25 (in Japanese).

(3) Kawabata S., Ohno Y. (1983), Reports of the Central Customs Laboratory **23**: 9 (in Japanese).

(4) Sekikawa Y., Ohno Y. (1983), Reports of the Central Customs Laboratory **24**: 17 (in Japanese).

(5) Kawabata S., Inoue T. (1981), Reports of the Central Customs Laboratory **22**: 57 (in Japanese).

(6) 鈴木繁男, 中村道徳編 「澱粉科学実験法」 朝倉書店 (1979).