GENERAL TESTS, PROCESSES AND APPARATUS

General Tests, Processes and Apparatus includes common methods for tests and other articles related to them. Unless otherwise specified, the procedures for absorbance determination, absorbance ratio determination, acid-neutralizing capacity determination of gastrointestinal medicines, alcohol number determination, ammonium determination, arsenic determination, atomic absorption spectrophotometry, test for bacterial endotoxins, boiling point determination, distilling range determination, chloride determination, congealing point determination, test for content uniformity, digestion test, disintegration test, dissolution test, endpoint detection in titrimetry, flame coloration, fluorometry, foreign insoluble matter test for injections, gas chromatography, heavy metals determination, infrared spectrophotometry, insoluble particulate matter test for injections, insoluble particulate matter test for ophthalmic solutions, iron determination, liquid chromatography, loss on drying determination, loss on ignition determination, mass variation test, melting point determination, methanol determination, methoxyl assay, test for microbial limit, test for microbial linit for crude drugs, microbiological potency determination for antibiotics, mineral oil determination, nitrogen determination, nuclear magnetic resonance spectroscopy, optical rotation determination, osmolarity determination, oxygen flask combustion method, paper chromatography, particle size distribution test for preparations, pH determination, powder particle size determination, test for pyrogen, qualitative test, test for readily carbonizable substances, refractive index determination, residual solvents test, residue on ignition determination, specific gravity and density determination, specific surface area determination, test for sterility, sulfate determination, test for glass containers for injections, test for metal particles in ophthalmic ointments, test for plastic containers, test for rubber closure for aqueous infusions, test for total organic carbon, thermal analysis, thin-layer chromatography, viscosity determination, vitamin A assay, test for volatile contaminants in ethanol, water determination, and X-ray powder diffraction are performed as directed in the corresponding articles under the General Tests, Processes and Apparatus. The tests for melting point of fats, congealing point of fatty acids, specific gravity, acid value, saponification value, ester value, hydroxyl value, unsaponifiable matter and iodine value of fats and fatty oils are performed as directed in the corresponding items under the Fats and Fatty oils Test, and the tests for foreign matter and loss on drying, total ash, acid-insoluble ash, extract content, essential oil content of crude drugs are performed as directed in the corresponding items under the Crude Drugs Test.

1. Absorbance Ratio Method

The Absorbance Ratio Method is a method of quantitative analysis involving the determination of the equivalence point of an acid-base reaction by using spectrophotometry, and is applied to the determinations of substances whose content is previously estimated to be within a certain range.

To the sample solution add a definite volume of a standard solution for volumetric analysis in one portion to make the solution near the end-point of titration, and read the absorbances of this solution at the maximum wavelengths of the absorbance spectrum for the acidic and alkaline forms of the added indicator. From the relationship between the ratio of these absorbances and the inverse of the molarity, determine the true volume of the standard solution for volumetric analysis consumed to complete the titration of the sample.

For an acid-base reaction select an indicator which has a *pKa* value around the pH of the equivalence point and has a marked difference between the absorption spectrum of acid color and that of alkali color; the maximum wavelengths of these absorption spectra are taken as λ_1 and λ_2, respectively (λ_1 is the shorter). When the absorbances of the reaction solution which contains this indicator at λ_1 and λ_2 are A_1 and A_2, respectively, the absorbance ratio (r) is defined by the following equation:

$$r = \frac{A_2}{A_1 + A_2}$$

The volume of the standard solution for volumetric analysis corresponding to the true equivalence point is supposed to be V mL, the actually added volume of the standard solution for volumetric analysis is to be V' mL, and the ratio of them is to be x.

$$x = \frac{V}{V'}$$

Prepare a series of standard solutions with various values of x in advance under definite conditions using the standard substance and the prescribed pH indicator, measure the absorbances, A_1 and A_2, to make a table of relationships between x and r, and draw the x-r curve. The table of x-r relationships is specified in each monograph. As for the sample, A_1 and A_2 are measured under the same conditions, the r value is calculated from the above formula, the x value is estimated from the x-r curve, and the content S (g) of the chemically pure substance in the sample taken is calculated from the following equation.

$$\text{S (g)} = V' \times f \times x \times \text{M}$$

f: The molarity coefficient of the standard solution for volumetric analysis.

M: The amount (g) of the chemically pure substance corresponding to 1 mL of the standard solution for volumetric analysis in the case of $f = 1$.

Procedure

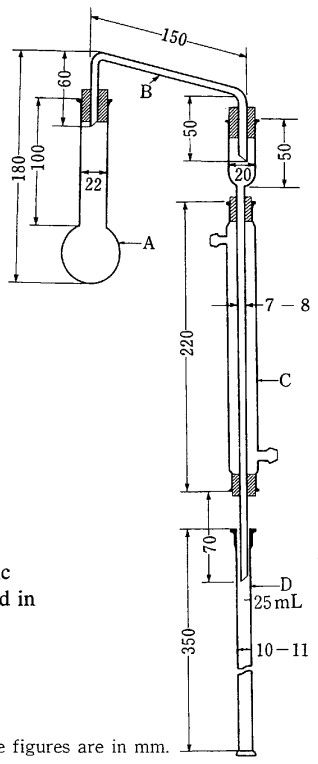
Under the conditions directed in the monograph, proceed as follows. Dry and weigh accurately the sample, and transfer it into a volumetric flask. Add V' mL of the standard solution for volumetric analysis with a pipet, and if directed in the monograph, add the specified solvent with a pipet. Then add the pH indicator, and dissolve the sample in water to make a definite volume. Use a spectrophotometer, and read the absorbances, A_1 and A_2, of this solution at λ_1 and λ_2 in a 10-mm cell, unless otherwise specified, at ordinary temperature using water as a blank. Calculate the value of r by using the formula. Estimate the value of x, using the obtained value of r and the x-r curve obtained from the table of relationships between x and r described in the monograph. Calculate the amount (g) of the chemically pure substance in the sample taken.

2. Alcohol Number Determination

The Alcohol Number Determination represents the number of milliliters of ethanol at 15°C obtained from 10 mL of tincture or other preparations containing ethanol by the following procedures.

Method 1 Distilling method

This is a method to determine the Alcohol Number by reading the number of milliliters of ethanol distillate at 15°C obtained from 10 mL of a sample measured at 15°C by the following procedures.



A: Distilling flask (50 mL)
B: Delivery tube
C: Condenser
D: Glass-stoppered volumetric cylinder (25 mL, graduated in 0.1 mL)

The figures are in mm.

Ethanol content in the sample (vol%)	Distillate to be collected (mL)
above 80	13
80 - 70	12
70 - 60	11
60 - 50	10
50 - 40	9
40 - 30	8
below 30	7

(1) Apparatus

Use hard glass apparatus as illustrated herein. Ground glass may be used for the joints.

(2) Reagent

Alkaline phenolphthalein solution: To 1 g of phenolphthalein add 7 mL of sodium hydroxide TS and water to make 100 mL.

(3) Procedure

Transfer 10 mL of the sample preparation, accurately measured at 15 ± 2°C, to the distilling flask A, add 5 mL of water and boiling chips. Distil ethanol carefully into the glass-stoppered, volumetric cylinder D.

By reference to the following Table, a suitable volume of distillate (mL) should be collected, according to the content of ethanol in the sample preparation.

Prevent bumping during distillation by rendering the sample strongly acidic with phosphoric acid or sulfuric acid, or by adding a small amount of paraffin, beeswax or silicone resin before starting the distillation.

When the samples contain the following substances, carry out pretreatment as follows before distillation.

(i) Glycerin: Add sufficient water to the sample so that the residue in the distilling flask, after distillation, contains at least 50% of water.

(ii) Iodine: Decolorize the sample with zinc powder.

(iii) Volatile substances: Preparations containing appreciable proportions of essential oil, chloroform, diethyl ether or camphor require treatment as follows. Mix 10 mL of the sample, accurately measured, with 10 mL of saturated sodium chloride solution in a separator, add 10 mL of petroleum benzin, and shake. Collect the separated aqueous layer. The petroleum benzin layer was extracted with two 5 mL portions of saturated sodium chloride solution. Combine the aqueous layers, and distill. According to the ethanol content in the sample, collect a volume of distillate 2 to 3 mL more than that shown in the above Table.

(iv) Other substances: Render preparations containing free ammonia slightly acidic with dilute sulfuric acid. If volatile acids are present, render the preparation slightly alkaline with sodium hydroxide TS, and if the preparations contain soap along with volatile substances, decompose the soap with an excess of dilute sulfuric acid before the extraction with petroleum benzin in the treatment described in (iii).

To the distillate add 4 to 6 g of potassium carbonate and 1 to 2 drops of alkaline phenolphthalein solution, and shake vigorously. If the aqueous layer shows no white turbidity, agitate the distillate with additional potassium carbonate. After allowing to stand in water at 15 ± 2°C for 30 minutes, read the volume of the upper reddish ethanol layer in mL, and regard it as the Alcohol Number. If there is no clear boundary surface between these two layers, shake vigorously