ethanol (95) and in diethyl ether.

It effloresces in warm, dry air.

Identification A solution of Dibasic Sodium Phosphate (1 in 10) responds to the Qualitative Tests (1) and (2) for sodium salt, and the Qualitative Tests for phosphate.

pH Dissolve 1.0 g of Dibasic Sodium Phosphate in 50 mL of water: the pH of this solution is between 9.0 and 9.4.

Purity **(1)** Clarity and color of solution—Dissolve 1.0 g of Dibasic Sodium Phosphate in 20 mL of water: the solution is clear and colorless.

(2) Chloride—Dissolve 1.0 g of Dibasic Sodium Phosphate in 7 mL of dilute nitric acid and water to make 50 mL. Perform the test using this solution as the test solution. Prepare the control solution with 0.40 mL of 0.01 mol/L hydrochloric acid VS (not more than 0.014%).

(3) Sulfate—Dissolve 0.5 g of Dibasic Sodium Phosphate in 2 mL of dilute hydrochloric acid and water to make 50 mL. Perform the test using this solution as the test solution. Prepare the control solution with 0.40 mL of 0.005 mol/L sulfuric acid VS (not more than 0.038%).

(4) Carbonate—To 2.0 g of Dibasic Sodium Phosphate add 5 mL of water, boil, and add 2 mL of hydrochloric acid after cooling: the solution does not effervesce.

(5) Heavy metals—Dissolve 2.0 g of Dibasic Sodium Phosphate in 4 mL of acetic acid (31) and water to make 50 mL. Perform the test using this solution as the test solution. Prepare the control solution with 2.0 mL of Standard Lead Solution by adding 2 mL of dilute acetic acid and water to make 50 mL (not more than 10 ppm).

(6) Arsenic—Prepare the test solution with 1.0 g of Dibasic Sodium Phosphate according to Method 1, and perform the test using Apparatus B (not more than 2 ppm).

Loss on drying 57.0 – 61.0% (10 g, at 40°C for 3 hours at first and then at 105°C for 5 hours).

Assay Dissolve about 3 g of Dibasic Sodium Phosphate, previously dried and accurately weighed, in 50 mL of water. Titrate it with 0.5 mol/L sulfuric acid VS at 15°C until the green color of the solution changes to dark-greenish red-purple (indicator: 3 to 4 drops of methyl orange-xylenecyanol FF TS).

Each mL of 0.5 mol/L sulfuric acid VS
= 141.96 mg of Na_2HPO_4

Containers and storage Containers—Tight containers.

Digenea

Digenea

マクリ

Digenea is the whole algae of *Digenea simplex* C. Agardh (*Rhodomelaceae*).

Description Rounded, string-like algae, 2 – 3 mm in diameter; externally, dark red-purple to dark grayish red or grayish brown; a few branched rods irregularly forked, covered with short hairy twigs; calcified weeds and other small algae often attached. Odor, seaweed-like; taste, disagreeable and slightly salty.

Identification To 5 g of Digenea add 50 mL of water, macerate between 50°C and 60°C for 1 hour, and filter while warm. Add 50 mL of water to the residue, macerate again between 50°C and 60°C for 1 hour, and filter while warm. Evaporate all the filtrate on a water bath to about 25 mL, and use this solution as the sample solution. Separately, dissolve 0.05 g of kainic acid in 10 mL of water, and use this solution as the standard solution. Perform the test with these solutions as directed under the Thin-layer Chromatography. Spot 20 μL each of the sample solution and the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with the upper layer of a mixture of water, 1-butanol and acetic acid (100) (5:4:1) to a distance of about 10 cm, and air-dry the plate. Spray evenly a water-saturated solution of ninhydrin in 1-butanol (1 in 500) upon the plate, and heat at 90°C for 10 minutes: the spots obtained from the sample solution and the standard solution show a light yellow color and the same *Rf* values.

Purity Foreign matter—The amount of other algae in Digenea does not exceed 20.0%.

Loss on drying Not more than 22.0%.

Acid-insoluble ash Not more than 8.0%.

Digitalis

Digitalis

ジギタリス

Digitalis is the leaf of *Digitalis purpurea* Linné (*Scrophulariaceae*), dried at a temperature not exceeding 60°C, with petiole and midrib removed, and finely cut.

It contains not less than 8 and not more than 15 Digitalis Units in 1 g.

Description Digitalis occurs as grayish green to grayish yellow-green, fine fragments of thin lamina. The upper surface is glabrous and dented along the vein, and the lower surface is generally densely pubescent and protruded along the vein. It has a slight odor, and a very bitter taste.

Under a microscope, Digitalis reveals upper epidermal cells possessing almost linear or slightly wavy walls, and rarely stomata; lower epidermal cells with prominently wavy walls, and numerous stomata, each being accompanied by 3 to 4 subsidiary cells; the acute multicelluar hairs of 2 to 8 cells and glandular hairs are on both surfaces but more prominently along the veins on the lower surface; crystals of calcium oxalate are absent in the tissues.

Identification To 1 g of Digitalis add 10 mL of diluted ethanol (7 in 10), boil for 2 minutes, and filter. To 5 mL of the filtrate add 10 mL of water and 0.5 mL of lead subacetate TS, shake, and filter. To the filtrate add 5 mL of chloroform, shake, and separate the chloroform layer. Evaporate the chloroform gently on a water bath. Cool, add 1 mL of a solution of iron (III) chloride hexahydrate in acetic acid (100) (1 in 10000) to the residue, shake well, and underlay gently with 1 mL of sulfuric acid: at the zone of contact of the two liquids

a red-brown ring is produced, and the upper layer near the zone of contact gradually changes to dark green, and finally to a dark color.

Purity Foreign matter—The amount of browned leaves, petiole, midrib and other foreign matter contained in Digitalis does not exceed 2.0%.

Loss on drying Not more than 5.0%.

Total ash Not more than 12.0%.

Acid-insoluble ash Not more than 5.0%.

Assay (i) Animals—Select healthy adult pigeons, the heaviest of which weighs less than twice the mass of the lightest. Divide the selected pigeons into two groups, each consisting of more than six pigeons which are as nearly alike as practicable with respect to sex and mass, so that average mass of the group receiving the standard solution does not differ by more than 30% from the average mass of the group receiving the sample solution. Withhold food but allow water for 16 to 28 hours prior to the test.

(ii) Standard stock solution—Transfer about 1 g of Digitalis Reference Standard, accurately weighed, to a 50-mL glass-stoppered, hard-glass bottle, and add 10 mL of diluted ethanol (4 in 5) per 1 g of the standard. Insert the stopper, the upper one-third of which is greased lightly with petrolatum. Shake the mixture between 20°C and 30°C for 24 hours by mechanical means which continuously brings the solid material into fresh contact with the liquid phase. Promptly centrifuge the mixture, decant the supernatant liquid into a suitable container, and use this solution as the standard stock solution. Preserve this solution between 1°C and 10°C before use, and use within 30 days.

(iii) Standard solution—On the day of the assay, dilute a portion of the standard stock solution with isotonic sodium chloride solution so that the estimated fatal dose per kg of body mass of the animals will be about 15 mL.

(iv) Sample stock solution—Transfer about 1 g of Digitalis, reduced to a fine powder and accurately weighed, to a 50-mL glass-stoppered, hard-glass bottle, and proceed as directed in the standard stock solution. Use this solution as the sample stock solution. Preserve this solution between 1°C and 10°C, and use within 30 days.

(v) Sample solution—On the day of the assay, dilute a portion of the sample stock solution with isotonic sodium chloride solution so that the estimated fatal dose per kg of body mass of the animals will be about 15 mL.

(vi) Procedure—Fix the animals, and, if necessary, lightly anesthetize with diethyl ether. Expose an alar vein, and cannulate with a suitable cannula, previously filled with isotonic sodium chloride solution. Place the standard solution and the sample solution in a burette calibrated to 0.05 mL, and connect it with the cannula using a vinyl tube. Inject the standard solution and the sample solution from the burette after ensuring the absence of air bubbles in the burette, vinyl tubes and cannula, or inject them using an injection syringe calibrated to 0.01 mL through the cannula. The injection volume of the standard solution and the sample solution is equivalent to 1 mL per kg of body mass of the animals, and the duration of the injection is 2 to 3 seconds. Repeat this dose at 5-minute intervals thereafter until the animals die of cardiac arrest. If the average number of doses for any given solution required to produce death is less than 13 or more than 19, or if the larger average exceeds the smaller in the same assay by more than 4 doses, regard these data as preliminary. Use them as a guide, and repeat with a fresh, higher or lower dilution.

(vii) Calculation—Designate the number of animals in the groups injected with the standard solution and the sample solution as N_S and N_T, respectively. Designate the total number of doses required to produce death in each group as Y_S and Y_T, respectively, and designate their average values as $\overline{Y}_S$ and $\overline{Y}_T$, respectively.

$$\text{Units in each g of Digitalis} = \frac{\text{units in each mL of the standard solution}}{\text{number of g of Digitalis in each mL of the sample solution}} \times \frac{\overline{Y}_S}{\overline{Y}_T}$$

Compute L (P = 0.95) by using following equation: L should be not more than 0.30. If it exceeds 0.30, repeat the assay by increasing the number of animals or improving the assay conditions, until L becomes not more than 0.30.

$$L = 2\sqrt{(C-1)\left[C \times \left(\frac{\overline{Y}_S}{\overline{Y}_T}\right)^2 + \frac{N_T}{N_S}\right]}$$

$$C = \frac{\overline{Y}_T^2}{\overline{Y}_T^2 - \frac{s^2 t^2}{N_T}}$$

$$s^2 = \frac{\Sigma y^2 - \frac{Y_S^2}{N_S} - \frac{Y_T^2}{N_T}}{n}$$

Σy^2: The sum of values which are obtained by squaring separately the number of injection for the standard solution and the sample solution.

$n = N_S + N_T - 2$

t^2: Value shown in the following table against n for which s^2 is calculated.

n	$t^2 = F_1$	n	$t^2 = F_1$	n	$t^2 = F_1$
1	161.45	13	4.667	25	4.242
2	18.51	14	4.600	26	4.225
3	10.129	15	4.543	27	4.210
4	7.709	16	4.494	28	4.196
5	6.608	17	4.451	29	4.183
6	5.987	18	4.414	30	4.171
7	5.591	19	4.381	40	4.085
8	5.318	20	4.351	60	4.001
9	5.117	21	4.325	120	3.920
10	4.965	22	4.301	∞	3.841
11	4.844	23	4.279		
12	4.747	24	4.260		

Containers and storage Containers—Tight containers.
Storage—Light-resistant.