(4) Benzene—Warm 5 drops of Petroleum Benzin with 2 mL of sulfuric acid and 0.5 mL of nitric acid for about 10 minutes, allow to stand for 30 minutes, transfer the mixture to a porcelain dish, and dilute with water: no odor of nitrobenzene is perceptible.

(5) Residue on evaporation—Evaporate 140 mL of Petroleum Benzin on a water bath to dryness, and heat the residue at 105°C to constant mass: the mass is not more than 1 mg.

(6) Readily carbonizable substances—Shake vigorously 5 mL of Petroleum Benzin with 5 mL of sulfuric acid for readily carbonizable substances for 5 minutes in a Nessler tube, and allow to stand: the sulfuric acid layer has no more color than Matching Fluid A.

Distilling range 50 - 80°C, not less than 90 vol%.

Containers and storage Containers—Tight containers.
Storage—Remote from fire, and not exceeding 30°C.

Pharbitis Seed

Pharbitidis Semen

ケンゴシ

Pharbitis Seed is the seed of *Pharbitis nil* Choisy (*Convolvulaceae*).

Description Longitudinally quartered or sexpartite globe, 6 - 8 mm in length, 3 - 5 mm in width; externally black to grayish red-brown or grayish white, smooth, but slightly shrunken and coarsely wrinkled. The transverse section almost fan-shaped, light yellow-brown to light grayish brown, and dense in texture. Under a magnifying glass, the surface of the seed coat reveals dense, short hairs; dented hilum at the bottom of the ridge. Seed coat thin, the outer layer dark gray, and the inner layer light gray; two irregularly folded cotyledons in the transverse section at one end; two thin membranes from the center of the dorsal side to the ridge separating cotyledons but unrecognizable in the transverse section of the other end having hilum; dark gray secretory pits in the section of the cotyledon. 100 seeds weighing about 4.5 g. When cracked, odor, slight; taste, oily and slightly pungent.

Total ash Not more than 6.0%.

Phellodendron Bark

Phellodendri Cortex

オウバク

Phellodendron Bark is the bark of *Phellodendron amurense* Ruprecht or *Phellodendron chinense* Schneider (*Rutaceae*), from which the periderm has been removed.

It contains not less than 1.2% of berberine [as berberine chloride ($C_{20}H_{18}ClNO_4$: 371.81)], calculated on the basis of dried material.

Description Flat or rolled semi-tubular pieces of bark, 2 - 4 mm in thickness; externally grayish yellow-brown to grayish brown, with numerous traces of lenticel; the internal surface yellow to dark yellow-brown in color, with fine vertical lines, and smooth; fractured surface fibrous and bright yellow. Under a magnifying glass, the transverse section of Phellodendron Bark reveals a thin and yellow outer cortex, scattered with stone cells appearing as yellow-brown dots; inner cortex thick; primary medullary rays expanding its width towards the outer end, the phloem appearing as a nearly triangular part between these medullary rays in secondary cortex, and many secondary medullary rays radiating and gathering to the tip of the triangle; brown phloem fiber bundles lined in tangential direction, crossed over the secondary medullary rays, and then these tissues show a latticework. Odor, slight; taste, extremely bitter; mucilaginous; it colors the saliva yellow on chewing.

Identification (1) To 1 g of pulverized Phellodendron Bark add 10 mL of diethyl ether, allow to stand for 10 minutes with occasional shaking, and filter to remove the diethyl ether. Collect the powder on the filter paper, add 10 mL of ethanol (95), allow to stand for 10 minutes with occasional shaking, and filter. To 2 to 3 drops of the filtrate add 1 mL of hydrochloric acid, add 1 to 2 drops of hydrogen peroxide TS, and shake: a red-purple color develops.

(2) Use the filtrate obtained in (1) as the sample solution. Separately, dissolve 1 mg of berberine chloride for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under the Thin-layer Chromatography. Spot 5 μL each of the sample solution and the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of 1-butanol, water and acetic acid (100) (7:2:1) to a distance of about 10 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one spot among the spots from the sample solution and a spot with yellow to yellow-green fluorescence from the standard solution show the same color tone and the same Rf value.

(3) Stir up pulverized Phellodendron Bark with water: the solution becomes gelatinous owing to mucilage.

Loss on drying Not more than 9.0% (60°C, 8 hours).

Total ash Not more than 7.5%.

Acid-insoluble ash Not more than 0.5%.

Assay Weigh accurately about 0.5 g of pulverized Phellodendron Bark, add 30 mL of a mixture of methanol and dilute hydrochloric acid (100:1), heat under a reflux condenser on a water bath for 30 minutes, cool, and filter. Repeat the above procedure twice with the residue, using 30-mL and 20-mL portions of a mixture of methanol and dilute hydrochloric acid (100:1). To the last residue add 10 mL of methanol, shake well, and filter. Combine the whole filtrates, add methanol to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 0.01 g of Berberine Chloride Reference Standard (separately determine the water content), dissolve in methanol to make exactly 100 mL, and use this solution as the standard solution. Perform the test with 20 μL each of the sample solution and the standard solution as directed under the Liquid Chromatography according to the following conditions, and determine the peak areas, A_T and A_S, of berberine in each solu-