

rapidly changes through purple and blue to green.

(2) Determine the infrared absorption spectrum of Cholecalciferol as directed in the potassium bromide disk method under the Infrared Spectrophotometry, and compare the spectrum with the Reference Spectrum or the spectrum of Cholecalciferol Reference Standard: both spectra exhibit similar intensities of absorption at the same wave numbers.

**Absorbance**  $E_{1\text{cm}}^{1\%}$  (265 nm): 450 – 490 (0.01 g, ethanol (95), 1000 mL).

**Optical rotation**  $[\alpha]_D^{20} + 103 - +112^\circ$  (0.05 g, ethanol (95), 10 mL, 100 mm). Prepare the solution without delay, using Cholecalciferol from a container opened not longer than 30 minutes, previously, and determine the rotation within 30 minutes after the solution has been prepared.

**Purity** 7-Dehydrocholesterol—Dissolve 0.010 g of Cholecalciferol in 2.0 mL of diluted ethanol (95) (9 in 10), add a solution prepared by dissolving 0.020 g of digitonin in 2.0 mL of diluted ethanol (95) (9 in 10), and allow the mixture to stand for 18 hours: no precipitate is formed.

**Assay** Dissolve separately about 0.03 g each of Cholecalciferol and Cholecalciferol Reference Standard, accurately weighed, in isoctane to make exactly 50 mL. Pipet 10 mL each of these solutions, add 3 mL each of the internal standard solution, then add the mobile phase to make 50 mL, and use these solutions as the sample solution and the standard solution. Perform the test with 10  $\mu\text{L}$  each of the sample solution and the standard solution as directed under the Liquid Chromatography according to the following conditions, and calculate the ratios,  $Q_T$  and  $Q_S$ , of the peak area of cholecalciferol to that of the internal standard. Proceed with the operation avoiding contact with air or other oxidizing agents and using light-resistant containers.

$$\begin{aligned} \text{Amount (mg) of } C_{27}H_{44}O \\ &= \text{amount (mg) of Cholecalciferol Reference Standard} \\ &\quad \times \frac{Q_T}{Q_S} \end{aligned}$$

**Internal standard solution**—A solution of dimethyl phthalate in isoctane (1 in 100).

**Operating conditions**—

**Detector:** An ultraviolet absorption photometer (wavelength: 254 nm).

**Column:** A stainless steel column about 4 mm in inside diameter and 10 to 30 cm in length, packed with silica gel for liquid chromatography (5 to 10  $\mu\text{m}$  in particle diameter).

**Column temperature:** Ordinary temperature.

**Mobile phase:** A mixture of hexane and *n*-amylalcohol (997:3).

**Flow rate:** Adjust the flow rate so that the retention time of cholecalciferol is about 25 minutes.

**Selection of column:** Dissolve 0.015 g of Cholecalciferol Reference Standard in 25 mL of isoctane. Transfer this solution to a flask, heat under a reflux condenser in an oil bath for 2 hours, and cool to room temperature rapidly. Transfer this solution to a quartz test tube, and irradiate under a short-wave lamp (main wavelength: 254 nm) and a long-wave lamp (main wavelength: 365 nm) for 3 hours. To this solution add the mobile phase to make 50 mL. Proceed with 10  $\mu\text{L}$  of this solution under the above operating conditions. Use a column with the ratios of the retention time of pre-*vitamin*

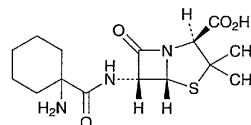
*min* D<sub>3</sub>, *trans*-*vitamin* D<sub>3</sub> and *tachysterol*<sub>3</sub> to that of *cholecalciferol* being about 0.5, about 0.6 and about 1.1, respectively, and with resolution between *previtamin* D<sub>3</sub> and *trans*-*vitamin* D<sub>3</sub>, and that between *cholecalciferol* and *tachysterol*<sub>3</sub> being not less than 1.0.

**Containers and storage** Containers—Hermetic containers.

Storage—Light-resistant, under nitrogen atmosphere, and in a cold place.

## Ciclacillin

シクラシリン



$C_{15}H_{23}N_3O_4S$ : 341.43  
(2*S*,5*R*,6*R*)-6-[(1-Aminocyclohexanecarbonyl)amino]-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0]heptane-2-carboxylic acid [3485-14-1]

Ciclacillin conforms to the requirements of Ciclacillin in the Requirements for Antibiotic Products of Japan.

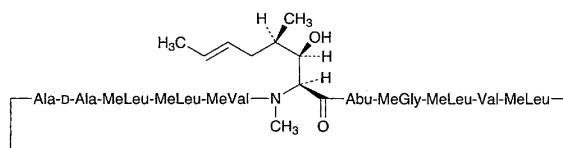
**Description** Ciclacillin occurs as a white to light yellowish white, crystalline powder.

It is sparingly soluble in water, slightly soluble in methanol, very slightly soluble in ethanol (95), and practically insoluble in diethyl ether.

## Ciclosporin

### Ciclosporin A

シクロスポリン



Abu = (2*S*)-2- Abu = (2*S*)-2-Aminobutyric acid

MeGly = *N*- MeGly = *N*-Methylglycine

MeLeu = *N*- MeLeu = *N*-Methylleucine

MeVal = *N*- MeVal = *N*-Methylvaline

$C_{62}H_{111}N_{11}O_{12}$ : 1202.61  
*cyclo*-{-(2*S*,3*R*,4*R*,6*E*)-3-Hydroxy-4-methyl-2-methylamino-6-enoylethyl-L-2-aminobutanoyl-N-methylglycyl-N-methyl-L-leucyl-L-valyl-N-methyl-L-leucyl-L-alanyl-D-alanyl-N-methyl-L-leucyl-N-methyl-L-leucyl-N-methyl-L-valyl-} [59865-13-3]

Ciclosporin contains not less than 98.5% and not more than 101.5% of  $C_{62}H_{111}N_{11}O_{12}$ , calculated on