AMENDMENTS TO THP 2021 VOLUME I AND THP 2021 SUPPLEMENT 2024

MONOGRAPHS

บัวบก (BUABOK)

[pp. 29–37 (THP 2021 VOLUME I), pp. XXIX–XXXVII (THP 2021 SUPPLEMENT 2024)]

Replace with the following:

บัวบก, ส่วนเหนือดิน (BUABOK, SUAN NUEA DIN)

ผักหนอก, ส่วนเหนือดิน (PHAK NOK, SUAN NUEA DIN)
Centellae Asiaticae Herba
Centella

Synonyms Asiatic Pennywort, Gotu Kola, Indian Pennywort, Indian Water Navelwort

Category Mild diuretic, anti-inflammatory, wound healing (topical).

Centella is the dried aerial part of *Centella asiatica* (L.) Urb. (*C. coriacea* Nannf., *Hydrocotyle asiatica* L., *H. lunata* Lam., *Trisanthus cochinchinensis* Lour.) (Family Umbelliferae), Herbarium Specimen Number: DMSC 1461, Crude Drug Number: DMSc 1261.

Constituents Centella contains triterpenoid saponins, including asiaticoside and madecassoside and their aglycones which are asiatic acid and madecassic acid, respectively. It also contains volatile oil, pectin, trace of alkaloids, etc.

Description of the plant (Figs. 1a, 1b) Slender trailing herb, rooting at nodes. Leaves simple, 1 to 6 in rosette at each node, orbicular to reniform, more or less cupped, glabrous and shiny above, paler beneath, 1 to 7 cm in diameter, apex rounded, base cordate, margin entire, crenate, or usually repand-dentate; petiole (1–)4 to 10(–50) cm long. Inflorescence in single umbel, bearing solitary or 2 to 5 together in the axils; peduncle shorter than petiole. Flowers usually 3, middle one sessile, lateral ones pedicellate; involucres 2, ovate; petals 5, minute, white or rose-tinged; ovary laterally flattened, style filiform. Fruit small, compressed, about 8 mm long, orbicular to ellipsoid, manifestly ribbed, slightly hairy when young.

Description Odour, characteristic; taste, slightly bitter-sweet.

Macroscopical (Fig. 1a) Aerial part, greenish brown, rough and brittle; stem thin, long, twisted; leaves rennate or cordate, brittle; petiole long.

Microscopical (Figs. 2a–2d) Transverse section of the fresh leaf shows upper epidermis, a layer of rectangular cells, polygonal and straight-walled in surface view; stomata, anisocytic, some paracytic and rarely anomocytic. Palisade cells, a layer of large columnar cells. Spongy cells, parenchymatous, some containing calcium oxalate crystals in the forms of rosette aggregate or prism. Collenchyma, occurring beneath upper and lower epidermises in the midrib. Vascular bundles, xylem in the upper part and phloem in the lower part; vessels, annular, spiral, scalariform, or reticulate. Lower epidermis, a layer of rectangular cells, slightly wavy-walled in surface view; stomata, anisocytic, paracytic, or anomocytic. Oil ducts, occurring beneath collenchyma in the middle of midrib.

Transverse sections of the fresh petiole and stolon show epidermal layer with cuticle. Collenchyma, present. Parenchyma containing chloroplastids, oil droplets, spreading circularly beneath collenchyma. Vascular bundles, collateral. The centre of petiole, hollow. Unicellular trichomes may also be found, but rare, in the section near the base of petiole.

Centella in powder possesses the diagnostic microscopical characters of the unground drug.

1

2

3

1 cm

4

5

Fig. 1a *Centella asiatica* (L.) Urb.
1. habit 2. leaves 3. flowers and fruits 4. inflorescence 5. leaves, flowers and fruits

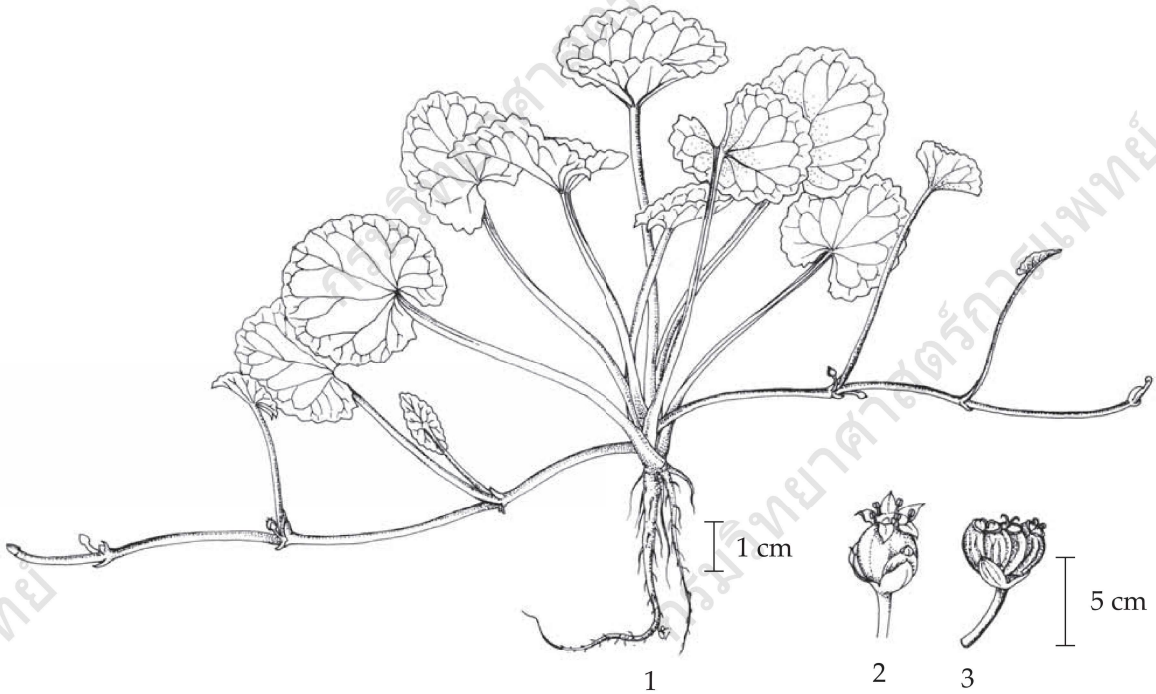
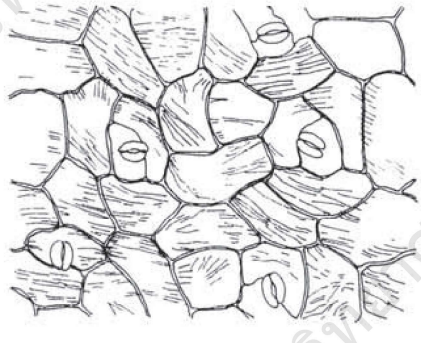
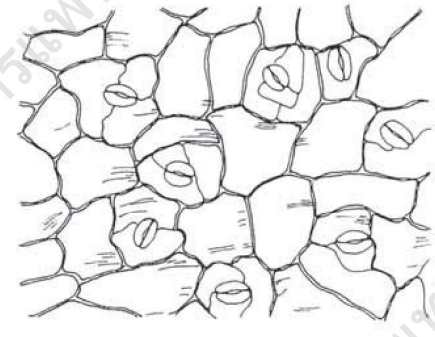


Fig. 1b *Centella asiatica* (L.) Urb.
1. habit 2. inflorescence 3. fruits



50 μm

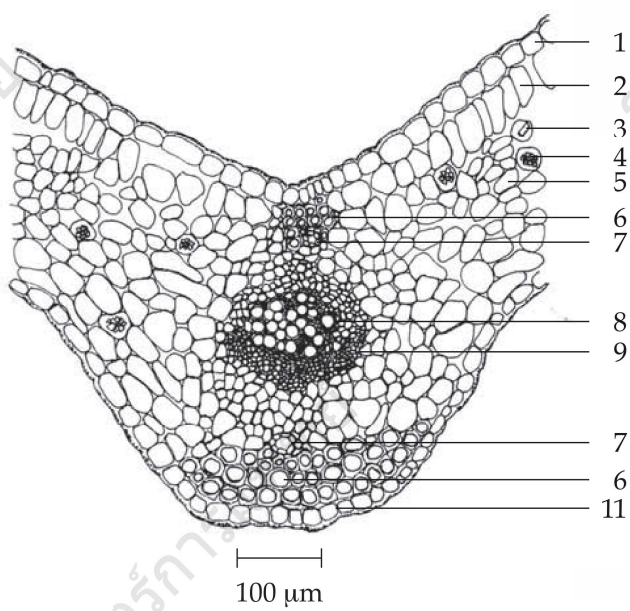
Upper Epidermis of the Lamina



50 μm

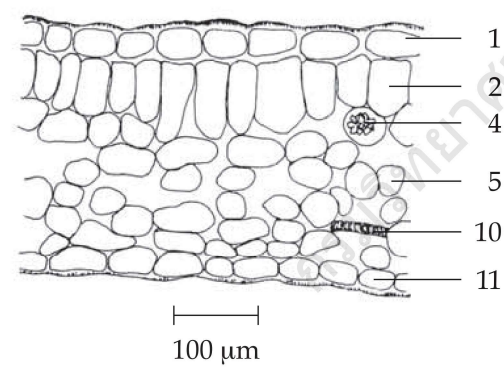
Lower Epidermis of the Lamina

Fig. 2a Line Drawings of Epidermises of the Fresh Leaf of *Centella asiatica* (L.) Urb.



100 μm

Transverse Section of the Midrib

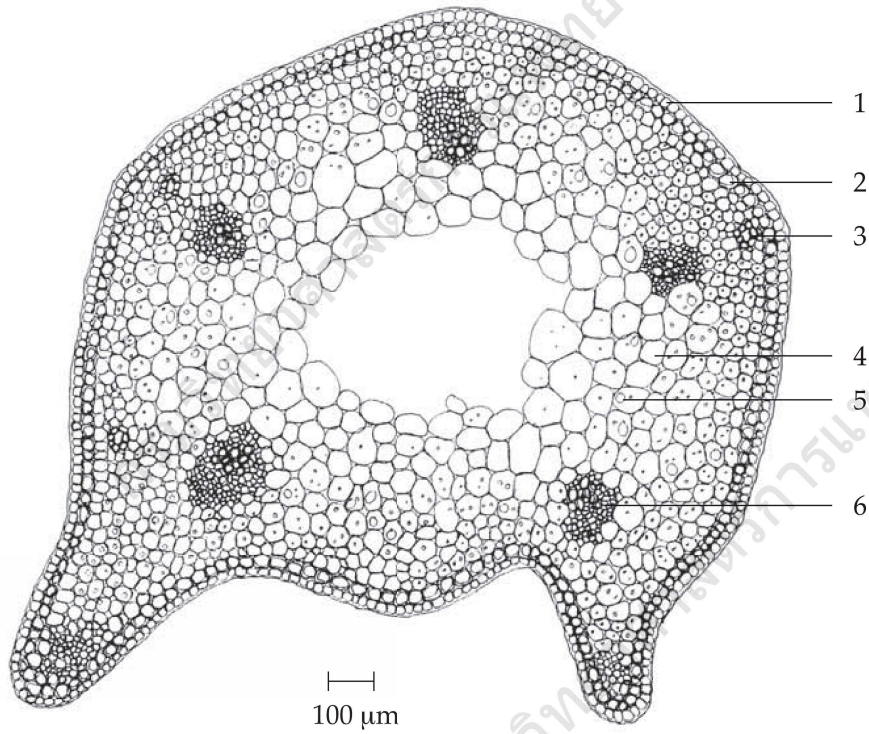


100 μm

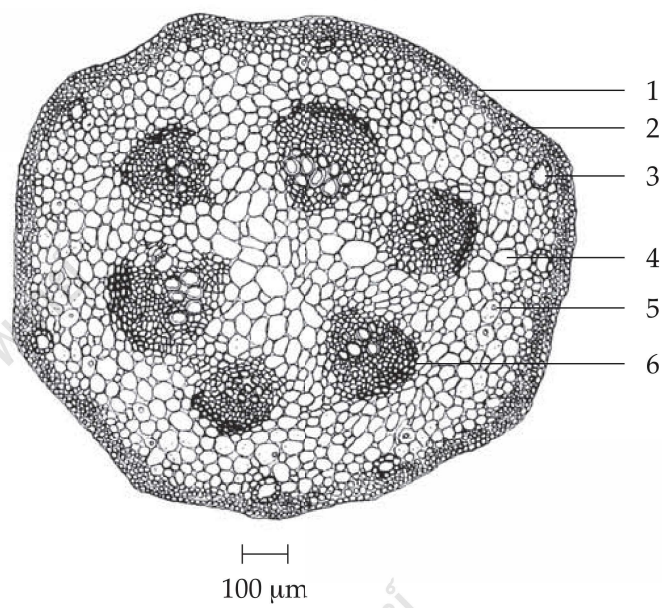
Transverse Section of the Lamina

Fig. 2b Line Drawings of Transverse Sections of the Fresh Leaf of *Centella asiatica* (L.) Urb.

1. upper epidermis
2. palisade cell
3. prismatic crystal
4. rosette aggregate crystal
5. spongy cell
6. collenchyma
7. oil duct
8. xylem
9. phloem
10. vessel
11. lower epidermis



Transverse Section of the Petiole



Transverse Section of the Stolon

Fig. 2c Line Drawings of Transverse Sections of the Fresh Petiole and Stolon of *Centella asiatica* (L.) Urb.

1. epidermis
2. collenchyma
3. oil duct
4. parenchyma
5. oil droplet
6. vascular bundle

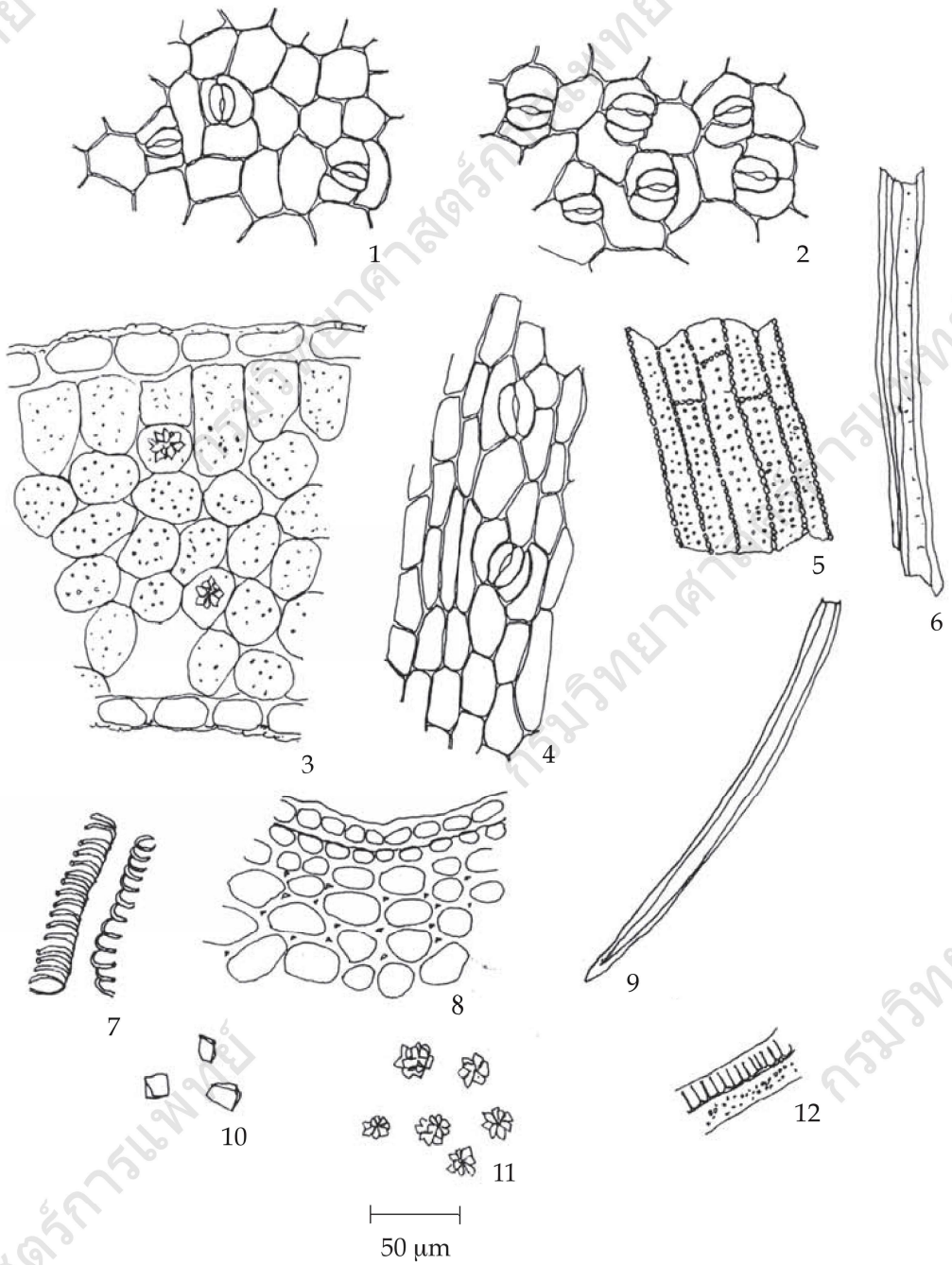


Fig. 2d Line Drawings of Powdered Drug of the Aerial Parts of *Centella asiatica* (L.) Urb.

1. upper epidermis
2. lower epidermis
3. lamina in sectional view
4. epidermis with stomata from petiole
5. pitted vessels
6. fibres
7. spiral vessels
8. epidermis and collenchyma, in sectional view
9. unicellular trichome
10. prismatic crystals
11. rosette aggregate crystals
12. scalariform and pitted vessels

Warning Excessive oral administration should be avoided during pregnancy and lactation.

Packaging and storage Centella shall be kept in well-closed containers, protected from light, and stored in a dry place.

Identification

A. Warm 500 mg of the sample, in powder, with 5 mL of *ethanol* for 5 minutes and filter (solution 1). To 2 mL of solution 1, add a few drops of *sulfuric acid*: a green colour develops.

B. Evaporate 2 mL of solution 1 to dryness and dissolve the residue in 2 mL of *acetic anhydride*. Add slowly 1 mL of *sulfuric acid* to form two layers: a green colour develops in the upper layer and a brownish red ring forms at the zone of contact.

C. Shake vigorously 500 mg of the sample, in powder, with 10 mL of *water*: a long lasting foam is produced.

D. Carry out the test as described in the "Thin-Layer Chromatography" (Appendix 3.1), using a high-performance plate with *silica gel GF254* as the coating substance and a mixture of 60 volumes of *dichloromethane*, 28 volumes of *methanol*, and 12 volumes of *water* as the mobile phase and allowing the solvent front to ascend 8.5 cm above the line of application. Apply separately to the plate as bands of 8 mm, 3 µL each of the following solutions. Prepare solution (A) by refluxing 500 mg of the sample, in powder, with 10 mL of *ethanol* for 10 minutes and filtering. Evaporate the filtrate under reduced pressure at 40° until dry and dissolve the residue in 4 mL of *ethanol*. For solution (B), dissolve 1 mg of *madecassoside* in 1 mL of *ethanol*. For solution (C), dissolve 1 mg of *asiaticoside* in 1 mL of *ethanol*. After removal of the plate, allow it to dry in air. Spray the plate with *anisaldehyde TS* and heat at 105° for 3 minutes. The chromatogram obtained from solution (A) shows a yellowish brown band (hR_f value 12 to 18) and a blue band (hR_f value 20 to 26) corresponding to the madecassoside and asiaticoside bands from solutions (B) and (C), respectively. Ten violet, two yellowish brown, and one blue bands are also observed (Fig. 3).

E. The chromatogram of the Sample preparation shows several peaks, two of which correspond to the asiaticoside and madecassoside peaks of the Standard preparations, as obtained in the *Contents of asiaticoside and madecassoside*.

Loss on drying Not more than 14.0 per cent w/w after drying at 105° to constant weight (Appendix 4.15).

Foreign matter Not more than 2.0 per cent w/w (Appendix 7.2).

Acid-insoluble ash Not more than 7.0 per cent w/w (Appendix 7.6).

Total ash Not more than 17.0 per cent w/w (Appendix 7.7).

Ethanol-soluble extractive Not less than 15.0 per cent w/w (Appendix 7.12).

Water-soluble extractive Not less than 24.0 per cent w/w (Appendix 7.12).

Contents of asiaticoside and madecassoside Not less than 3.0 per cent w/w for the sum of asiaticoside and madecassoside, calculated on the dried basis. Carry out the determination as described in the "Liquid Chromatography" (Appendix 3.5).

Mobile phase A Use *acetonitrile*.

Mobile phase B Use *water*.

Standard preparation A Dissolve a suitable quantity of *asiaticoside*, accurately weighed, in sufficient *methanol* to obtain a stock solution having a known concentration of about 250 μg of asiaticoside per mL. Dilute the solution quantitatively and stepwise with the same solvent to obtain six solutions having known concentrations ranging from 10 to 60 μg per mL.

Standard preparation B Dissolve a suitable quantity of *madecassoside*, accurately weighed, in sufficient *methanol* to obtain a stock solution having a known concentration of about 300 μg of madecassoside per mL. Dilute the solution quantitatively and stepwise with the same solvent to obtain six solutions having known concentrations ranging from 30 to 180 μg per mL.

Sample preparation Transfer about 200 mg of Centella, in *coarse powder*, accurately weighed, into a 50-mL round-bottomed flask and add 25 mL of *methanol*. Heat under a reflux condenser for 1 hour, filter into a 50-mL volumetric flask, and add the same solvent to volume. Filter through a membrane having a 0.22-μm porosity.

Chromatographic system The chromatographic procedure may be carried out using (a) a stainless steel column (5 cm × 2.1 mm) packed with octadecylsilane chemically bonded to porous silica or ceramic microparticles (1.7 μm), (b) *Mobile phase* at a flow rate of about 0.6 mL per minute, and (c) an ultraviolet photometer set at 205 nm.

The step gradient of mobile phases is as follows:

Time (Minutes)	*Mobile Phase A* (Per Cent V/V)	*Mobile Phase B* (Per Cent V/V)
0	15	85
1.5	60	40
2	0	100
3	0	100
4	15	85

To determine the suitability of the chromatographic system, chromatograph *Standard preparation A* and *Standard preparation B* having known concentrations of 30 μg per mL of asiaticoside and 90 μg per mL of madecassoside, respectively, and record the peak responses as directed under *Procedure* and *Calculation*: the relative standard deviation for replicate injections is not more than 2.0 per cent.

Procedure Separately inject equal volumes (about 4 μL) of *Standard preparation A* and *Standard preparation B* into the chromatograph, record the chromatograms, and measure the responses for asiaticoside and madecassoside peaks. Plot the readings and draw the standard curves of best fit: the curves show the correlation coefficient of not less than 0.999. Inject about 4 μL of *Sample preparation* into the chromatograph, record the chromatogram, and measure the responses for asiaticoside and madecassoside peaks.

Calculation By reference to the standard curves, calculate the sum of asiaticoside ($C_{48}H_{78}O_{19}$) and madecassoside ($C_{48}H_{78}O_{20}$) contents, in the portion of the Centella taken.

Dose 0.6 g three times a day.

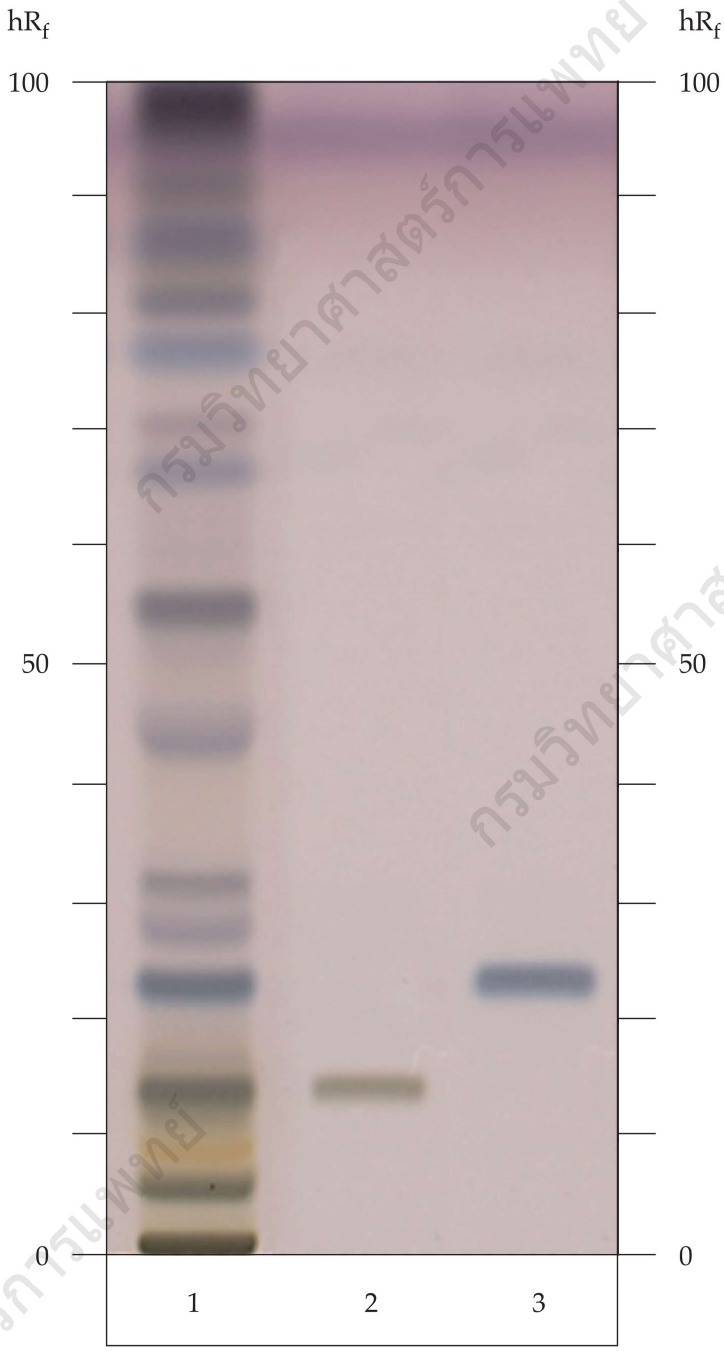


Fig. 3 Thin-Layer Chromatogram of Ethanolic Extract of the Aerial Parts of *Centella asiatica* (L.) Urb., Detected with *Anisaldehyde TS*

1 = solution (A)
2 = solution (B)
3 = solution (C)