

## กระตุกไก่ดำ, ใบ (KRADUK KAI DAM, BAI)

Justiciae Gendarussae Folium

Justicia Gendarussa Leaf

**Category** Anti-inflammatory.

**Justicia Gendarussa Leaf is the dried leaf of *Justicia gendarussa* Burm. f. (*Gendarussa vulgaris* Nee) (Family Acanthaceae), Herbarium Specimen Number: DMSC 5347, Crude Drug Number: DMSc 1260.**

**Constituents** Justicia Gendarussa Leaf contains amino acids such as alanine. It also contains flavonoids (e.g., gendarusins A-E), aromatic amines, triterpenoids, alkaloids, sterols, etc.

**Description of the plant** (Fig. 1) Shrub up to 1.5 m tall; stem erect, much branched, obtusely quadrangular, swollen at nodes, glabrous, purplish to blackish purple. Leaves simple, opposite decussate, narrowly lanceolate, 6 to 14 cm long, 1 to 2.5 cm wide, apex acute to acuminate, base cuneate or attenuate, margin slightly sinuate, glabrous on both sides, midrib prominently blackish red; petiole 1 to 2.5 cm long. Inflorescence spike, terminal or axillary, 3 to 12 cm long; rachis glabrous or sparsely puberulous; peduncle 0.5 to 1.5 cm long; bract leaf-like, triangular, 2 to 6 mm long, 1 to 2.5 mm wide, basal bract longer than calyx, margin ciliate, apex acute; bracteole caducous, elliptic to linear-lanceolate, about 3 mm long, about 1 mm wide, apex acute, margin ciliate. Flower: calyx deeply 5-lobed, subequal, 4 to 7 mm long, lobe linear-lanceolate, 3 to 4 mm long, about 0.5 mm wide, apex acuminate; corolla creamy white to light purple, with purple spots or blotches on corolla lips, bilabiate, 1.2 to 1.6 cm long, glabrous, tube basally cylindric, 8 to 9 mm long, about 2 mm wide, upper lip flat or slightly hooded, triangular-ovate, 5 to 6 mm long, about 3.5 mm wide, apex emarginate, lower lip spreading, 3-lobed, elliptic-ovate, 0.6 to 1 cm long; stamens 2, filament 3 to 6 mm long, attached on upper part of corolla tube, glabrous, anther thecae oblong, about 1.2 mm long, superposed, upper one muticous at base, lower one spurred; ovary superior, 2-loculed, ovules 2 per locule, glabrous, style about 1 cm long, glabrous, stigma capitate, shortly 2-lobed. Fruit a capsule, clavate, 1 to 1.2 cm long. Seeds 2 to 4, dark brownish, orbicular, flattened.

**Description** Odour, mild; taste, slightly bitter.

**Macroscopical** (Fig. 1) Whole or broken leaves; leaf petiolate, yellowish green, lanceolate, glabrous, apex acute to acuminate, base cuneate or attenuate, margin slightly sinuate.

**Microscopical** (Figs. 2a–2d) Transverse section of the leaf through the midrib shows upper epidermis, mesophyll, vascular tissue, and lower epidermis. Upper epidermis: 1 to 3 layers of cells containing purple substances, cuticle layer, unicellular warty-walled trichome, multicellular glandular trichomes, multicellular trichomes, and lithocyst. Mesophyll: 1 to 3 layers of palisade cells, containing starch grains or some containing purple substances; spongy cells, containing starch grains or some containing purple substances; angular collenchyma and parenchyma, containing starch grains or some containing purple substances. Vascular tissue: phloem and xylem. Lower epidermis: layers of rectangular cells, some containing purple substances, cuticle layer, multicellular glandular trichomes, multicellular trichomes, lithocyst, and stomata.

In surface view, upper epidermis shows wavy-walled cells, multicellular glandular trichomes; lower epidermis, slightly wavy-walled cells, diacytic stomata, multicellular glandular trichomes, and lithocyst.



1



2



3



4



5

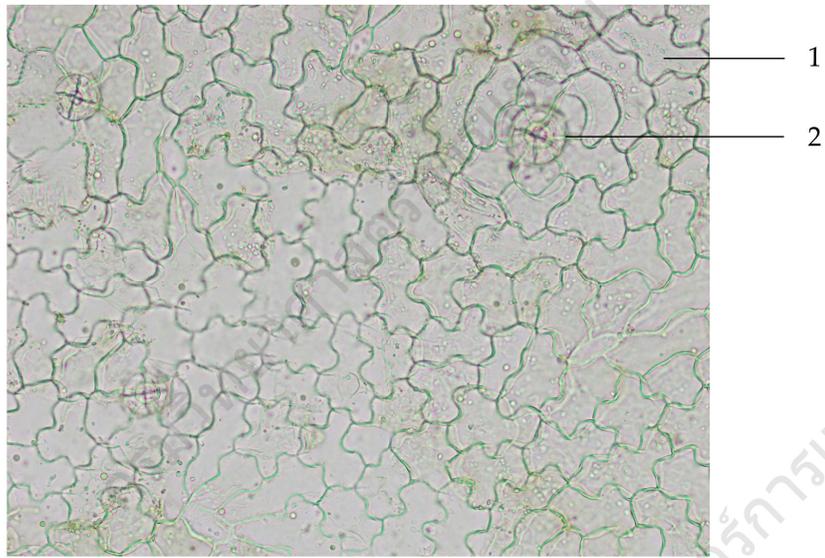


2 cm

6

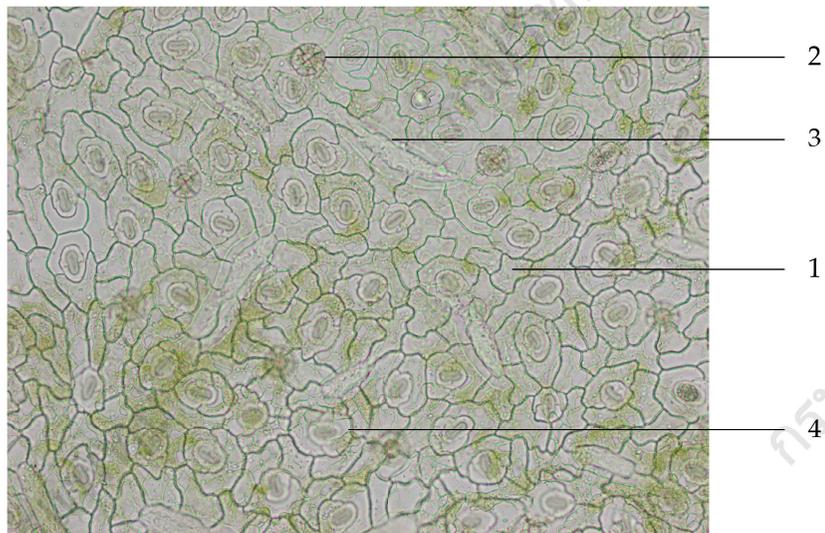
**Fig. 1** *Justicia gendarussa* Burm. f.

1. habit 2. leafy twigs 3. inflorescences 4. flowers 5. fruits and seeds 6. crude drug



100  $\mu$ m

Upper Epidermis of the Lamina

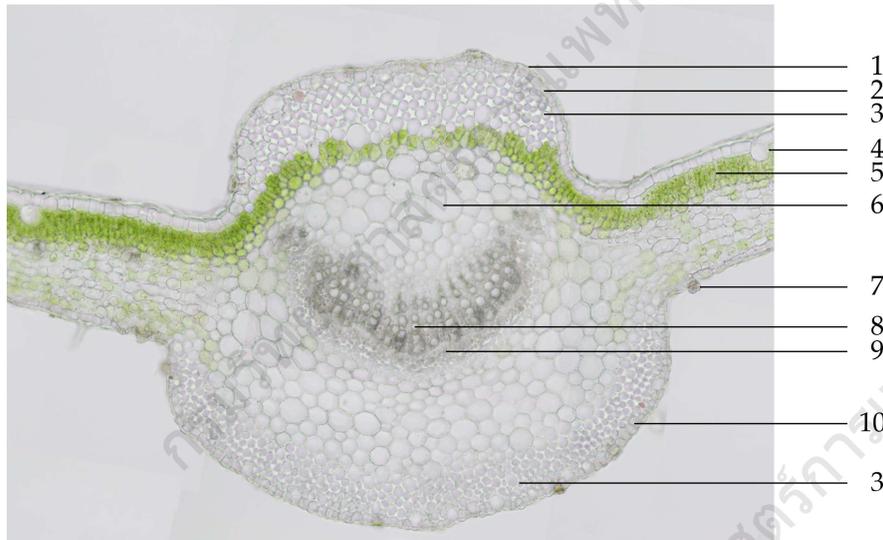


100  $\mu$ m

Lower Epidermis of the Lamina

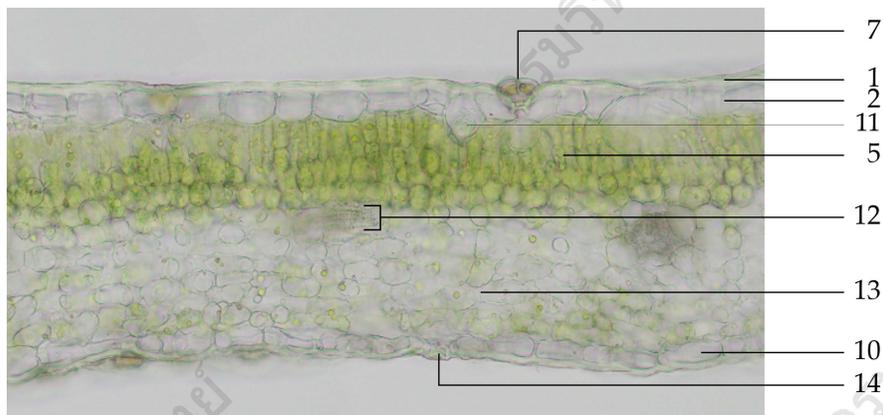
**Fig. 2a** Photomicrographs of Epidermises of the Leaf of *Justicia gendarussa* Burm. f.

- |                       |              |
|-----------------------|--------------|
| 1. epidermal cell     | 3. lithocyst |
| 2. glandular trichome | 4. stoma     |



100 μm

Transverse Section of the Midrib

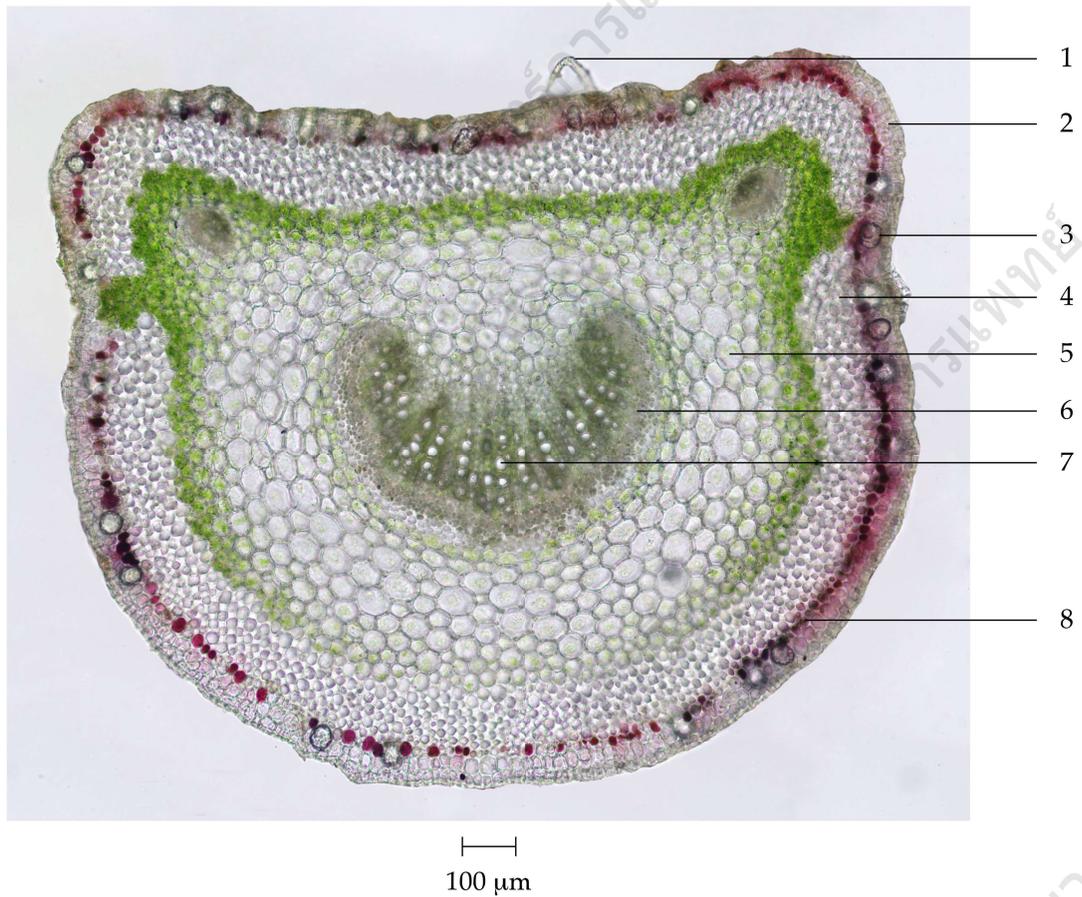


100 μm

Transverse Section of the Lamina

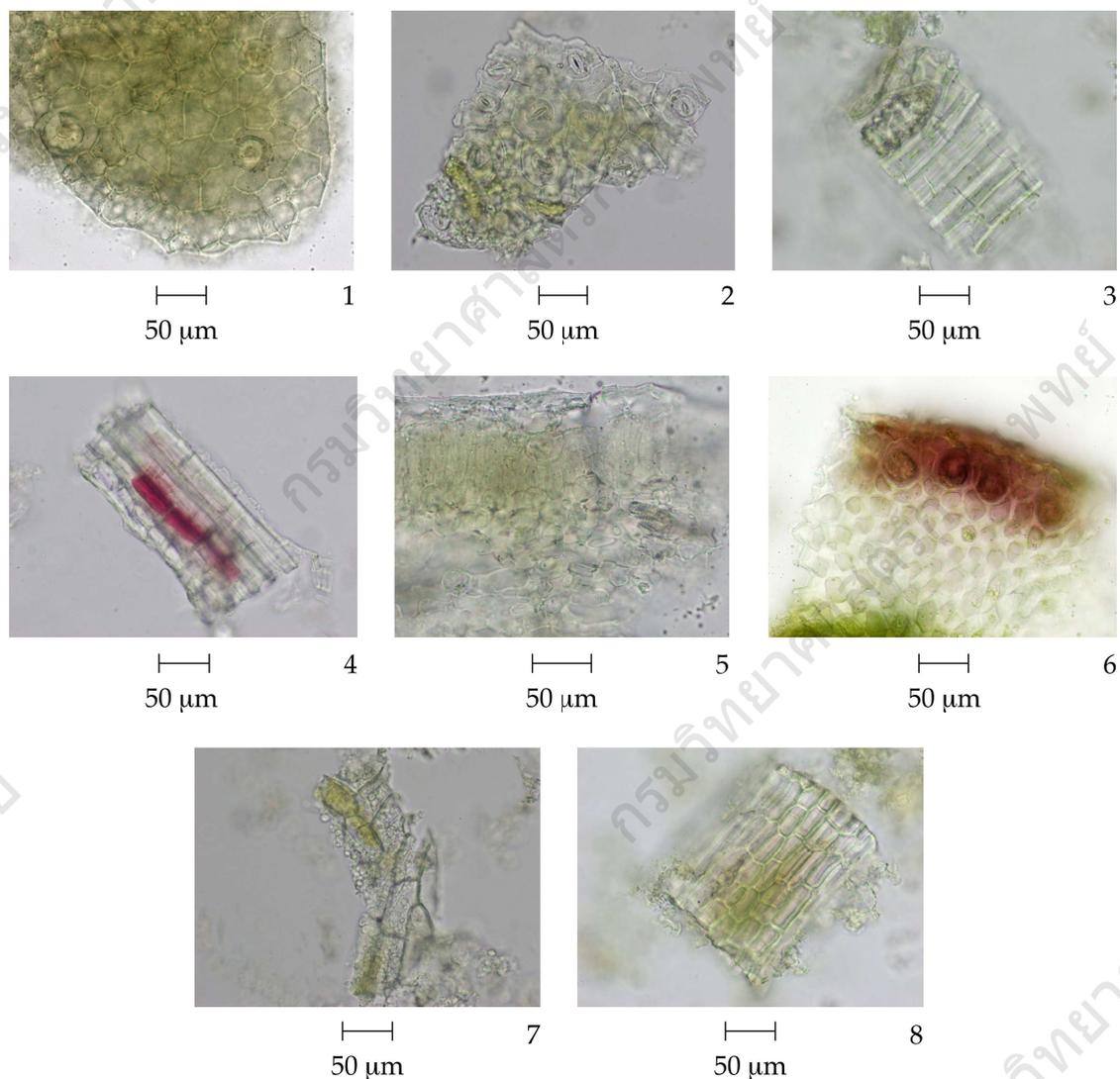
**Fig. 2b** Photomicrographs of Transverse Sections of the Leaf of *Justicia gendarussa* Burm. f.

- |   |  |
|---|--|
| 1. cuticle                                | 8. vessel                                |
| 2. upper epidermis                        | 9. phloem                                |
| 3. collenchyma                            | 10. lower epidermis                      |
| 4. lithocyst                              | 11. cystolith                            |
| 5. palisade cell containing starch grains | 12. vascular tissue                      |
| 6. parenchyma                             | 13. spongy cell containing starch grains |
| 7. multicellular glandular trichome       | 14. stoma                                |



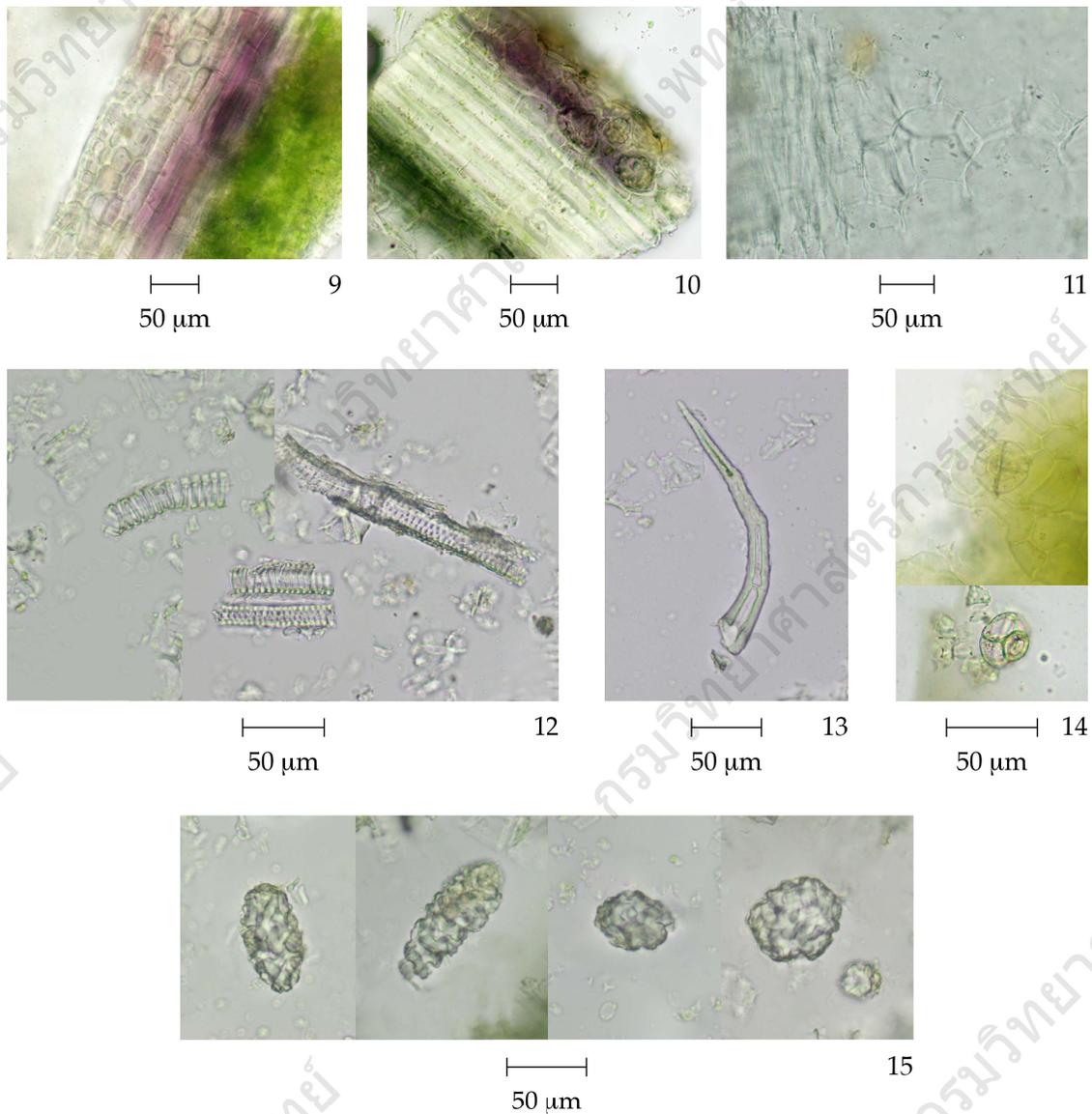
**Fig. 2c** Photomicrograph of Transverse Section of the Petiole of *Justicia gendarussa* Burm. f.

- |                   |                      |
|-------------------|----------------------|
| 1. trichome       | 5. parenchyma        |
| 2. epidermal cell | 6. phloem            |
| 3. lithocyst      | 7. vessel            |
| 4. collenchyma    | 8. purple substances |



**Fig. 2d** Photomicrographs of Powdered Drug of the Leaves of *Justicia gendarussa* Burm. f.

1. upper epidermis, glandular trichome and underlying palisade cells, in surface view
2. lower epidermis with diacytic stomata and lithocysts, in surface view
3. epidermis with lithocyst and collenchyma, in longitudinal view
4. epidermis and collenchyma, some containing purple substances, in longitudinal view
5. lamina, in sectional view, showing upper epidermis, palisade cells, vascular tissues, and spongy cells
6. epidermis, collenchyma, and lithocyst, some containing purple substances, in sectional view
7. parenchyma containing starch grains
8. epidermal cells of petiole in surface view



**Fig. 2d** (continued)

- |  |  |
|--|--|
| <p>9. epidermis and collenchyma, some containing purple substances and chlorenchyma, in longitudinal view</p> <p>10. epidermis, cystoliths, collenchyma, some containing purple substances, and chlorenchyma, in longitudinal view</p> | <p>11. parenchyma containing rod-shaped crystals and fibres</p> <p>12. spiral, reticulate, and pitted vessels</p> <p>13. multicellular trichome</p> <p>14. glandular trichomes</p> <p>15. cystoliths</p> |
|--|--|

Transverse section of the petiole shows epidermis, cortex, and vascular tissue. Epidermis: 1 to 2 layers of epidermal cells, some containing purple substances, cuticle layer, multicellular glandular trichomes, multicellular trichomes, lithocyst, and stomata. Cortex: angular collenchyma, some containing purple substances and parenchyma, some containing purple substances and/or starch grains and/or crystals. Vascular tissue: phloem and xylem.

Justicia Gendarussa Leaf in powder possesses the diagnostic microscopical of the unground drug. The purple substance, although characteristic, is visible only in freshly dried and ground samples. The combination of lithocysts, multicellular glandular trichomes, and collenchyma is also uniquely present.

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

#### Identification

A. Reflux 1 g of the sample, in *fine powder*, with 20 mL of *ethanol* for 15 minutes and filter. To 2 mL of the filtrate, add a few drops of *ninhydrin TS* and warm in a water-bath for about 5 minutes: a violet colour is produced.

B. 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 48 volumes of *butanol*, 26 volumes of *glacial acetic acid*, 16 volumes of *2-propanol*, and 4 volumes of *water* as the mobile phase and allowing the solvent front to ascend 8 cm above the line of application. Apply separately to the plate as bands of 8 mm, 8  $\mu$ L of solution (A) and 1  $\mu$ L of solution (B). Prepare solution (A) by refluxing 1 g of the sample, in *fine powder*, with 30 mL of *ethanol* for 30 minutes and filtering. Evaporate the filtrate to dryness and dissolve the residue in 3 mL of *ethanol*. For solution (B), dissolve 1 mg of *alanine* in 1 mL of *water*, add 9 mL of *ethanol*, and mix. After removal of the plate, allow it to dry in air. Examine the plate under ultraviolet light (254 nm); marking the quenching bands. Subsequently examine the plate under ultraviolet light (366 nm); two red and three blue fluorescent bands are observed. Spray the plate with *ninhydrin TS* and heat at 110° for 10 minutes; the chromatogram obtained from solution (A) shows a pink band ( $hR_f$  value 29 to 35) corresponding to the alanine band from solution (B). Other five pink bands are observed (Fig. 3).

Repeat the same procedure on another plate but omitting solution (B). After removal of the plate, allow it to dry in air. Heat the plate at 80° for 10 minutes and then spray the plate with *natural products (NP) TS* while the plate is still warm. Subsequently spray the plate with *polyethyleneglycol (PEG) TS* and observe the colours of the bands under ultraviolet light (366 nm) within 5 to 15 minutes. One blue and three yellow fluorescent bands are observed (Fig. 3).

**Loss on drying** Not more than 10.0 per cent w/w after drying at 105° to constant weight (Appendix 4.15).

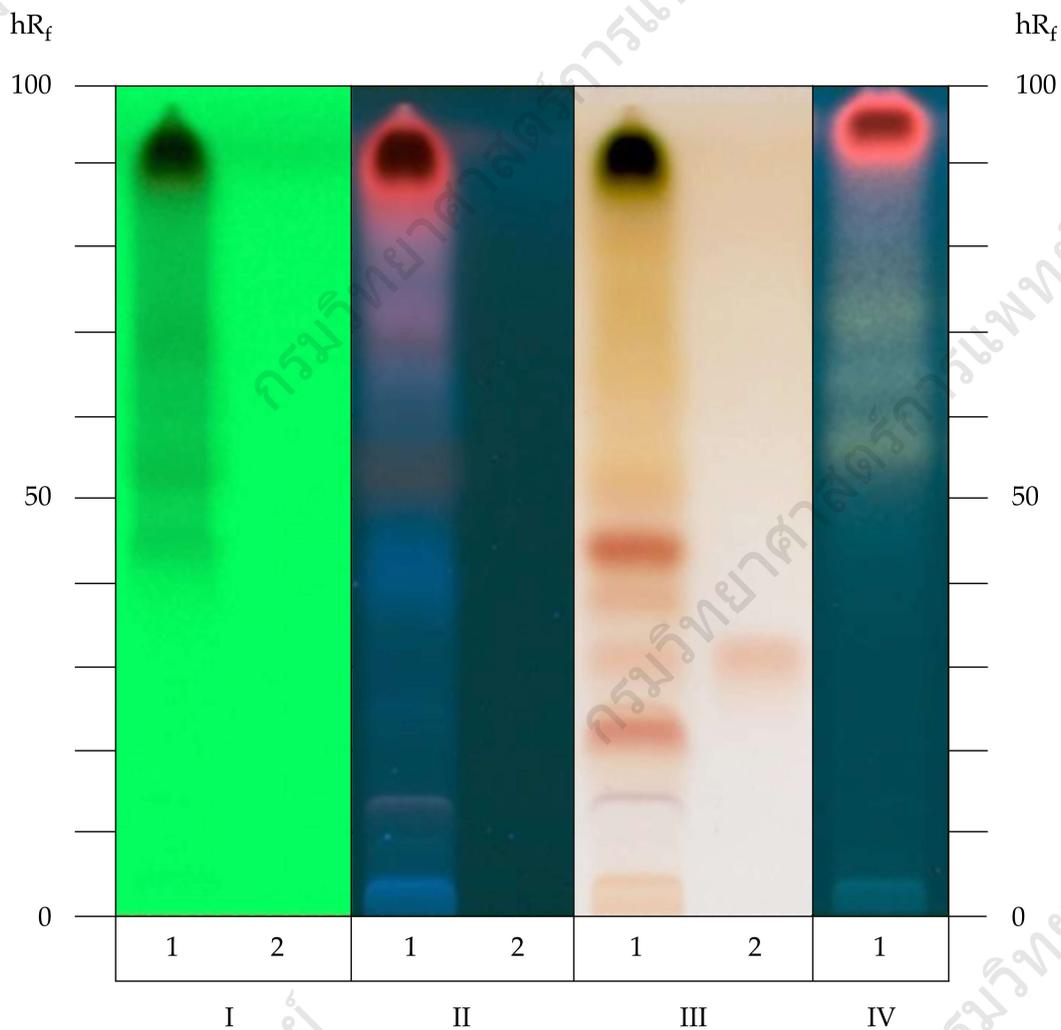
**Foreign matter** Not more than 2.0 per cent w/w (Appendix 7.2).

**Acid-insoluble ash** Not more than 3.0 per cent w/w (Appendix 7.6).

**Total ash** Not more than 15.0 per cent w/w (Appendix 7.7).

**Ethanol-soluble extractive** Not less than 9.0 per cent w/w (Appendix 7.12).

**Water-soluble extractive** Not less than 20.0 per cent w/w (Appendix 7.12).



**Fig. 3** Thin-Layer Chromatogram of Ethanolic Extract of the Leaves of *Justicia gendarussa* Burm. f.

1 = solution A

2 = solution B

I = detection under UV light (254 nm)

II = detection under UV light (366 nm)

III = detection with *ninhydrin* TS

IV = detection under UV light (366 nm) after spraying with *NP/PEG* TS