

# Identification and Detection Limits for Yao Medicinal Material *Gendarussa vulgaris*

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**Abstract** [Objectives] To further revise and enhance the quality standards of the Yao medicinal material *Gendarussa vulgaris*, and to provide empirical data to ensure the safety of *G. vulgaris* medicinal resources and the advancement of medicinal material standards. [Methods] The identification of *G. vulgaris* was conducted through morphological, microscopic, and physicochemical analyses. Parameters such as moisture content, total ash, acid-insoluble ash, extract content, heavy metal concentrations, and organochlorine pesticide residues were determined in accordance with the 2025 edition of the *Chinese Pharmacopoeia*. [Results] The distinguishing characteristics of *G. vulgaris* were clearly evident, allowing for the establishment of an identification method for this species. The thin-layer chromatogram exhibited clear and well-resolved separation. Additionally, the permissible ranges for moisture content, total ash, and water-soluble extract in the medicinal materials were defined, while new limit standards for acid-insoluble ash and heavy metal content were concurrently established. [Conclusions] The test method established in this study is both accurate and reliable, thereby enhancing the quality standards of the medicinal material *G. vulgaris*.

**Key words** *Gendarussa vulgaris*, Quality standard, Identification, Heavy metal, Pesticide residue

## 1 Introduction

Yao medicinal material *Gendarussa vulgaris* Nees., derived from the dried aerial parts of the plant *Gendarussa* Herba belonging to the Acanthaceae family<sup>[1]</sup>, is included in the first volume of the *Quality Standards for Yao Medicinal Materials in the Guangxi Zhuang Autonomous Region*<sup>[2]</sup>. *G. vulgaris* is traditionally recognized for its therapeutic properties, including the promotion of tendon and bone extension, the removal of blood stasis, the facilitation of tissue regeneration, as well as the reduction of swelling and alleviation of pain. Furthermore, *G. vulgaris* is documented in several sources, including the *List of Medicinal Plants in Guangxi*<sup>[3]</sup>, *Medicinal Plant Resources of the Yao Ethnic Group's Dragon Boat Festival Medicine Market in Gongcheng, Guangxi*<sup>[4]</sup>, and *Flora of Guangdong* (Volume 9)<sup>[5]</sup>. This species is traditionally employed in the treatment of various conditions, such as traumatic injuries, fractures, rheumatoid arthritis, as well as abscesses, carbuncles, and sores of unknown etiology<sup>[2]</sup>. The *Lingnan Herbal Medicine Record*<sup>[6]</sup> documents that *G. vulgaris* exhibits multiple properties, including the promotion of blood circulation, the alleviation of blood stasis, and the facilitation of bone regeneration and tendon repair. It is particularly effective in treating the middle stage of fractures, specifically during callus formation, indicating

its considerable therapeutic value.

The ongoing revisions of the *Chinese Pharmacopoeia* have established a drug quality supervision framework characterized by distinct Chinese features. Since the implementation of the fifth edition in 1990, the included medicinal material varieties encompass only standardized evaluation systems for morphological identification, microscopic examination, moisture and ash content determination, and extract tests. However, these systems are inadequate for quantifying the content of active ingredients and are even less effective in evaluating variations in the quality of medicinal materials. The 2025 edition of the *Chinese Pharmacopoeia* has enhanced the threshold limits for heavy metals and harmful elements in Chinese medicinal materials and decoction pieces, thereby further standardizing the regulatory framework for these substances<sup>[7]</sup>. To improve the quality standards of the Yao medicinal material *G. vulgaris*, this study conducted morphological identification, microscopic analysis, thin-layer chromatography (TLC), and measured moisture content, total ash, acid-insoluble ash, and extract content in 24 batches of *G. vulgaris* medicinal materials. Comprehensive quality control measures were thereby established for *G. vulgaris* medicinal materials. Furthermore, the concentrations of heavy metals and pesticide residues in *G. vulgaris* were analyzed to ensure its safety. The specifications for identification and inspection items of *G. vulgaris*, as outlined in the *Chinese Pharmacopoeia* and the first volume of the *Quality Standards for Yao Medicinal Materials in the Guangxi Zhuang Autonomous Region*, were enhanced to provide empirical data supporting quality control of the medicinal material. This effort aims to establish a robust foundation for the standardized development and clinical ap-

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plication of the Yao medicinal material *G. vulgaris*.

## 2 Materials

**2.1 Main instruments** The main instruments utilized in this study comprised the Ni-U system microscope (Nikon, Japan), DS-Ri2 imaging system (Nikon, Japan), ZF-20C dark box three-in-one ultraviolet analyzer (Shanghai Precision Instrument Co., Ltd.), SQP, BSA224S electronic balance (Sartorius, Germany), DHG-9240A electric heating blower drying oven (Shanghai Yiheng Scientific Instrument Co., Ltd.), 7800 ICP-MS (Agilent, USA), and GC-2010PLUS gas chromatograph (Shimadzu, Japan).

**2.2 Main reagents** The main reagents utilized in this study were as follows: phosphomolybdic acid hydrate (Shanghai Alading Biochemical Technology Co., Ltd., batch No.: H2307209); silica gel G plates (Qingdao Marine Chemical Plant Branch, batch No.: 20240806); glycerin (Sinopharm Chemical Reagent Co., Ltd., batch No.: 20210720); ethanol (Nanjing Quanlong Biotechnology Co., Ltd., batch No.: 20220301); chloral hydrate

(Qingdao Yulong Seaweed Co., Ltd., batch No.: 20190902); dichloromethane (Shanghai Meryer Biochemical Technology Co., Ltd., batch No.: M82187); hydrochloric acid (Sinopharm Chemical Reagent Co., Ltd., batch No.: 2024101502); ICP analysis multi-element standard solution (National Nonferrous Metals and Electronic Materials Analysis and Testing Center, batch No.: GNM-M241289-2013); pentachloronitrobenzene (Tianjin Alta Technology Co., Ltd., batch No.: FS1604246); and DDT and hexachlorocyclohexane (TMRM Quality Inspection Technology Co., Ltd., batch No.: A22110634b).

**2.3 Medicinal materials** The samples of the medicinal material *G. vulgaris* used in this test were collected from various locations. These samples were identified as the dried aerial part of *G. vulgaris*, belonging to the Acanthaceae family, by Associate Professor Dai Zhonghua of Guangxi University of Chinese Medicine<sup>[1]</sup>. Detailed information regarding the samples is presented in Table 1. The reference medicinal material of *G. vulgaris* (batch No.: ZSNF-TKBM) was obtained from China National Institute for Food and Drug Control.

Table 1 Information of *Gendarussa vulgaris* samples from various production areas

Sample No.		Place of origin	Sample No.		Place of origin
S-1	Minzhu South Road, Yuzhou District, Yulin City, Guangxi		S-13	Jinli Town, Zhaoqing City, Guangdong	
S-2	Zhenzhong Village, Nanjiang Street, Yuzhou District, Yulin City, Guangxi		S-14	Jiangnan District, Nanning City, Guangxi	
S-3	Tanfu Village, Qingtang Town, Qinbei District, Qinzhou City, Guangxi		S-15	Shijiao Town, Qingyuan City, Guangdong	
S-4	Shuzai Town, Dianbai District, Maoming City, Guangdong		S-16	Jiedong District, Jieyang City, Guangdong	
S-5	Kangqiao District 1, Qinbei District, Qinzhou City, Guangxi		S-17	Pingyuan County, Meizhou City, Guangdong	
S-6	Gaozhou City, Maoming City, Guangdong		S-18	Tianquan County, Chengdu City, Sichuan	
S-7	Puning City, Jieyang City, Guangdong		S-19	Xiangzhou County, Laibin City, Guangxi	
S-8	Qiaocheng District, Bozhou City, Anhui		S-20	Chengzhong District, Liuzhou City, Guangxi	
S-9	Xiashan District, Zhanjiang City, Guangdong		S-21	Qintang District, Guigang City, Guangxi	
S-10	Shalang Town, Dianbai District, Maoming City, Guangdong		S-22	Litang Town, Binyang County, Guangxi	
S-11	Rongcheng County, Baoding City, Hebei		S-23	Dalang Town, Dongguan City, Guangdong	
S-12	Yizhou District, Hechi City, Guangxi		S-24	Liucheng County, Liudong District, Liuzhou City, Guangxi	

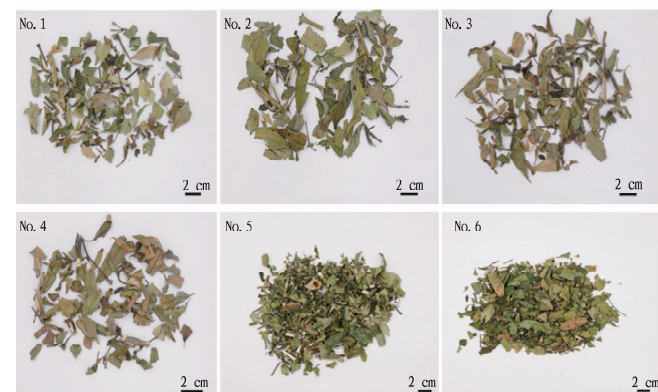
## 3 Methods and results

**3.1 Morphological identification** Morphological identification of *G. vulgaris* was conducted across 24 production areas. According to the 2025 edition of the *Chinese Pharmacopoeia*, the primary identification characteristics are as follows. Stems are cylindrical and branched, 40–90 cm in length and 0.2–3.0 cm in diameter. The stem surface displays colors ranging from yellowish-green to light greenish-brown or brownish-green and is marked by sparse, small yellow lenticels. The twigs are slightly quadrangular with swollen nodes. It is brittle and prone to breakage, exhibiting a yellowish-white cross section. The leaves are arranged oppositely, appearing curled and fragmented. When flattened, they are narrowly lanceolate or linear-lanceolate in shape, 4–14 cm in length and 1–2 cm in width. The apex tapers gradually, while the base is cuneate; the leaf margins are entire, and the veins display a slight purple coloration. Some inflorescences are spicate, occurring terminally or in the upper leaf axils, characterized by narrow and

slender bracts and a bilabiate corolla (Fig. 1). The specimen emits a faint odor and possesses a mildly pungent and sour taste. The stem diameters, as well as the lengths and widths of leaves from 24 sample batches, were measured using a ruler. More than 80% of these measurements fell within the ranges specified in the 2025 edition of the *Chinese Pharmacopoeia*.

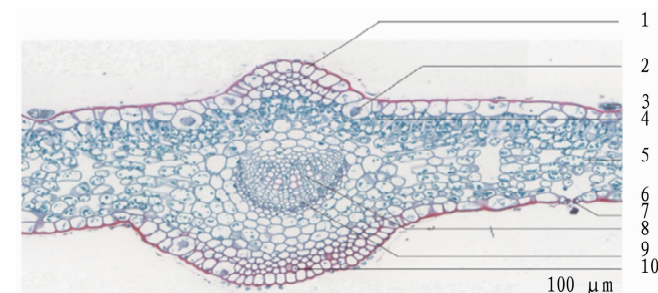
**3.2 Microscopic identification** Through microscopic examination of *G. vulgaris* samples collected from 24 production areas and with reference to the 2025 edition of the *Chinese Pharmacopoeia*, the following microscopic characteristics with diagnostic significance were identified.

**3.2.1** Cross-sectional characteristics of leaves. The upper and lower epidermis each consists of a single row of cells. The outer stratum corneum of the upper epidermis contains slightly larger cells that are square or deltoid in shape. The cells in the lower epidermis are smaller and contain stomata. Additionally, both the upper and lower epidermis possess slightly larger cells that contain cystoliths and glandular scales. The mesophyll palisade tissue is



**Fig. 1** *Gendarussa vulgaris* medicinal materials

relatively short, comprising 12 cell columns and constituting approximately 1/4 of the entire leaf. The spongy mesophyll consists of 56 cell columns characterized by irregular shapes and large intercellular spaces. The main vein extends laterally on both sides of the leaf. Multiple columns of xylem vessels are present, with 13 vessels per column. The cambium is well-defined, and the vascular bundles are ectophloic (Fig. 2).



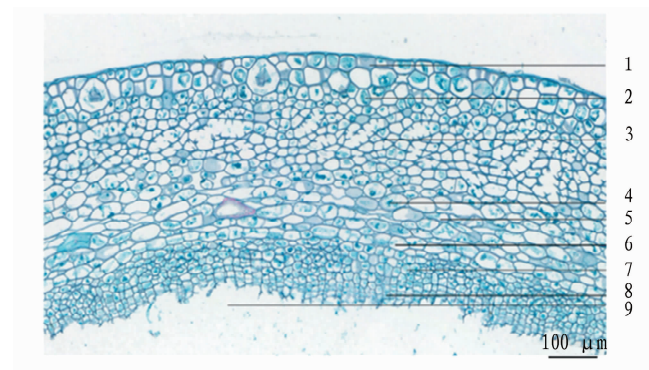
**NOTE** 1. Upper epidermis; 2. Cystoliths; 3. Glandular scales; 4. Palisade tissues; 5. Spongy tissues; 6. Lower epidermis; 7. Stomata; 8. Xylem; 9. Phloem; 10. Collenchyma.

**Fig. 2** Cross section of a leaf blade of *Gendarussa vulgaris*

**3.2.2** Cross-sectional characteristics of the stem. The outermost layer of the tender stem comprises a complex epidermis, consisting of 23 layers of epidermal cells that are approximately square or nearly round in shape. The outer wall cells are keratinized, and the epidermal cells contain crystals (cystoliths) that are conical or nearly round. Glandular scales are frequently observed. Chloroplast-containing cells are arranged in a ring pattern and are secondarily present within the complex epidermis. The middle layer is composed of 57 layers of cortical cells, which are encased by sclereids. The inner cortex is distinctly defined; the phloem is narrow, while the xylem forms a ring consisting of 13 columns of cells. The medullary region is broad, with polygonal parenchymal cells occupying this area (Fig. 3).

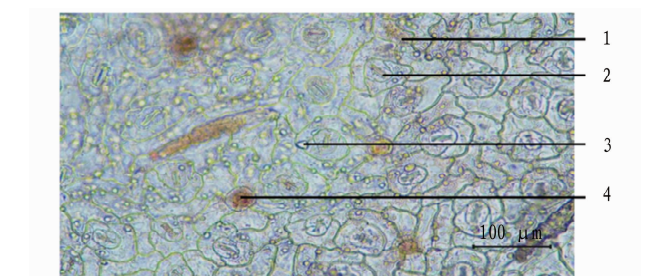
**3.2.3** Characteristics of leaf surface slices. The epidermal cells exhibit irregular, triangular, or polygonal shapes. The anticlinal walls are polygonal in form. Oval or elongated oval cystoliths, glandular scales, and crystals are frequently observed within the epidermis. The stomata are characterized as anisocytic or anomocytic. The glandular scale head is flat, round, and spherical,

comprising four cells (Fig. 4).



**NOTE** 1. Epidermis; 2. Cystoliths; 3. Collenchyma; 4. Cortex; 5. Sclereids; 6. Endodermis; 7. Phloem; 8. Xylem; 9. Pith.

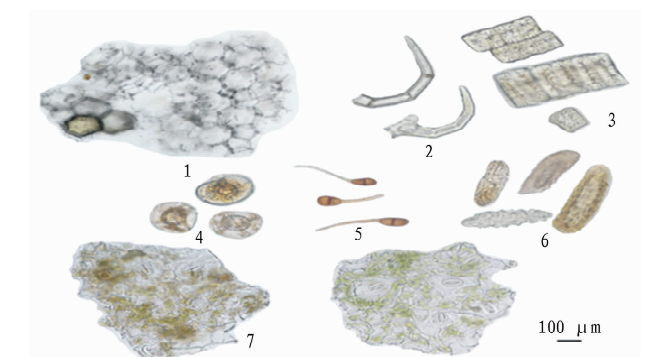
**Fig. 3** Cross section of the stem of *Gendarussa vulgaris*



**NOTE** 1. Cystoliths; 2. Stomata; 3. Square crystals; 4. Glandular scales.

**Fig. 4** Leaf surface slices of *Gendarussa vulgaris*

**3.2.4** Powder characteristics. The powder exhibits a color range from yellowish-green to yellowish-brown. Cystoliths are oval or nearly rectangular in shape. Numerous sclereids, which are yellow in color, measure between 20 and 80  $\mu\text{m}$  in diameter and display distinct stratification. The glandular scales possess 4-celled heads and a unicellular stalk. Non-glandular hairs consist of 2–4 cells. The epidermal cells beneath the leaves are rectangular or polygonal, characterized by slightly curved anticlinal walls. The stomata are either diacytic or anisocytic. Parenchymal cells contain calcium oxalate square crystals, with diameters ranging from 2 to 10  $\mu\text{m}$  (Fig. 5).

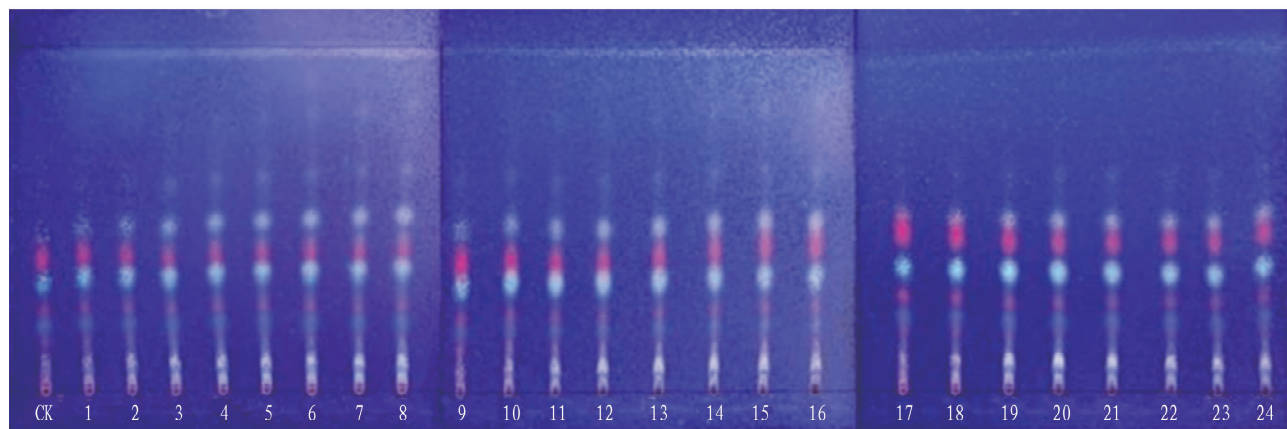


**NOTE** 1. Parenchymal cells; 2. Non-glandular hairs; 3. Sclereids; 4. Glandular scales; 5. Small glandular hairs; 6. Cystoliths; 7. Stomata.

**Fig. 5** Microscopic characteristics of *Gendarussa vulgaris*

**3.3 Thin-layer chromatography identification** According to the Identification Section for *G. vulgaris* in the 2025 edition of the *Chinese Pharmacopoeia*<sup>[1]</sup>, 2 g of the powdered sample was combined with 50 mL of ethanol and subjected to ultrasonic treatment for 30 min, followed by filtration. The filtrate was evaporated to dryness, and the resulting residue was dissolved in 1 mL of ethanol to prepare the test solution. Similarly, 2 g of the reference medicinal material of *G. vulgaris* was processed in the same manner to obtain the reference solution. The test was performed in accordance with the TLC method outlined in General Rule 0502. The sample application conditions were as follows: silica gel G thin-

layer plate, sample volume of 2  $\mu$ L, temperature maintained at 23.2  $^{\circ}$ C, relative humidity at 38%, developer solvent composed of dichloromethane-methanol (16 : 5, *v/v*), and chromogenic agent consisting of a 10% sulfuric acid ethanol solution. Chromogenic detection was conducted under ultraviolet light at 365 nm. The TLC identification was carried out on *G. vulgaris* medicinal materials obtained from 24 different production areas. In the chromatogram of the test sample, fluorescent spots exhibiting the same color were observed at positions corresponding to those of the reference herbal material in the chromatograms of test solutions derived from 24 distinct production areas of *G. vulgaris* (Fig. 6).



NOTE CK. Reference medicinal material; 1–24. S–1 ~ S–24.

Fig. 6 TLC chromatograms of *Gendarussa vulgaris* from 24 production areas

### 3.4 Determination indicators

**3.4.1 Moisture content.** According to the drying method specified in Item 0832 of General Rule IV in the 2025 edition of the *Chinese Pharmacopoeia*<sup>[8]</sup>, the moisture content of *G. vulgaris* powder was determined. The moisture content of *G. vulgaris* samples from 24 production areas ranged from 2.90% to 9.33%, with an average value of 6.81%, and the relative standard deviation (*RSD*) was below 3% for all samples (Table 2). Based on the moisture limit outlined in Part I of the 2025 edition of the *Chinese Pharmacopoeia*, the moisture content limit for *G. vulgaris* was established at a maximum of 10.0%. All samples from the 24 production areas complied with this specification.

**3.4.2 Total ash content.** The total ash content was determined in accordance with Item 2302 of General Rule IV in the 2025 edition of the *Chinese Pharmacopoeia*<sup>[8]</sup>, with calculations based on the weight of the residue. The total ash content of *G. vulgaris* samples from 24 production areas ranged from 9.29% to 10.97%, with an average value of 10.46% and *RSD* below 3% for all samples (Table 2). According to the limit specified in Part I of the 2025 edition of the *Chinese Pharmacopoeia*, the maximum allowable total ash content for *G. vulgaris* was set at 11.0%. All 24 batches complied with this requirement.

**3.4.3 Acid-insoluble ash content.** The acid-insoluble ash content was determined following Item 2302 of General Rule IV in the

2025 edition of the *Chinese Pharmacopoeia*<sup>[8]</sup>, with calculations based on the weight of the residue. The measured acid-insoluble ash values ranged from 0.53% to 1.97%, averaging 1.57%, and the *RSD* was consistently below 3% (Table 2). According to the total ash content limit specified in Part I of the 2025 edition of the *Chinese Pharmacopoeia*, the maximum allowable acid-insoluble ash content was set at 2.0%. All samples of *G. vulgaris* collected from 24 production areas complied with this regulatory standard.

**3.4.4 Extract content.** In accordance with the provisions of Item 2201 of General Rule IV in the 2025 edition of the *Chinese Pharmacopoeia* (hot dipping method)<sup>[8]</sup>, the contents of water-soluble and alcohol-soluble extracts in *G. vulgaris* powder were determined. The water-soluble extract content ranged from 12.17% to 29.69%, with a mean value of 23.07% and *RSD* below 3%. All samples from various production areas exhibited water-soluble extract contents exceeding 8.0%, consistent with the minimum requirement specified in the 2025 edition of the *Chinese Pharmacopoeia*. The alcohol-soluble extract content ranged from 12.79% to 22.73%, with an average of 18.28% and *RSD* also below 3% (Table 2). Comparative analysis revealed no significant overall difference between the water-soluble and alcohol-soluble extract contents; however, the majority of samples demonstrated slightly higher water-soluble extract levels. Consequently, the extract content standard may continue to be based on the water-soluble

extract criterion established in the 2025 edition of the *Chinese Pharmacopoeia*.

**Table 2** Measurement results of the examination indicators of *Gendarussa vulgaris* ( $n=3$ ,  $x$ , %,  $RSD<3\%$ )

No.	Moisture	Total ash	Acid-insoluble ash	Water-soluble extract	Alcohol-soluble extract
S-1	8.52	9.71	0.75	29.69	19.07
S-2	9.14	10.96	0.83	24.87	18.64
S-3	9.12	10.21	0.93	26.04	18.54
S-4	5.42	10.93	1.84	24.09	19.07
S-5	6.28	10.97	1.66	28.62	19.22
S-6	6.26	10.96	1.97	24.88	17.80
S-7	8.22	10.87	1.84	19.95	13.34
S-8	6.12	9.64	1.94	25.56	20.11
S-9	2.90	10.92	1.90	23.95	17.85
S-10	6.06	10.88	1.94	19.27	12.79
S-11	8.11	10.09	1.52	26.55	19.52
S-12	5.70	10.57	1.35	25.68	21.61
S-13	6.11	10.67	1.81	19.21	22.73
S-14	9.16	10.73	1.94	24.46	20.47
S-15	6.22	9.58	1.86	21.74	20.16
S-16	3.20	10.87	1.93	27.19	20.24
S-17	4.88	10.91	1.47	26.18	21.36
S-18	6.30	9.77	1.87	19.75	22.65
S-19	9.20	10.82	0.53	22.93	17.24
S-20	6.03	10.77	1.14	21.39	17.76
S-21	6.29	10.32	1.52	19.36	13.51
S-22	8.84	10.91	1.86	12.17	13.25
S-23	6.00	9.72	1.86	19.63	18.15
S-24	9.33	9.29	1.47	20.44	13.57

**3.4.5** Heavy metals and pesticide residues. *G. vulgaris* medicinal materials were collected from 18 production areas characterized by distinct production environments and geographically distant locations for this study. In accordance with Method 2321 of Part IV in the 2025 edition of the *Chinese Pharmacopoeia*<sup>[8]</sup>, concentrations of five heavy metals, including lead, cadmium, arsenic, mercury, and copper, were quantified using inductively coupled plasma mass spectrometry<sup>[9]</sup>. Additionally, residues of nine pesticides were analyzed by gas chromatography following Method 2341 of Part IV in the 2025 edition of the *Chinese Pharmacopoeia*<sup>[8,10]</sup>. The results indicated lead contents ranging from 0.188 to 2.86 mg/kg, cadmium from 0.00 to 0.046 7 mg/kg, total arsenic from 0.00 to 0.233 mg/kg, copper from 2.62 to 5.12 mg/kg, and mercury from 0.000 to 0.043 1 mg/kg. None of the nine organochlorine pesticides, including total DDT (pp'-DDE, pp'-DDD, op'-DDT, pp'-DDT), total hexachlorocyclohexane ( $\alpha$ -BHC,  $\beta$ -BHC,  $\gamma$ -BHC,  $\delta$ -BHC), and pentachloronitrobenzene, were detected (Table 3).

4 Discussion

Microscopic analysis revealed that the combined presence of cystoliths (elliptical or rectangular), glandular scales (comprising a 4-celled head and a unicellular stalk), sclereids (exhibiting distinct stratification), and calcium oxalate square crystals within the leaves, cross sections of young stems, and powdered samples of *G. vulgaris* demonstrates significant specificity. Notably, the cystoliths and glandular scales located in the upper and lower epider-

**Table 3** Heavy metal and pesticide residue results of *Gendarussa vulgaris* mg/kg

No.	DDT	Hexachlorocyclohexane	Pentachloronitrobenzene	Lead	Cadmium	Total arsenic	Mercury	Copper
S-1	-	-	-	0.769	0.036 5	0.116	0.010 2	4.37
S-2	-	-	-	0.686	0.046 7	0.171	0.039 8	3.17
S-4	-	-	-	0.307	0.036 3	0.146	0.022 5	3.84
S-5	-	-	-	0.531	0.020 8	0.117	0.025 9	3.60
S-6	-	-	-	0.514	0.025 5	0.200	0.020 1	4.28
S-7	-	-	-	0.477	0.035 1	0.160	0.013 4	4.27
S-8	-	-	-	0.463	0.034 2	0.122	0.012 0	4.32
S-10	-	-	-	0.305	0.023 9	0.084 3	0.013 0	4.23
S-11	-	-	-	0.389	0.029 1	0.233	0.015 6	5.12
S-12	-	-	-	0.318	0.020 8	0.097 2	0.016 1	3.77
S-13	-	-	-	0.441	0.046 7	0.135	0.011 2	4.15
S-14	-	-	-	2.86	0.040 9	0.201	0.034 0	4.69
S-15	-	-	-	0.274	0.025 5	-	-	3.01
S-16	-	-	-	0.309	0.023 9	0.061 2	0.010 7	2.84
S-18	-	-	-	0.446	0.025 1	0.116	0.022 1	4.00
S-19	-	-	-	0.274	-	0.059 0	-	4.25
S-20	-	-	-	0.554	0.025 1	0.123	0.043 1	3.06
S-21	-	-	-	0.188	0.015 3	-	-	2.62

**NOTE** -: Not detected; the detection limits are as follows: cadmium, 0.01 mg/kg; mercury, 0.01 mg/kg; lead, 0.05 mg/kg; copper, 1 mg/kg; and total arsenic, 0.05 mg/kg.

mal cells of the stem and leaf cross sections serve as primary diagnostic features for identification. The TLC identification results

demonstrated that the medicinal material powder was subjected to ultrasonic extraction using ethanol, followed by spotting and devel-

opment with a dichloromethane-methanol solvent system (16 : 5, *v/v*). Subsequently, the plate were sprayed with a 10% sulfuric acid ethanol solution and visualized under a 365 nm ultraviolet lamp. Samples obtained from 24 production regions, along with the reference medicinal materials, exhibited fluorescent spots of identical color at corresponding positions, characterized by distinct spots and effective separation. According to the current quality standards for medicinal materials, *G. vulgaris* has incorporated an additional test item for acid-insoluble ash. Based on the test results, the established limit standards are as follows: moisture content must not exceed 10.0%, total ash content must not exceed 11.0%, acid-insoluble ash content must not exceed 2.0%, and water-soluble extract content must be no less than 8.0%.

The primary pathway for heavy metals to enter the human body involves contaminated soil leading to the accumulation of medicinal materials, followed by prolonged low-dose intake. This exposure frequently results in chronic, multisystem, and irreversible damage<sup>[11]</sup>. According to the fifth item of the *Guiding Principles for Setting Limits of Harmful Residues in Traditional Chinese Medicine*, as outlined in Item 9302 of Part IV in the 2025 edition of the *Chinese Pharmacopoeia*<sup>[8]</sup>, the permissible limits for heavy metals and harmful elements in medicinal materials and decoction pieces (plant-based) are as follows: lead, cadmium, arsenic, mercury, and copper shall not exceed 5, 1, 2, 0.2, and 20 mg/kg, respectively. Based on the test results, the concentrations of these five heavy metals did not exceed the specified limits, and none of the nine organochlorine compounds were detected. Given the variability in heavy metal content across the 18 production areas, it is recommended to implement a heavy metal limit test for *G. vulgaris* in accordance with the standardized criteria outlined in the *Chinese Pharmacopoeia*. Specifically, the concentrations of lead, cadmium, arsenic, mercury, and copper should not exceed 5, 1, 2, 0.2, and 20 mg/kg, respectively.

The medicinal component of *G. vulgaris* consists of the dried aerial parts, which are susceptible to dust accumulation. To effectively manage inorganic contaminants, including sand and mud, an additional test for acid-insoluble ash was incorporated. For the first time, heavy metal analyses were performed on *G. vulgaris* samples collected from 18 production areas, revealing significant variability in heavy metal content across different locations. This variation is hypothesized to be associated with environmental factors, such as soil heavy metal concentrations, as well as cultivation practices, including fertilization and pesticide application. Although the pesticide residue levels in this test did not exceed the

established standards, given the variability in heavy metal content across different production areas, it is recommended to incorporate heavy metal limit tests into the standards for medicinal materials by adhering to the general rules of the *Chinese Pharmacopoeia*. This approach would provide valuable data to ensure the safety of *G. vulgaris* medicinal resources and support the enhancement of medicinal material standards.

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