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ABSTRACT

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Introduction: Although there is a wide range of studies on phytochemistry and pharmacology, there currently needs to be a standard quality control method or index for assessing the components of X. sorbifolium Bunge to ensure quality. Objectives: This study aimed to determine some quality parameters of the standardization of trunks and branches of Xanthoceras sorbifolia Bunge. Methods: The cellular diagnostic structures of *trunks and branches* of the plant were defined by "Olympus" light microscopy. Biologically active compounds were identified using TLC and HPLC, and the main biologically active compounds' contents were determined by HPLC methods. Some quality parameters of trunks and branches of X. sorbifolium Bunge were determined by Chinese Pharmacopoeia methods. Results: Microstructure of trunk and branch of the X. sorbifolia Bunge was defined. Some quality parameters of trunks and branches of X. sorbifolia Bunge collected from seven locations in Inner Mongolia were defined as: moisture 4.8-6.1%, total ash 5.57-7.3%, acid-insoluble ash 1.3-2.7%, alcohol-soluble extractive 8.97-11.63%. The fingerprints of TLC to reveal epicatechin and HPLC to reveal epicatechin, dihydromyricetin, dihydroquercetin, myricetin, quercetin, and naringenin were defined. Some flavonoid contents in trunks and branches of X. sorbifolia Bunge collected from seven locations in Inner Mongolia were determined according to the developed and validated method as: epicatechin 6.39-11.687 mg/g, dihydroquercetin 1.02-1.833 mg/g, and myricetin 0.02-2.693 mg/g. Conclusion: The standardization criteria for the trunk and branches of X. sorbifolia Bunge were defined. Quality parameters and contents of epicatechin, dihydroquercetin, and myricetin were different in Xanthoceras sorbifolia Bunge collected from seven locations in Inner Mongolia.

Key words: Epicatechin, Dihydroquercetin, Myricetin, Mongolian medicine.

INTRODUCTION

Plant materials are used throughout developed and developing countries as home remedies, over-thecounter drug products, and raw materials for the pharmaceutical industry, representing a substantial proportion of the global drug market. The World Health Assembly - in resolutions WHA31.33 (1978), WHA40.33 (1987), and WHA42.43 (1989) - has emphasized the need to ensure the quality of medicinal plant products by using modern control techniques and applying suitable standards.1 The safety and efficacy of herbal medicines largely depend on their quality. Requirements and methods for quality control of finished herbal products, particularly for a mixture of herbal products, are far more complex than chemical medicines. The quality of the raw materials used also influences the quality of finished herbal products.² According to WHO guidelines, an herbal product must be standardized before being released into the market.3

Xanthoceras sorbifolium Bunge is the only species of the genus Xanthoceras in the family Sapindaceae and is native to Shaanxi, Shanxi, Hebei, Gansu, and Inner Mongolia in northwestern China.⁴⁻⁶ Xanthoceras sorbifolium Bunge is a multipurpose plant; all plant sections are edible, medicinal, economical, and of ecological value. The plant, commonly called the yellow horn or golden horn, is widely used in Chinese and Mongolian traditional medicines to treat arterial sclerosis, hyperlipidemia, hypertension, chronic hepatitis, rheumatism, and arthritis.⁷⁻¹⁰ Various parts of the plant with different medicinal values are used medicinally, and most of its recorded traditional applications are concentrated in northern China. The trunks and branches, fruits, leaves, and other parts are medicinally used, and in China, different sections, such as fruit, tea, and cooking oil, are utilized as food. Modern pharmacology has gradually verified the traditional efficacy of X.sorbifolium Bunge and explored its role in treating Alzheimer's disease, rheumatism, vasculitis, scabies, and other conditions.¹⁰ A total of 278 compounds have been isolated from different sections of X.sorbifolium Bunge, including triterpenoids (124), flavonoids (48), phenylpropanoids (14), steroids (17), phenols (17), fatty acids (29), alkaloids (9), quinones (4), and others (16).10-14 The main pharmacological activities of *X.sorbifolium Bunge* include improving learning and memory impairments, anti-inflammatory, antitumor, anti-oxidation, and lipid-lowering effects.10,15

The trunks and branches of X.sorbifolium Bunge, called "xi la sen deng" in traditional Mongolian medicine, are used to treat arthritis in Inner Mongolia.^{10,16} A total of 46 compounds have been isolated from trunks and branches of X.sorbifolium Bunge, including triterpenoids (7), flavonoids (27), steroids (2), phenols (3), fatty acids (3), quinones (2), and others (2).¹⁰ n-Butanol extract of the wood of X.sorbifolium Bunge has an antagonistic effect on adjuvant arthritis in rats, and its mechanism may be related to the inhibition of immune function. 3-Oxotirucalla-7,24-dien-21-oic acid, oleanolic acid, and epicatechin were found to be inhibitory

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substances against human immunodeficiency virus (HIV-1) protease, and catechin, epicatechin, myricetin, and dihydromyricetin isolated from the wood have an anti-oxidation effect.¹⁷⁻¹⁹

Although there is a wide range of studies on phytochemistry and pharmacology, there currently needs to be a standard quality control method or index for assessing the components of *X.sorbifolium Bunge*. Therefore, considerable research should be devoted to creating a standard quality assessment approach to ensure the quality of *X. sorbifolium Bunge*. Specifically, it is necessary to determine *X.sorbifolium Bunge* contents or produce standardized fingerprints to index the components of this species.¹⁰ This study aimed to determine some quality parameters of the standardization of trunks and branches of *X.anthoceras sorbifolia* Bunge to ensure quality.

MATERIALS AND METHODS

Plants material

The crude herbal medicines from *Xanthoceras sorbifolia* Bunge collected or purchased from seven locations in Inner Mongolia, including Baotou, Daban, Jarud, Jalaid, Ordos, Fuxin, and Hospital of Inner Mongolia Minzu University (IMMU), were used in the study. The plants were identified by Laxinamujila, professor of the Department of Mongolian Medicine, Mongolian Medical College of the (IMMU), and voucher specimens have been deposited in the Herbarium of the IMMU, China.

Standards and chemicals

Reference epicatechin (\geq 99.65% HPLC, A0049), dihydromyricetin (\geq 99.57% HPLC, A00771), dihydroquercetin (\geq 98.23% HPLC, A140811), myricetin (\geq 98.31% HPLC, A0048), quercetin (\geq 99.65% HPLC, A0082), and naringenin (\geq 99.18% HPLC, A0147) from *Beijing Putian* Tongchuang Biological Technology Co., Ltd, China were used for the study. Solvents used for the HPLC study were of HPLC grade, and all other reagents and solvents were of analytical grade.

Powder microscopy

The powdered plant raw material from *trunks and branches* of *X.sorbifolium* Bunge purchased from the hospital of IMMU was softened in chloral hydrate $(C_2H_3Cl_3O_2)$ for 15 minutes. The small amount of powder was placed on a microscope slide with a drop of choral hydrate and covered with cover glass. The microstructure was determined using Olympus light microscopy equipped with a digital camera.

Determination of quality parameters

Moisture, total ash, acid-insoluble ash, and alcohol-soluble extractive value were determined by Chinese Pharmacopoeia methods.

Chemical analysis

TLC identification of biologically active substances

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The powders (1g) of *trunks and branches* of *Xanthoceras sorbifolia* Bunge from seven places in Inner Mongolia were extracted with 25 mL of methanol using ultrasonic for 20 minutes, respectively. 4 mg of reference epicatechin was precisely weighed to an accuracy of 0.001 g and solved in 1 mL of methanol. The sample and reference solutions were applied to TLC plates (Silica gel 60 GF 254, *Qingdao Haiyang* Chemical Co., Ltd., China) at 5µL at a 1.5 cm distance from the lower border of the plate. The development of the plate has been performed with a mobile phase of cyclohexane-acetone-glacial acetic acid (10:5:1, v/v). After drying at room temperature, the plate was visualized under 254 UV light.

HPLC identification of biologically active substances

Preparation of sample solution

2 g of the powders of *Xanthoceras sorbifolia* Bunge was precisely weighed to an accuracy of 0.001 g and placed in a 100 mL volumetric flask, and ultrasonically extracted with 25 mL of methanol for 20 minutes at a temperature of 30°C, respectively. After putting it at room temperature for 30 minutes, the extract was weighed. The lost weight was made up with methanol, shaken well, and filtered through 0.22 μ m filter membranes before being injected into the HPLC instrument.

Standard solution preparation for the qualitative determination

4 mg of reference epicatechin, 0.23 mg of reference dihydroquercetin, 0.38 mg of reference quercetin, 0.2 mg of reference myricetin, 4.93 mg of reference dihydromyricetin, and 0.31 mg of reference naringenin were precisely weighed an accuracy of 0.0001 g into a 10 mL volumetric flask and dissolved in 10 mL of methanol, respectively. Mix 1 mL of each solution evenly to prepare a mixture solution. The solution was filtered through a 0.22 μ L filter before injecting it into the HPLC instrument.

Standard solution preparation for the quantitative determination

4 mg of reference epicatechin, 0.23 mg of reference dihydroquercetin, and 0.2 mg of reference myricetin were precisely weighed to an accuracy of 0.001 g and placed in a 10 mL volumetric flask. The mixed stock solution was prepared in 10 mL of methanol and filtered through a 0.22 μ L filter.

Chromatographic procedures: The C18 column (300 mm x 4.6 mm x 5 μ m, Venusil MP) was used as the stationary phase. Acetonitrile (A) and acetic acid (B) were used as mobile phase with gradient system: 0-20 min, 90-80% A; 20-40 min 80-70% A; 40-45 min, 70-60% A; 45-50 min, 60-45% A; 50-60 min, 45-25% A; 60-80 min, 25-20% A; 80-95 min, 20-15% A. The total runtime: 95 min; flow rate: 1.0 mL/min; column temperature: 30°C; injection volume: 5 μ L; UV detection wavelength: 274 nm.

Validation of the developed HPLC method for the Quantitative determination of epicatechin, dihydroquercetin, and myricetin

ICH guideline was followed to validate developed methods with precision, repeatability, accuracy, LOD, and LOQ.

Selectivity and specificity: The selectivity and specificity of the developed method to measure epicatechin, dihydroquercetin, and myricetin in *trunks and branches of Xanthoceras sorbifolia* Bunge were defined.

Linearity

Preparation of stock solution: An accurately weighed 4 mg of reference epicatechin, 0.23 mg of reference dihydroquercetin, and 0.2 mg reference myricetin were transferred to a 10 mL volumetric flask, and 10 mL of methanol was added, respectively.

Precision: 4 μ L, 8 μ L, 12 μ L, 16 μ L, and 20 μ L of each stock solution were injected into the HPLC apparatus. The analysis was carried out at different intervals on the same day with six repeats.

Accuracy: For the accuracy of the proposed method, recovery studies were performed by the standard addition method. A known amount of reference epicatechin, dihydroquercetin, and myricetin was added to the pre-analyzed plant powder, and the sample was then analyzed by the proposed method.

Statistical analysis

The data were reported as the mean±standard deviation. Quality parameters experiments were repeated three times, and quantitative determination of the content of epicatechin, dihydroquercetin, and myricetin in a sample was repeated six times using linear regression analysis. Analyzes were performed using SPSS Statistics 26.0 software.

RESULTS

Microstructural study

The powder of *trunks and branches of Xanthoceras sorbifolia* Bunge is red-brown, brown-yellow, or yellowish-white in color. Many colorless or pale yellow woody fibers (Figure 1-A), some cells around some fibrous bundles contain square crystals of calcium oxalate and aggregates of calcium oxalate (Figure 1-B), crystal fibrous bundles are formed (Figure 1-C), woody rays (Figure 1-D) and thin-walled woody cells are rectangular or round (Figure 1-E), pale yellow reticular canals are well visible (Figure 1-F), and irregular brown spots are common (Figure 1-G).

Quality parameters

Quality parameters of *Xanthoceras sorbifolia* Bunge were defined according to Chinese Pharmacopoea, and the results are shown in Table 1.

TLC fingerprinting

Epicatechin was revealed in trunks and branches of *Xanthoceras sorbifolia* Bunge using cyclohexane-acetone-glacial acetic acid (4:4:0.2 v/v) solvent system (Figure 2).

The spots in the chromatogram obtained with the test solutions correspond in Rf value and color to those obtained with the standard solution.

HPLC analysis

Standard and Sample solutions were prepared according to the Section on preparing the sample and standard solutions. Five μL of

each solution was injected into the HPLC system according to the chromatographic conditions given in the section on chromatographic procedures, and the chromatograms were recorded. A retention time was 20.33 min for epicatechin, 22.67 min for dihydromyricetin, 32.32 min for dihydroquercetin, 40.76 min for myricetin, 50.27 min for quercetin, 53.47 min for naringenin (Figure 3).

The chromatogram of *Xanthoceras sorbifolia* Bunge indicated the presence of epicatechin, dihydromyricetin, dihydroquercetin, myricetin, quercetin, and naringenin, compared with reference substances (Figure 4).

The HPLC fingerprint of the methanol extract of the trunk and branch of *Xanthoceras sorbifolia* Bunge (Figure 5) was obtained, and compared to the chromatograms of 7 samples, the similarity was more significant than 0.99.

Method validation

The HPLC method for estimating epicatechin, dihydroquercetin, and myricetin content was developed and validated according to ICH Q2 (R1) guidelines (Table 2).

Specificity

According to ICH Q2 (R1) guidelines, specificity is the ability to assess unequivocally the analyte in the presence of components that may be expected to be present. The results of specificity are shown in Figure 6.

Linearity

According to ICH Q2 (R1) guideline, the linearity of an analytical procedure is its ability (within a given range) to obtain test results that are directly proportional to the concentration (amount) of analyte in the sample. The results of linearity are shown in Figure 7.

Accuracy

ICH Q2 (R1) guidelines specified that the accuracy of an analytical procedure expresses the closeness of agreement between the value, which is accepted either as an actual conventional value or an accepted reference value, and the value found. The results of accuracy are shown



Figure 1: Microscopic analysis of Xanthoceras sorbifolia Bunge

A. Woody fibers, B. Crystals of calcium oxalate, C. Crystal fibers, D. Wood-ray cells, E. Thin-walled woody cells, F. Reticular canals, G. Irregular brown spot

Table 1: Quality parameters of Xanthoceras sorbifolia Bunge.

| N | Sample | Moisture content, % | Total ash, % | Acid-insoluble ash, % | Alcohol soluble extractive value, % |
|---|------------------|---------------------|-----------------|-----------------------|--|
| 1 | Baotou | 4.8±0.1 | 7.1±0.00 | 2.4±0.00 | 9.6±0.00 |
| 2 | Daban | 5.13±0.12 | 5.57±0.06 | 1.3±0.1 | 11.63±0.15 |
| 3 | Hospital of IMMU | 5.23±0.126 | 5.73±0.21 | 1.5±0.1 | 8.97±0.12 |
| 4 | Jarud | 6.1 ± 0.00 | 6.47 ± 0.06 | $2.27{\pm}~0.06$ | 10.7 ± 0.17 |
| 5 | Jalaid | 5.6 ± 0.00 | 6.37 ± 0.21 | $2.47{\pm}~0.06$ | 9.7 ± 0.00 |
| 6 | Ordos | 5.43±0.06 | 6.77±0.06 | 2.3±0.00 | 10.5±0.00 |
| 7 | Fuxin | 5.7±0.1 | 7.3±0.00 | 2.7±0.1 | 9.5±0.00 |
| | p-value | < 0.001 | < 0.001 | < 0.001 | < 0.001 |

Quality parameters of Xanthoceras sorbifolia Bunge collected from seven places in Inner Mongolia were different (p<0.001).

Table 2: Summary of validation parameters of the proposed HPLC method.

| Parameters | Results | | | |
|--|---------------------|---------------------------|----------------------|--|
| | Epicatechin | Dihydroquercetin | Myricetin | |
| Retention time | 20.33 min | 32.32 min | 40.46 min | |
| Beer's law limit (µg/mL) | 4-20 | 0.092-0.46 | 0.08-0.48 | |
| Wavelength | 274 nm | 274 nm | 274 nm | |
| Regression equation | Y = 665395x - 18959 | Y=1458825.7764X+1115.9048 | Y = 750711x - 1460.5 | |
| Slope | 665394.5 | 1458825.77 | 750711.16 | |
| Intercept | -18959.1 | 1115.9 | -1460.6 | |
| Coefficient of correlation (r ²) | 0.9999 | 1.000 | 0.9997 | |
| Limit of detection (LOD) | 0.49 μg/mL | 0.0071 μg/mL | 0.0073 μg/mL | |
| Limit of quantification (LOQ) | 1.5 μg/mL | 0.021 μg/mL | 0.0222 μg/mL | |
| Accuracy (% RSD) | 96.55-101.63% (1.9) | 96.77-103.33% (2.5) | 98.71-101.3% (1.3) | |
| Precision (% RSD) | 0.24 | 0.42 | 0.81 | |

Table 3: Accuracy studies of the proposed HPLC method.

| Amount of compound in the sample (μg/mL) | The added amount of reference compound (μg/mL) | Observed concentration (µg/mL) | Recovery, % | Mean recovery, % | % RSD |
|--|---|---|--|--|---|
| 4.22 | 4.21 | 8.43 | 100.00 | 99.68 | 1.99 |
| 4.22 | 4.23 | 8.50 | 101.18 | | |
| 4.22 | 4.24 | 8.32 | 96.7 | | |
| 4.22 | 4.24 | 8.38 | 98.11 | | |
| 4.22 | 4.22 | 8.53 | 102.13 | | |
| 4.22 | 4.22 | 8.44 | 100.00 | | |
| 0.60 | 0.61 | 1.22 | 101.64 | 100.02 | 2.54 |
| 0.61 | 0.61 | 1.21 | 98.36 | | |
| 0.61 | 0.61 | 1.21 | 98.36 | | |
| 0.61 | 0.62 | 1.21 | 96.77 | | |
| 0.60 | 0.60 | 1.21 | 101.67 | | |
| 0.60 | 0.60 | 1.22 | 103.33 | | |
| 0.77 | 0.77 | 1.55 | 101.30 | 99.79 | 1.3 |
| 0.78 | 0.78 | 1.55 | 98.71 | | |
| 0.78 | 0.78 | 1.55 | 98.71 | | |
| 0.78 | 0.78 | 1.55 | 98.71 | | |
| 0.77 | 0.78 | 1.55 | 100.00 | | |
| 0.77 | 0.77 | 1.55 | 101.30 | | |
| | Amount of compound in the sample (µg/mL) 4.22 4.22 4.22 4.22 4.22 4.22 0.60 0.61 0.61 0.61 0.61 0.61 0.61 0.60 0.60 | Amount of compound in the sample (µg/mL) The added amount of reference compound (µg/mL) 4.22 4.21 4.22 4.23 4.22 4.24 4.22 4.24 4.22 4.22 4.22 4.22 6.60 0.61 0.61 0.61 0.61 0.61 0.61 0.61 0.61 0.61 0.61 0.61 0.61 0.61 0.61 0.61 0.61 0.61 0.62 0.60 0.60 0.60 0.77 0.77 0.78 0.78 0.78 0.78 0.77 0.78 0.77 0.78 | Amount of compound in the sample (µg/mL) The added amount of reference compound (µg/mL) Observed concentration (µg/mL) 4.22 4.21 8.43 4.22 4.23 8.50 4.22 4.24 8.32 4.22 4.24 8.38 4.22 4.24 8.53 4.22 4.22 8.44 0.60 0.61 1.21 0.61 1.21 1.21 0.61 1.21 1.21 0.61 0.61 1.21 0.61 0.61 1.21 0.61 0.61 1.21 0.61 0.61 1.21 0.61 0.61 1.21 0.61 0.62 1.21 0.60 0.60 1.21 0.60 0.60 1.21 0.60 0.61 1.21 0.77 0.78 1.55 0.78 0.78 1.55 0.78 0.78 1.55 0.77 0.78 1.55< | Amount of compound in the sample (ug/mL)The added amount of reference compound (ug/m)Observed concentration (ug/m)Recovery, %4.224.218.43100.004.224.238.50101.184.224.248.3296.74.224.228.53102.134.224.228.44100.000.600.611.2198.360.611.2198.36101.640.611.2198.36101.640.610.611.2191.640.610.611.2191.670.610.611.2191.670.610.611.21101.670.610.611.2191.330.750.601.22103.330.770.770.781.5598.710.780.781.5598.710.780.781.5598.710.770.781.55100.000.770.781.55101.000.770.781.55101.000.770.781.55101.000.770.781.55101.000.770.781.55101.000.770.781.55101.00 | Amount of compound in the sample (µg/m)He added amount of reference compound (µg/m)Observed oncentration (µg/m)Mean recovery, %4.224.218.43100.009.684.224.238.50101.1814.224.224.248.3296.714.224.224.248.3898.1114.224.224.228.44100.0014.224.224.228.44100.0010.020.600.611.2198.3614.140.611.2198.3614.140.611.2196.7714.140.611.2196.7714.140.611.2196.7714.140.611.2191.6714.140.611.2110.6714.140.611.2196.7714.140.611.2196.7714.140.611.2196.7714.140.611.2110.6714.140.611.2110.6714.140.611.2110.3314.140.611.5598.7114.140.770.781.5598.710.780.781.5598.710.790.781.5590.000.770.781.5510.000.770.781.5510.300.770.781.5510.300.770.781.5510.30 <trr>0.770.781.5510.30</trr> |

Table 4: Epicatechin, dihydroquercetin, and myricetin contents in Xanthoceras sorbifolia Bunge.

| N | Sample | Epicatechin | | Dihydroquercetin | | Myricetin | |
|---|------------------|--------------------|--------|---------------------|--------|---------------------|--------|
| | | Content, mg/g | RSD, % | Content, mg/g | RSD, % | Content, mg/g | RSD, % |
| 1 | Baotao | 9.339±0.001 | 0.01 | 1.507 ± 0.006 | 0.39 | 0.28±0.00 | 0.00 |
| 2 | Daban | 11.687 ± 0.006 | 0.05 | $1.357 {\pm} 0.005$ | 0.36 | 0.04 ± 0.00 | 0.00 |
| 3 | Hospital of IMMU | 7.61±0.121 | 1.59 | 1.02 ± 0.00 | 0.00 | 2.693 ± 0.006 | 0.22 |
| 4 | Jarud | 6.39 ± 0.06 | 0.94 | 1.13 ± 0.00 | 0.00 | 0.02 ± 0.00 | 0.00 |
| 5 | Jalaid | 6.64 ± 0.00 | 0 | 1.057 ± 0.006 | 0.57 | 0.46 ± 0.005 | 1.08 |
| 6 | Ordos | 8.406 ± 0.058 | 0.69 | $1.833 {\pm} 0.005$ | 0.69 | $0.513 {\pm} 0.005$ | 0.97 |
| 7 | Fuxin | 6.397±0.006 | 0.09 | 1.02 ± 0.000 | 0.00 | 1.703 ± 0.006 | 0.35 |
| | p-value | < 0.001 | | < 0.001 | | < 0.001 | |

The content of epicatechin, dihydroquercetin, and myricetin in trunks and branches of *Xanthoceras sorbifolia* Bunge collected from seven places in Inner Mongolia were different (p<0.001).



Figure 2: TLC of trunks and branches of *Xanthoceras sorbifolia* Bunge (in UV 254 nm) 1. Baotou, 2. Daban, 3. The hospital of IMMU, 4. Jarud, 5. Jalaid, 6. Ordos, 7. Fuxin



Peaks: 1. Epicatechin, 2. Dihydromyricetin, 3. Dihydroquercetin, 4. Myricetin, 5. Quercetin, 6.Naringenin

in Table 3.

Epicatechin, dihydroquercetin, and myricetin contents

Epicatechin, dihydroquercetin, and myricetin contents in trunks and branches of *Xanthoceras sorbifolia* Bunge. from seven places in Inner Mongolia were calculated from the calibration curve (Table 4).

DISCUSSION

Standardization of herbal raw drugs includes passport data of raw plant drugs, botanical authentification, microscopic & molecular examination, identification of chemical composition by various chromatographic techniques, and biological activity of the whole plant²⁰. One of the quality parameters of medicinal plants and traditional powder medicines is the determination of microstructure based on anatomical characterization.²¹ Within the framework of this study, the anatomical characteristics of the trunks and branches of the *Xanthoceras sorbifolia Bunge* were determined, which can be used to detect the raw material in the traditional medicine containing this plant.

Xanthoceras sorbifolia Bunge is rich in biologically active substances, including flavonoids, and researchers isolated many flavonoids from the plant, including epicatechin, dihydromyricetin, myricetin, and



Figure 4: Comparison of *Xanthoceras sorbifolia* Bunge's ethanol extract collected from several parts of Inner Mongolia Peaks: 1. Epicatechin, 2. Dihydromyricetin, 3. Dihydroquercetin, 4. Myricetin, 5. Quercetin, 6. Naringenin; Plants: S1. Baotou, S2. Daban, S3. The hospital of IMUN, S4. Jarud, S5. Jalaid, S6. Ordos, S7. Fuxin



quercetin by Zhang (2000) and Wu (2017), and naringenin by Zhao (2012), and Wu (2017).²²⁻²⁴ Our findings have established TLC \ condition to reveal epicatechin, HPLC condition to reveal epicatechin, dihydroquercetin, quercetin, and naringenin in *Xanthoceras sorbifolia Bunge*. This result can be applied in the quality control of this plant's trunks and branches' raw material. Plant raw material quality parameters, including ash, moisture, extractive value, and the plant's substance content, may differ depending on the growing environment and location. The quality parameters and the content of some flavonoids in the trunks and branches of *X. sorbifolia Bunge* collected from seven different places in Inner Mongolia

were determined, and there were statistically significant differences. As the X. sorbifolia Bunge plant is prepared and used from different places, this should be considered when developing quality documents for this plant. In this study, some flavonoids contained in the X. sorbifolia Bunge were revealed and quantified by HPLC, and the results of this study can be used to develop Xanthoceras sorbifolia Bunge—quality documents. Identifying the plant's marker substance may be necessary by clarifying the active substances by conducting pharmacological studies. Zhang (2015) reported that the compounds epicatechin, catechin, myricetin, and dihydromyricetin, which exist in lignum xanthocerais, showed remarkable protective effects against peroxyl



Figure 6: HPLC Chromatogram of the sample of *Xanthoceras sorbifolia* Bunge (A), standard solution of epicatechin, dihydroquercetin, myricetin (B), and methanol (C)



radical-induced DNA strand scission.¹⁸ Epicatechin was found to be an inhibitory substance against human immunodeficiency virus (HIV-1) protease, and catechin, epicatechin, myricetin, and dihydromyricetin isolated from the wood have an anti-oxidation effect.¹⁷⁻¹⁹ Currently, there is no pharmacopoeia article for *X. sorbifolia Bunge*, so further development of this document is necessary, in which the results of this study can be used.

CONCLUSION

The standardization criteria for the trunk and branches of *Xanthoceras sorbifolia* Bunge were defined. Quality parameters and content of epicatechin, dihydroquercetin, and myricetin were different in trunks and branches of the *Xanthoceras sorbifolia* Bunge collected from seven locations in Inner Mongolia.

CONFLICTS OF INTEREST

The authors have declared no conflicts of interest.

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