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Article

Quantitative Determination and Fingerprint of Phenolic Compounds with Anti-inflammatory Activities for the Quality Control of Mangosteen Extract

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Abstract: The extracts of *Garcinia mangostana* L. (mangosteen) are traditionally used for medicinal purposes in Asia. Phenols and xanthones are the two main phenolic compounds with anti-inflammatory activities in the pericarps of mangosteen. A simple and economic reverse-phase high-performance liquid chromatography (RP-HPLC) method has been developed for the simultaneous quantification of phenols (catechin, epicatechin, and procyanidin B2) in mangosteen. The mobile phase was acetonitrile: water (12:88) at 30°C, the flow rate was 1 mL/min, and the detection wavelength was 230 nm. The results showed good linear relationships for catechin from 50-250 ng and for epicatechin and procyanidin B2 from 250-1250 ng. The RP-HPLC fingerprint determination method of total xanthones in mangosteen was also established to provide a new method for quality control of mangosteen extract. The mobile phase was a methanol-water gradient elution at 30°C, the flow rate was 1 mL/min, and the detection wavelength was 320 nm. The similarity of fingerprints of 10 batches of total xanthones was more than 0.90. These methods had good precision, reproducibility, and stability and can be applied in the quality control of mangosteen extracts for medicinal purposes.

Keywords: mangosteen; phenolic components; HPLC; quantification; fingerprint

1. Introduction

At present, medicinal plants are still an important source of new drug discovery. For example, in recent years, numerous pharmacologically active ingredients have been found in plants, such as mangosteen [1-3]. Mangosteen (*Garcinia mangostana* L.) is a plant of genus Garcinia in the family Guttiferae. It is a well-known tropical fruit referred to as the "Queen of Fruits". Mangosteen is native to the Malay Peninsula and the Malay Archipelago; it is widely cultivated in tropical regions of Asia and Africa, including Thailand, Vietnam, Malaysia, Indonesia, Philippines, and other Southeast Asian countries, and has also been introduced in southern China. In Southeast Asia, the pericarp of mangosteen, which accounts for approximately three-quarters of the total weight of the fruit, is used as medicine and is widely used to treat abdominal pain, diarrhea, trauma, dysentery, purulence, skin infection, ulcer, etc. [4,5]. Researchers have conducted extensive research on the chemical components in mangosteen pericarp and their pharmacological activities, and found that they are rich in xanthone components, such as α-, β-, and γ-mangostin, and confirmed that these components have various pharmacological effects, including

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anti-inflammatory, antitumor, antioxidant, antibacterial, anti-HIV, and anti-Alzheimer's disease activities, and are often used as a plant-based dietary supplement in Western countries [6-11]. Additionally, in recent years, many polyphenols have been found in mangosteen pericarp, including a 4-aryl-2-flavanylbenzopyran derivative, 3,4,3′,5′-tetrahydroxy-5-methoxybenzophenone, and a 2,3-dihydrochromone derivative [12]. Our previous research found that mangosteen pericarp also contains large quantities of polyphenols, such as procyanidins, which possess the same anti-inflammatory activity as α-mangostin and other xanthones. The difference was that these procyanidins had a high affinity with lipopolysaccharide (LPS) and exerted anti-inflammatory effects by binding and neutralizing LPS [13]. In addition, mangosteen pericarp contains volatile components, such as 3-hexen-1-ol, as well as trace elements, including phosphorus, sulfur, magnesium, calcium, and zinc [14]. These studies provide further justification for the use of mangosteen pericarp as a traditional medicine.

Quality control is an important research topic in the process of developing natural medicines, and its purpose is to ensure that the quality of natural medicines meets the requirements for medicinal use. The quality control of natural medicines includes research on the properties, identification, and compositional determination of medicines. Commonly used quality control methods include microscopic identification, thin layer chromatography (TLC), high-performance liquid chromatography (HPLC), gas chromatography (GC), infrared spectroscopy (IR), nuclear magnetic resonance spectroscopy (NMR), and DNA fingerprinting. According to the natural medicines and the types and structural characteristics of their components, the appropriate detection methods are selected [15-17]. With the development of chemical research methods, the study of the quality control of natural medicines is no longer limited to macroscopic research, such as property identification of natural medicines, but includes in-depth research on effective chemical components. The chemical composition that exerts its medicinal effect is the basis for the medicinal effect of natural medicine. It may be a single component or a combination of components. When the quality of the active ingredients of a natural medicine is controlled, the quality of the whole medicine can be well controlled, which can ensure the safety and efficacy of medicines. Therefore, the key to the quality control research of natural medicines is the quality control of their active ingredients.

To better develop mangosteen as a medicinal plant resource, a quality control method is supposed be established for the active ingredients in its pericarp. In this study, according to the characteristics of the main active ingredients in mangosteen pericarp, the HPLC technique was used to establish a method for determining the contents of polyphenols (catechin, epicatechin, and procyanidin B2) and the HPLC chromatographic fingerprint of the total xanthones. This study provides a simple and economic method for the quality control of the active ingredients of mangosteen pericarp.

2. Materials and Methods

2.1. Materials

Mangosteen was purchased from local fruit supermarkets in Chongqing, China. Acetonitrile and methanol (HPLC grade) were purchased from Honeywell Co., Ltd (USA), anhydrous ethanol was purchased from Chongqing Chuandong Chemical (Group) Co., Ltd. (China), and ultrapure water was prepared by the ARE1-500L-U00 pure water system (Aquapro, USA). Catechin, epicatechin, and procyanidin B2 were purchased from Sigma-Aldrich Co., Ltd (USA). α-Mangostin was prepared by our laboratory, and the structure and purity (greater than 98%) were confirmed by ultraviolet (UV) spectrum, HPLC, and NMR measurements (Supplementary Materials Figure S1-S3). The software "Similarity Evaluation System for Chromatographic Fingerprint of Traditional Chinese Medicine" was developed by the Chinese Pharmacopoeia Commission.

2.2. Determination of the polyphenol components in a mangosteen pericarp by HPLC

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2.2.1. HPLC analysis

An Agilent 1200 HPLC system (Agilent, USA) equipped with a G1312A binary pump, G1316A thermostatted column compartment, and G1315D diode array detector (DAD) was used for HPLC analysis. The chromatography column, Agilent ZORBAX SB-C18 reversed-phase column (150 mm × 4.6 mm, 5 μm), was utilized for chromatographic separation. The mobile phase was water:acetonitrile (88:12), the column temperature was 30°C, the flow rate was 1 mL/min, the detection wavelength was 230 nm, and the injection volume was 10 μL.

2.2.2. Preparation of the standard solution

After the appropriate amounts of catechin, epicatechin, and procyanidin B2 were accurately weighed, methanol was added to dissolve the samples, and the solution was mixed evenly to prepare a mixed standard stock solution with a concentration of 0.125 mg/mL. To 10-mLvolumetric flasks, 0.4, 0.8, 1.2, 1.6, 2, 4, 6, 8, or 10 mL of the stock solutions was added, methanol was added to dilute the solution to the concentrations of 0.005, 0.010, 0.015, 0.020, 0.025, 0.050, 0.075, 0.100, and 0.125 mg/mL, respectively. All the solutions were filtered with a 0.22 μm microporous membrane before use.

2.2.3. Preparation of the sample solution

Three parts of mangosteen pericarp samples were crushed, and 5 g was taken from each part and placed in separate 100-mL conical flask, followed by adding 50 mL of methanol. Then, the three samples were subjected to ultrasonic-assisted extraction for 1 h, reflux extraction for 1 h, and cold maceration for 24 h. After the extraction, the samples were allowed to stand at room temperature to cool down and then filtered, and the filtrate was concentrated under reduced pressure using Rotavapor R-205 rotary evaporator (Büchi Company, Switzerland) and dried to obtain the extract. An appropriate amount of the extract was dissolved in methanol to prepare a solution with a concentration of 5 mg/mL. The solution was shaken well and filtered with a 0.22 μm microporous membrane, and 10 μL was accurately injected for HPLC analysis to investigate the extraction effect of different extraction methods.

Two parts of the mangosteen pericarp sample were crushed, and 5 g was taken from each part and placed in separate 100-mL conical flask, followed by adding 50 mL of methanol and ethyl acetate, respectively, and the two samples were subjected to ultrasonic-assisted extraction for 1 h. Then, the samples were allowed to stand at room temperature to cool down, and filtered, and the filtrate was concentrated under reduced pressure and dried to obtain the extract. An appropriate amount of the extract was dissolved in methanol to prepare a solution with a concentration of 5 mg/mL. The solution was shaken well and filtered with a 0.22 μm microporous membrane, and 10 μL was injected for HPLC analysis to investigate the extraction effect of different solvents.

Three parts of mangosteen pericarp samples were crushed, and 5 g was taken from each part and placed in a separate 100-mL conical flask, followed by adding 50 mL of methanol. The three samples were subjected to ultrasonic-assisted extraction for 0.5, 1, and 1.5 h. Subsequently, the samples were allowed to stand at room temperature to cool down, and filtered, and the filtrate was concentrated under reduced pressure and dried to obtain the extract. An appropriate amount of the extract was dissolved in methanol to prepare a solution with a concentration of 5 mg/mL. The solution was shaken well and filtered with a 0.22 μm microporous membrane, and 10 μL was injected for HPLC analysis to investigate the extraction effect of different extraction times.

According to the results of investigating the extraction method, solvent, and time, a method for preparing the sample solution was determined. The mangosteen pericarp sample was crushed, and 5 g of the crushed sample was placed in a 100-mL conical flask, followed by adding 50 mL of methanol. Then, the sample was subjected to ultrasonic-assisted extraction for 1 h. The sample was allowed to stand at room temperature to cool down, and filtered, and the filtrate was concentrated under reduced pressure and dried to obtain the extract. An appropriate amount of the extract was weighed and dissolved in

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methanol to prepare a solution with a concentration of 5 mg/mL. The solution was shaken well and filtered with a 0.22 μm microporous membrane, and 10 μL was injected for HPLC analysis.

2.2.4. Linearity, precision, stability, and repeatability

HPLC analysis was performed on 10 μL of the standard solution for linearity test. The linear regression on the injection amount (x) versus the peak area (y) was performed based upon the chromatograms recorded. A standard curve was plotted, and the regression equation was calculated accordingly.

For precision examination, 10 μL of the standard solution was analyzed for six replicates. The relative standard deviation (RSD) of the peak area of each standard substance was calculated based on the chromatograms recorded.

Stability test were performed on 10 μL of the sample solution that was collected at 0, 2, 4, 6, and 8 h, respectively, after preparation. The RSD of the peak area of each substance was calculated based on the chromatograms recorded.

In terms of repeatability, six replicates of the same batch of mangosteen were prepared, and 10 μL of each sample solution was injected for HPLC analysis. The RSD of the peak area of each substance was calculated based on the chromatograms recorded.

2.2.5. Spike and recovery

Six mangosteen samples with known content were crushed, and 5 g from each sample was placed in a separate conical flask, followed by adding an appropriate amount of standard substance and 50 mL of methanol. The sample was prepared according to the method of sample solution preparation. Then, an appropriate amount of the extract was dissolved in methanol to prepare a solution with a concentration of 5 mg/mL. The solution was shaken well and filtered with a 0.22 μm microporous membrane, and 10 μL was injected for HPLC analysis. The chromatogram was recorded, and calculations were carried out according to the recovery equation to examine the accuracy.

2.2.6. Quantification of catechin, epicatechin, and procyanidin B2

Two batches of mangosteen pericarp samples were randomly selected and crushed, and 5 g was used to prepare a solution of the test product. Subsequently, 10 μL of the solution was injected for HPLC analysis, the chromatogram was recorded, and the content of each component was calculated.

2.3. Establishment of the chromatographic fingerprint of total xanthones by HPLC

2.3.1. HPLC analysis

An Agilent ZORBAX SB-C18 reversed-phase column (150 mm × 4.6 mm, 5 μm) was used, gradient elution was performed with a mobile phase of methanol:water (0 to 32 min, 77:23; 32 to 55 min, 77:23 to 100:0), the column temperature was 30°C, the flow rate was 1 mL/min, and the detection wavelength was 320 nm.

2.3.2. Preparation of the reference solution

An appropriate amount of α-mangostin was dissolved by adding methanol to prepare a solution with a concentration of 0.5 mg/mL, which was used as a fingerprint reference substance.

2.3.3. Preparation of the sample solution

Appropriate amounts of 10 batches of mangosteen total xanthone extract were dissolved in methanol to prepare a solution with a concentration of 1 mg/mL, and filtered with a 0.22 μm microporous membrane before use.

2.3.4. Precision, stability, and reproducibility

HPLC analysis was performed on 10 μL of the sample solution that was analyzed for five replicates for precision test. The sample solutions were examined at 0, 2, 4, 6 and 8hrs, respectively, after preparation for stability test. Five replicates of the sample solution were prepared, and 10 μL of each solution was injected for HPLC analysis. The RSDs of the

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relative retention time and the peak area ratio of each common peak were calculated based on the chromatograms recorded.

2.3.5. Establishment of the chromatographic fingerprint and the similarity calculation

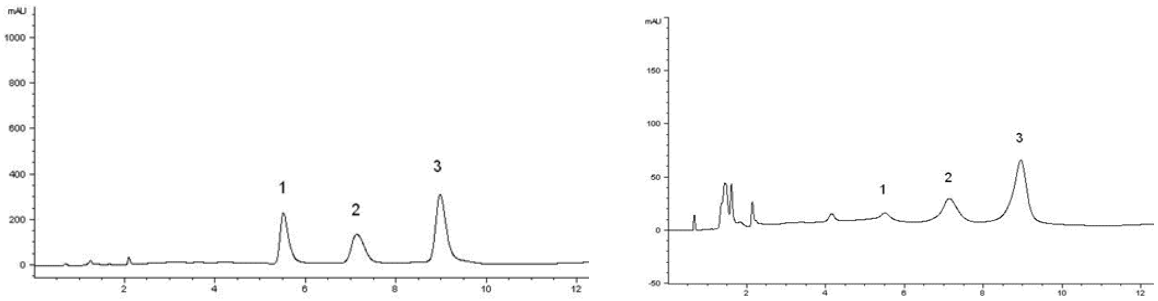
The 10 batches of the sample solution were analyzed by HPLC, the chromatograms were recorded, the average values of characteristic parameters, including the relative retention time and the peak area ratio, were calculated, and the pattern of the chromatographic fingerprint was established by using the "Similarity Evaluation System for Chromatographic Fingerprint of Traditional Chinese Medicine" developed by the Chinese Pharmacopoeia Commission. The similarity of each common peak in the sample solution and pattern of the chromatographic fingerprint was calculated by the cosine ratio method [18], and the similarity of each fingerprint was evaluated.

3. Results

3.1. Quantification of the polyphenols in mangosteen pericarp

3.1.1. HPLC analysis

A C18 reversed-phase chromatography column was used and eluted with water:acetonitrile (88:12), and 230 nm was selected as the detection wavelength based on the ultraviolet (UV) absorption spectrum acquired by DAD. Under this condition, each compound could be efficiently separated from other substances in the sample. Good separation was achieved, and the theoretical plate numbers of catechin, epicatechin, and procyanidin B2 were not less than 3000 (Figure 1).



(**a**) (**b**)

Figure 1. HPLC chromatograms of the standard substances and test samples. (a) Standard substances (1. catechin; 2. procyanidin B2; 3. epicatechin). (b) Test samples (1. catechin; 2. procyanidin B2; 3. epicatechin)

3.1.2. Preparation of the sample solution

The results of investigating the three extraction methods of ultrasound, reflux, and cold maceration showed that the content of each component (chromatographic peak area) in the ultrasonic-assisted extract was the highest (Supplementary Materials Table S1), indicating that the ultrasonic-assisted extraction effect was the best, followed by the reflux extraction method, so ultrasonic-assisted extraction was selected as the extraction method for preparing the sample solution. According to the solubility of the compounds, methanol and ethyl acetate were selected as the extraction solvents. The investigation results suggested that the content of each component (chromatographic peak area) was the highest in the methanol extract (Supplementary Materials Table S2), indicating that methanol used as the extraction solvent could provide the best effect. Therefore, methanol was selected as the extraction solvent for preparing the sample solution. The results of the investigation of the time of ultrasonic-assisted extraction with methanol showed that a satisfactory extraction effect could be achieved within 1 h, after which the content of each component (chromatographic peak area) in the extract did not increase (Supplementary Materials Table S3), so the extraction time for preparing the sample solution was 1 h.

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3.1.3. Linearity, precision, stability, and repeatability

The regression equation of catechin was y = 1.9662x - 5.17 (r = 0.9992), and the linear range was 50-250 ng (Supplementary Materials Table S4). The regression equation of epicatechin was y = 2.1752x - 113.07 (r = 0.9993), and the linear range was 250-1250 ng (Supplementary Materials Table S5). The regression equation of procyanidin B2 was y = 0.8531x + 6.81 (r = 0.9995), and the linear range was 250-1250 ng (Supplementary Materials Table S6).

The RSDs of the peak areas of the catechin, epicatechin, and procyanidin B2 reference substances were 2.26%, 1.88%, and 2.07% (Supplementary Materials Table S7), respectively, indicating a good instrument precision.

The RSD values of the peak areas of catechin, epicatechin, and procyanidin B2 in the sample solution within 8 h were 1.99%, 2.84%, and 1.73% (Supplementary Materials Table S8), respectively, indicating that the sample solution was stable within 8 h. The detection of all samples in the experiment was complete within 8 h.

The RSD values of the contents of catechin, epicatechin, and procyanidin B2 in the sample solution were 1.99%, 1.45%, and 2.96% (Supplementary Materials Table S9), respectively, indicating that the method was reproducible.

3.1.4. Spike and recovery

The experimental results of the spike-recovery test showed that the recoveries of catechin, epicatechin, and procyanidin B2 were 99.3%, 99.5%, and 99.8%, respectively, and the RSDs of the recoveries of catechin, epicatechin, and procyanidin B2 were 2.8%, 2.7%, and 2.7%, respectively, suggesting that the method was accurate (Supplementary Materials Table S10).

3.1.5. Quantification of catechin, epicatechin, and procyanidin B2

Two batches of mangosteen pericarp were measured, and the results showed that the average contents of catechin, epicatechin, and procyanidin B2 in mangosteen pericarp were 0.0065%, 0.0780%, and 0.0775%, respectively (Supplementary Materials Table S11).

3.2. Chromatographic fingerprint of total xanthones

3.2.1. HPLC analysis

A C18 reversed-phase chromatographic column was used, gradient elution was carried out with a mixture of methanol:water, the column temperature was 30°C, the flow rate was 1 mL/min, and 320 nm was finally selected as the detection wavelength based on the UV absorption spectrum acquired by DAD. The chromatograms of the reference substance (α-mangostin) and the test sample are shown in Figure 2.

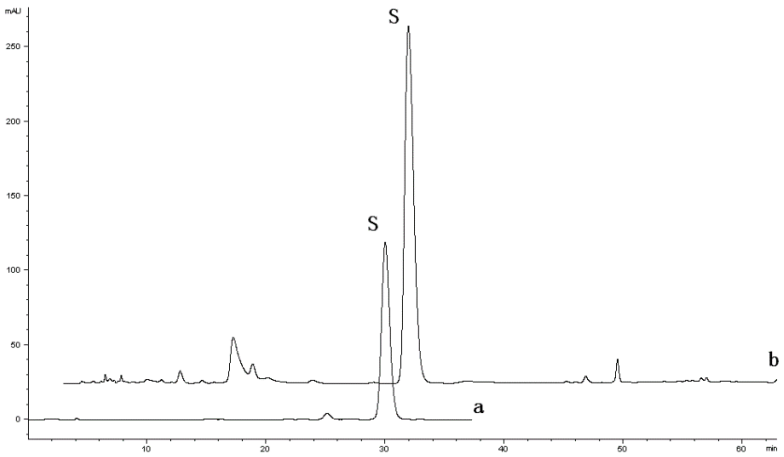

Figure 2. HPLC chromatograms of the reference substance (S) and the test sample. (a) Chromatogram of reference substance S (α-mangostin); (b) Chromatogram of the test sample.

3.2.2. Precision, stability, and reproducibility

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The RSD of the relative retention time and the peak area ratio of each common peak was less than 3% (Supplementary Materials Table S12 and S13), which met the requirements of the chromatographic fingerprint, demonstrating that the instrument had good precision.

The RSD of the relative retention time and the peak area ratio of each common peak of the sample solution within 8 h was less than 3% (Supplementary Materials Table S14 and S15), indicating that the test sample was stable within 8 h, so the detection of all samples in this experiment was complete within 8 h.

The RSD of the relative retention time and the peak area ratio of each common peak was less than 3% (Supplementary Materials Table S16 and S17), which met the requirements of the fingerprint, suggesting that the method had good reproducibility.

3.2.3. Pattern of the chromatographic fingerprint

The common pattern of chromatographic fingerprint of 10 batches of total xanthones in mangosteen pericarp was obtained by using "Similarity Evaluation System for Chromatographic Fingerprint of Traditional Chinese Medicine" (Figure 3), and the pattern of the chromatographic fingerprint (Table 1) was established using cosine ratio method. Among them, seven peaks were found as common fingerprint peaks, and the sum of the area percentages of noncommon peaks was less than 3%, which meets the requirements of the chromatographic fingerprint.

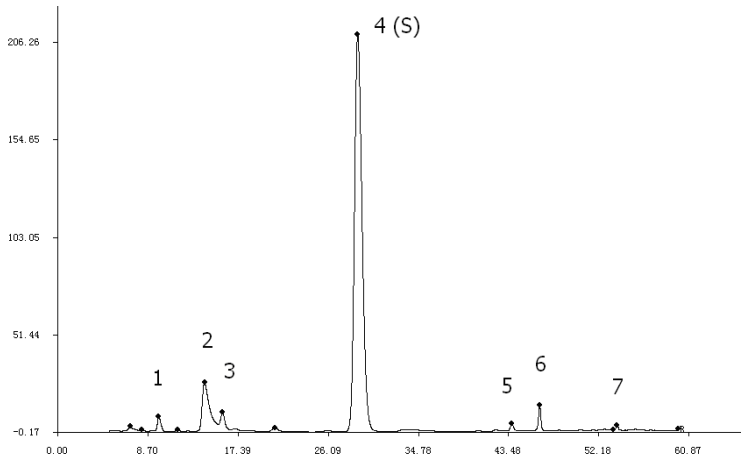

Figure 3. Common pattern of the chromatographic fingerprints of total xanthones in 10 batches of mangosteen pericarp. S (peak No. 4) indicates the reference substance (α-mangostin).

Table 1. Pattern of the chromatographic fingerprints of 10 batches of total xanthones of mangosteen pericarp.

Common peak	Relative retention time	Peak area ratio
1	0.337	0.017
2	0.489	0.115
3	0.549	0.025
4 (S) [1]	1.000	1.000
5	1.514	0.007
6	1.606	0.020
7	1.864	0.004

[1] S: the reference substance (α-mangostin)

3.2.4. Similarity

Using α-mangostin as a reference, the cosine ratio method was used to calculate the similarity between the common fingerprint peaks in the 10 batches of mangosteen samples and the corresponding common peaks in the pattern (Figure 4). The results showed that the 10 batches of mangosteen samples were similar according to the calculation of

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"Similarity Evaluation System for Chromatographic Fingerprint of Traditional Chinese Medicine" (Table 2).

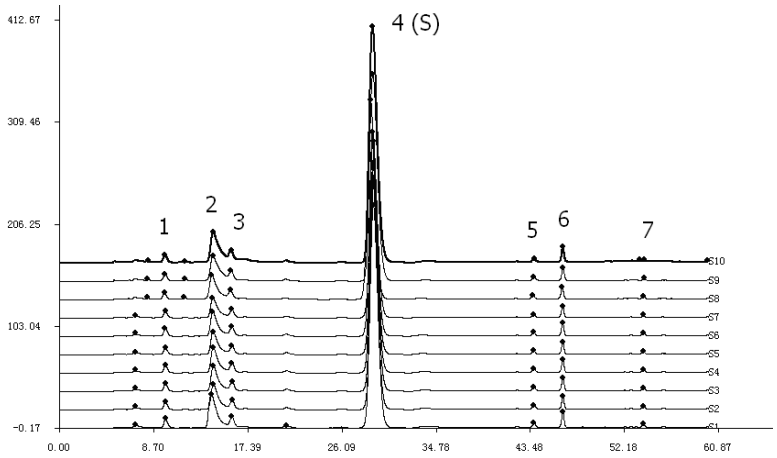

Figure 4. Chromatographic fingerprints of total xanthones in 10 batches of mangosteen pericarp. S (peak No. 4) indicates the reference substance (α-mangostin).

Table 2. Similarity of the chromatographic fingerprints of 10 batches of total xanthones of mangosteen pericarp.

Batch	1	2	3	4	5	6	7	8	9	10	Common pattern
1	/	0.9998	0.9997	0.9997	0.9996	0.9992	0.9990	0.9997	0.9998	0.9999	0.9997
2	0.9998	/	0.9999	0.9999	0.9999	0.9997	0.9996	0.9999	0.9999	0.9999	0.9999
3	0.9997	0.9999	/	0.9999	0.9999	0.9998	0.9997	0.9999	0.9999	0.9999	0.9999
4	0.9997	0.9999	0.9999	/	0.9999	0.9998	0.9997	0.9999	0.9999	0.9999	0.9999
5	0.9996	0.9999	0.9999	0.9999	/	0.9998	0.9997	0.9999	0.9999	0.9999	0.9999
6	0.9992	0.9997	0.9998	0.9998	0.9998	/	0.9999	0.9998	0.9996	0.9996	0.9998
7	0.9990	0.9996	0.9997	0.9997	0.9997	0.9999	/	0.9997	0.9995	0.9995	0.9997
8	0.9997	0.9999	0.9999	0.9999	0.9999	0.9998	0.9997	/	0.9999	0.9999	0.9999
9	0.9998	0.9999	0.9999	0.9999	0.9999	0.9996	0.9995	0.9999	/	0.9999	0.9999
10	0.9999	0.9999	0.9999	0.9999	0.9999	0.9996	0.9995	0.9999	0.9999	/	0.9999
Common pattern	0.9997	0.9999	0.9999	0.9999	0.9999	0.9998	0.9997	0.9999	0.9999	0.9999	/
Cosine ratio	0.9997	0.9999	0.9999	0.9999	0.9999	0.9998	0.9997	0.9999	0.9999	0.9999	/

4. Discussion

As a medicinal and edible plant resource that has been used for a long time in Southeast Asia, mangosteen has also entered other Asian countries and European and American countries in recent years. The study of its active ingredients provides evidence to clarify its pharmacological activity and mechanism of action and lays the basis for the development of new medicinal resources. In addition to the research on active ingredients and their activities, quality control research is also a crucial research work, and it is a necessary monitoring measure to ensure quality. HPLC has the advantages of high separation efficiency, fast analysis speed, high sensitivity, and wide application range, and it is especially suitable for the separation and qualitative and quantitative analysis of complex mixed components [19,20]. In this paper, this technique was used to explore the method for determining the contents of the main active ingredients in mangosteen and the corresponding chromatographic fingerprint, which provided a basis for the quality control and evaluation of mangosteen and was conducive to the further development of this important medicinal plant resource.

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In the study of the method for determining the content of polyphenols, the extraction method, solvent, and time were first investigated, showing that the effect of ultrasonic-assisted extraction with methanol was the best, and it was suitable for experimental operation. Second, in the screening of chromatographic conditions, mobile phases, such as water-methanol, water-acetonitrile, and glacial acetic acid-acetonitrile, were used successively. The results showed that the compounds could be efficiently separated with water:acetonitrile (88:12), the peak shape was good, and the retention time was appropriate. The UV absorption spectrum was acquired using the DAD, and the above four compounds had absorptions at 230 nm and 280 nm, with 230 nm the maximum absorption wavelength. Although 230 nm was close to the short-wavelength end absorption peak, the selected mobile phase (water:acetonitrile) had a low cutoff wavelength (190 nm). Additionally, under this chromatographic condition, the interference was small, and baseline separation could be achieved for the components, so 230 nm was finally selected as the detection wavelength for this method after considering the detection sensitivity. At present, no method has been reported for simultaneously determining the contents of catechin, epicatechin, and procyanidin B2 in mangosteen pericarp. In this study, an HPLC method was established to simultaneously determine the contents of the above three polyphenol components. The methodological investigation showed that the method was simple, accurate, and suitable for the quality control of the active ingredients of mangosteen, laying a foundation for the further development of the medicinal value of mangosteen.

A method for determining the content of individual xanthone components in mangosteen pericarp, such as α-mangostin, has been reported [21,22], and in the present study, the HPLC chromatographic fingerprint of the total xanthones in mangosteen pericarp was explored, including HPLC conditions and a method validation, and the quality control of total xanthones was studied more comprehensively. α-Mangostin is the indicator component with the highest content in the total xanthones of mangosteen, and it is the main active ingredient of this part. Therefore, it was selected as the reference substance for the chromatographic fingerprint of total mangosteen xanthone, and the chromatographic fingerprint of total xanthones in mangosteen pericarp was established by HPLC. In the experiment of chromatographic conditions screening, different ratios of mobile phases, such as water-methanol, water-acetonitrile, phosphoric acid-methanol, and phosphoric acid-acetonitrile, were used. The results showed that the water-methanol gradient elution could meet the experimental requirements, with many eluted peaks and good resolution, the recording time of the spectrum was 1 h, and the retention time of each component was suitable. The detection wavelength was selected in combination with the acquired UV absorption spectrum and literature reports. The absorbances of the xanthone components in mangosteen were mainly found at approximately 205 nm, 240 nm, and 320 nm. Considering the number of peaks, sensitivity, interference, and other factors, 320 nm was finally selected as the detection wavelength. Through the analysis of characteristic parameters, such as the relative retention time and the peak area ratio, seven common fingerprint peaks were calibrated, and the sum of noncommon peak areas was less than 3%, which met the technical requirements of the chromatographic fingerprint. Consequently, the "Similarity Evaluation System for Chromatographic Fingerprint of Traditional Chinese Medicine" was used to establish the pattern standard of the chromatographic fingerprint of total xanthones of mangosteen. The similarity of the fingerprint peaks of each batch was calculated by the cosine ratio method, and the similarity of 10 batches of samples was evaluated, providing a scientific method for the study of quality control of the total xanthones of mangosteen.

5. Conclusions

In this study, by exploring a method for quantitative determination and chromatographic fingerprint of the two main anti-inflammatory active ingredients, polyphenols and xanthone components, in mangosteen pericarp, a simple and economic HPLC method for the quality control of mangosteen pericarp extract has been developed, providing the

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theoretical and experimental basis for further development of this medicinal plant resource.

Supplementary Materials: The following supporting information can be downloaded at: www.mdpi.com/xxx/s1, Figure S1: The UV spectrum of α-mangostin; Figure S2: The HPLC chromatogram of α-mangostin; Figure S3: The ^{1}H NMR spectrum of α-mangostin; Table S1: The evaluation of different methods of extraction; Table S2: The evaluation of different solvents of extraction; Table S3: The evaluation of different extraction time; Table S4: The linear relationship of catechin; Table S5: The linear relationship of epicatechin; Table S6: The linear relationship of procyanidin B2; Table S7: The result of precision; Table S8: The result of stability; Table S9: The result of repeatability; Table S10: The result of recovery; Table S11: The results of quantitative determination of catechin, epicatechin, and procyanidin B2; Table S12: The result of precision (relative retention time of common peaks); Table S13: The result of precision (peak area ratio of common peaks); Table S14: The result of stability (relative retention time of common peaks); Table S15: The result of stability (peak area ratio of common peaks); Table S16: The result of reproducibility (relative retention time of common peaks); Table S17: The result of reproducibility (peak area ratio of common peaks).

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