Eur J Drug Metab Pharmacokinet
DOI 10.1007/s13318-015-0283-4

ORIGINAL PAPER

A Method of Effectively Improved α-Mangostin Bioavailability

Yan Zhao[1] · Guosheng Tang[1] · Qiang Tang[1] · Jing Zhang[1] · Yingying Hou[1] · Enbo Cai[1] · Shuangli Liu[1] · Daihong Lei[1] · Lianxue Zhang[1] · Shijie Wang[2]

Received: 18 December 2014 / Accepted: 30 April 2015

Abstract α-Mangostin, a major xanthone isolated from the pericarp of *Garcinia mangostana*, exhibits anti-inflammatory and antitumor effects. Its absolute bioavailability is low, with minimal oral absorption. In this article, a soft capsule, with vegetable oil as the dispersion matrix, was prepared to improve the bioavailability of α-mangostin. Its pharmacokinetics and tissue distribution were determined in rats. An HPLC assay was established to determine the concentration of α-mangostin in biological samples. The validated method was used successfully to support pharmacokinetic and tissue distribution studies of α-mangostin in rats after intravenous (i.v.) and oral administration. The pharmacokinetic study found the absolute bioavailabilities of low, medium and high doses were 61.1, 51.5 and 42.5 %, respectively, indicating that the absolute bioavailability was effectively improved.

Y. Zhao, G. Tang and Q. Tang contributed equally to this work.

Electronic supplementary material The online version of this article (doi:10.1007/s13318-015-0283-4) contains supplementary material, which is available to authorized users.

✉ Yan Zhao
zhyjlu79@163.com

✉ Lianxue Zhang
zlx863@163.com

[1] College of Chinese Medicinal Materials, Jilin Agriculture University, No. 2888 Xincheng Street, Changchun 130118, Jilin, China

[2] College of Traditional Chinese Medicine, Jilin Agricultural Science and Technology University, No 77, Hanlin Road, Jilin 132101, Jilin, China

1 Introduction

Mangosteen *Garcinia mangostana* is a common fruit found in Southeast Asia, e.g., in Thailand, Myanmar, Malaysia, Philippines, Sri Lanka and India, and can also be found in many other countries worldwide [1]. Mangosteen is known as "the queen of fruits" because it is one of the best tasting tropical fruits. The fruit is dark purple or reddish, with white, soft and juicy edible pulp with a slightly acidic and sweet flavor and a pleasant aroma [2]. Its pericarp has long been used as a traditional medicine for the treatment of diarrhea, trauma, skin infection and wounds [3–5]. Phytochemical studies have shown that it contains a variety of major secondary metabolites such as prenylated and oxygenated xanthones [6–10]. Currently, researchers have identified and isolated about 200 xanthones; of these, 40 species were found in mangosteen pericarps. α-Mangostin is one of the major xanthones (78 % total xanthone content) [11] and is isolated from mangosteen pericarps. Numerous studies have shown that α-mangostin possesses anti-inflammatory and antitumor activities [12–16]. The *G. mangostana* xanthones are gaining more interest because of their remarkable pharmacological effects, including not only anticancer [17], but also analgesic [18], antioxidant [19], anti-inflammatory [20], antiallergy [21], antimicrobial, antituberculosis [9], antifungal [22, 23], antiviral [24], cytotoxic [25], cardioprotective [26], neuroprotective [27] and immunomodulation [28] effects.

Pharmacokinetic studies are known to play an increasingly important role in the drug discovery and development process, from toxicity and clinical studies to the optimization of candidate drugs [29]. The pharmacokinetic study of a bioactive component can help us understand its in vivo actions and explain a variety of events related to the efficacy and toxicity of the relevant herbs or herbal

Published online: 20 May 2015

preparations in which this constituent is found [30]. Therefore, it is of clinical importance to explore the in vivo pharmacokinetic profiles of α-mangostin in rats. The pharmacokinetic (PK) characteristics of α-mangostin have been explored by Li L, Li G and Petiwala [31–34]. The studies of Petiwala [33] showed evidence of phase II metabolism and the presence of up to four glucuronides of α-mangostin. The studies of Li L [32] showed that the absolute bioavailability of α-mangostin was low and exhibited minimal oral absorption. Thus, the purpose of the present study was to explore the pharmacokinetics and tissue distribution of α-mangostin soft capsules. The results suggest that a capsule can improve the bioavailability and provide a meaningful basis for clinical application. On the other hand, because of other xanthones, such as α-mangostin analog, the achieved pharmacokinetic and tissue distribution results may be useful.

2 Materials and Methods

2.1 Materials and Reagents

Mangosteens were purchased from Vorrarat Fresh Fruit Co., Ltd. (Thailand), and identified as *G. mangostana* by Prof. Lianxue Zhang (Jilin Agriculture University, Jilin, China). α-Mangostin was isolated from ethyl acetate extract of the fruit hull of *G. mangostana* (1.0 kg) as follows: The dried and pulverized fruit hulls were powdered and extracted with ethyl acetate (EtOAc) three times at 50 °C. Afterwards, after filtration, the combined extract was concentrated under reduced pressure to yield it as a yellowish solid (538 g). A portion of the extract was subjected to repeated column chromatography over silica gel using a hexane/acetone gradient. This yielded the pure major compound, α-mangostin, including other minor xanthones. The structure was identified by comparing its physicochemical and spectroscopic (MS and NMR) data [35]. Its purity was determined to be over 98.0 % by HPLC. Vegetable oil (soybean oil, containing palmitic acid 6 %, oleic acid 29 %, stearic acid 3 %, linoleic acid 55 % and linolenic acid 2 %), gelatin, sorbitol, glycerol and titania (food grade) were used. Methanol (HPLC grade) was purchased from Honeywell Burdick & Jackson (Ulsan, Korea). The ultra-high-purified water used in this study was prepared in a Milli-Q water purification system (Millipore, Bedford, MA, USA). Other chemicals used were of analytical grade.

2.2 Capsules

α-Mangostin was dissolved in vegetable oil in 15 mg/ml proportions. Each capsule contained 11.25 mg α-mangostin and vegetable oil as a dispersion matrix. The capsule was suitable for human consumption. The capsule had the following composition: α-mangostin, vegetable oil, 40.5 % gelatin, 10.1 % sorbitol, 15.2 % glycerol, 33.4 % water and 0.8 % titania. The α-mangostin concentration was diluted with vegetable oil to 0.07 mg/ml, 0.27 mg/ml and 1.0 mg/ml, respectively. Then the rats were fed at 1.5 ml/100 g body weight at the beginning of the experiment, i.e., 1.025 mg/kg, 4.100 mg/kg and 16.400 mg/kg. We did this becuase we observed that the mangosteen extract, when administered at 50, 100 and 200 mg/kg, could decrease the blood lipids in hyperlipidemia (the content of α-mangostin in the mangosteen extract was about 10 %) [36]. The chosen of dosage for the *G. mangostana* capsules is also recommended (2009L11444, second category new drugs of traditional Chinese Medicine).

2.3 Animals

Sprague-Dawley rats weighing 200–220 g were supplied by the Animal Experiment Center of Jilin University (Jilin, China). The animals were housed under controlled temperature (25 ± 2 °C) and relative humidity (40–70 %), using a 12-h light/dark cycle for 7 days before starting the experiments, and fed with standard laboratory food and water ad libitum except for fasting 12 h prior to the experiment. All experimental procedures adopted in this study were previously approved by the ethics committee for research on laboratory animal use of the institution [no. SCXK(Ji)2008-0005].

2.4 Instrumentation and Chromatographic Conditions

The HPLC analysis was carried out on an ALLTECH series liquid chromatographic system equipped with a Model 626 HPLC Pump, Model 631 Column Heater thermostatted column compartment and UVIS-201 UV detector. Data acquisition was controlled by ATTACH ChemStation SS420X software. Chromatographic separation was accomplished on a Dikma Diamonsil^{TM} C_{18} (250 mm × 4.6 mm, 5 μm) analytical column (Dikma Technologies Co., Ltd., Beijing, China). The mobile phase was methanol–water (95:5, v/v) at a flow rate of 1.0 ml/min. Chromatograms were monitored at 317 nm, and the column temperature was maintained at 25 °C.

2.5 Calibration Standard and Quality Control (QC) Sample Preparation

Stock solution of α-mangostin was prepared in 95 % methanol to give a final concentration of 1.024 mg/ml. A series of working solutions was obtained by diluting the α-mangostin stock solution with 95 % methanol. All solutions were stored at 4 °C until used. Calibration standards

of α-mangostin were prepared by spiking the appropriate amount of the working solutions into 100 μl drug-free rat serum or tissue homogenates. The final concentrations of the calibration standard samples were 0.04, 0.2, 0.5, 2.5, 10 and 40 μg/ml. Quality control (QC) samples were prepared at low, medium and high concentrations of 0.04, 2.5 and 40 μg/ml for plasma and different tissue homogenates in the same manner as the calibration standards.

2.6 General Procedure of Sample Preparation for α-Mangostin Analysis in Serum and Tissue Samples

A liquid-liquid extraction (LLE) of α-mangostin in biosamples was performed prior to HPLC analysis. Briefly, 100 μl of serum or tissue was added to centrifuge tubes and extracted twice with 900 μl methanol by vortexing for 3 min. After centrifugation at 3000 rpm for 10 min, the upper organic layer was transferred to a clean tube and evaporated to dryness under a gentle stream of nitrogen at 40 °C. The residue was reconstituted in 100 μl of 95 % methanol and centrifuged at 3000 rpm for 10 min. An aliquot (20 μl) of supernatant was injected onto the HPLC system.

2.7 Method Validation

2.7.1 Specificity

Specificity was assessed by analyzing blank serum and tissue homogenate samples, serum and tissue homogenate samples spiked with α-mangostin, and rat serum and tissue samples after oral administration of α-mangostin.

2.7.2 Calibration Curves, Limit of Detection (LOD) and Lower Limit of Quantification (LLOQ)

The calibration standard was prepared as described above in triplicate and analyzed on 3 consecutive days. Calibration curves were constructed by plotting the peak area/concentration of α-mangostin. The limit of detection (LOD) was determined using a signal-to-noise ratio (S/N) of 3:1 by comparing test results from samples with known concentrations of the analyte with those from the blank samples. The lower limit of quantification (LLOQ) was defined as the lowest concentration of analyte in a sample that provided a peak area with a signal-to-noise ratio higher than 10.

2.7.3 Precision and Accuracy

Intra-day precision and accuracy were evaluated by analysis of the three QC samples with six determinations per concentration on the same day, while the inter-day precision and accuracy were measured over 3 consecutive days. The precision was defined as the standard deviation, mean ± SD (%), while accuracy was determined by calculating the percentage deviation observed in the analysis of QC samples and expressed by relative error (RE%). The accepted criteria for the data were that the precision and accuracy should not exceed 15 %, except at the LLOQ where it should not exceed 20 %.

2.7.4 Extraction Recovery and Stability

The extraction recoveries of α-mangostin were determined at low, medium and high levels of QC samples. Recoveries were calculated by comparing the observed peak area ratios in biosamples to those non-processed standard solutions at the same concentrations. The stability of α-mangostin in serum and tissue was determined under different storage or handling conditions. The short-term stability at ambient temperature (25 ± 2 °C) was performed by processing and analyzing rat plasma and tissue homogenate samples after 12 h exposure at room temperature. Freeze-thaw stability was investigated by freezing the QC samples at −80 °C and thawing at 25 °C for a total of three freeze-thaw cycles. Samples were frozen for at least 12 h at −80 °C in each cycle. The long-term stability was evaluated by determining QC samples of the plasma and tissues homogenate kept at low temperature (−80 °C) for 30 days.

2.8 In Vivo Pharmacokinetic Study

For pharmacokinetic studies, 36 male rats were randomly assigned to six groups ($n = 6$ per group). α-Mangostin was administered by i.v. injection via the lateral sublingual vein and oral administration at the dose of 1.025, 4.100 and 16.400 mg/kg, respectively. At the time points of 2, 5, 10, 20, 40 min, 1, 2, 4, 6, 8, 12 and 24 h post administration, blood samples (0.5 ml) were collected in tubes from the orbital vein (making sure to use six animals from one group at every time point). After a water bath at 37 °C for 30 min, the samples were centrifuged at 2,500 rpm for 10 min to obtain the serum. The serum was stored for HPLC analysis.

2.9 Tissue Distribution Study

To study the tissue distribution of α-mangostin, 18 rats were randomly divided into three groups (3 males and 3 females per group), and α-mangostin was administered orally at a dose of 16.400 mg/kg. The rats were killed 20 min, 1.0 and 4.0 h following administration under ether anesthesia. Tissue specimens, including the heart, liver, spleen, lung, kidney, stomach, small intestine, muscle, fat,

brain, bone marrow, bone, prostate, pancreas, testis, ovary and uterus, were collected. The heart, lung, kidney, stomach, small intestine and uterus were flushed to remove contents. Tissue samples were rinsed in saline and blotted dry with filter paper, then weighed for wet weight and homogenized in ice-cold physiological saline solution (500 mg/ml). The obtained tissue homogenates were stored at −80 °C until analysis was performed using the above-described procedure.

2.10 Statistical Analysis

The pharmacokinetic parameters were calculated using the 3P97 software (Chinese Pharmacology Society, Beijing, China). An appropriate pharmacokinetic model was chosen based on the lowest Akaike information criterion (AIC) value, lowest weighted squared residuals, lowest standard errors of the fitting parameters and dispersion of the residual under an equal weight scheme [37, 38]. All the data were expressed as the mean ± standard deviation, and the levels of statistical significance were assessed using Student's *t* test.

3 Results and Discussion

3.1 Preparation of Serum and Tissue Samples

Biosample preparation was performed using liquid-liquid extraction (LLE), and different extraction solvents, such as ethyl acetate, chloroform, diethyl ether, hexane, methyl tert-butyl ether, methanol and ethanol, were investigated. Finally, methanol was found to be optimal, because it could produce clean chromatograms for serum and tissues and yielded the highest recovery of the analytes. The recovery of ethyl acetate, chloroform, diethyl ether, hexane, methyl tert-butyl ether, methanol and ethanol are shown in Table S1.

3.2 Method Validation

3.2.1 Specificity

The representative chromatograms for determination of α-mangostin in serum and tissues were almost the same, so only one chromatogram was chosen as an example in Figure S1. The retention time of α-mangostin was about 4.5 min. The analytes were well separated, and no interference from endogenous substances or metabolites was detected.

3.2.2 Linearity of the Calibration Curve, Limit of Detection and Lower Limit of Quantification

The calibration curves showed good linearity over the concentration range of 0.04–40 μg/ml in rat serum and most of tissue homogenates, including the heart, spleen, lung, kidney, fat, testis, marrow, muscle, bone, uterus and prostate, and 0.08–80 μg/ml in the liver, brain, ovary, small intestine and stomach with a correlation coefficient (R) larger than 0.999. Typical linear regression equations, correlation coefficients and linear ranges of α-mangostin in serum and each tissue are listed in Table S2. The current assay offered a limit of detection of 0.01 μg/ml and a lower limit of quantification of 0.04 μg/ml in serum and tissue samples. The limits were sufficient for studies of pharmacokinetics and tissue distribution following oral and i.v. administration of α-mangostin.

3.2.3 Precision and Accuracy

The precision and accuracy values of the method are summarized in Table 1. All the samples were spiked with analytes at three concentration levels, the RSD% of both intra-day and inter-day precision was below 10.11 %, and the accuracy was within the range of −11.10 to 11.02 %. The results demonstrated that the method was accurate and reproducible for the determination of α-mangostin in rat serum and tissues.

3.2.4 Recovery and Stability

The extraction recoveries of α-mangostin ranged from 88.82 to 99.64 % in serum and tissue samples (Table 1). These data indicated the biosample preparation procedure was satisfactory and could achieve acceptable extraction recovery. The stability tests were designed to cover the anticipated conditions that the samples might experience. The results are summarized in Table 2. The results show the α-mangostin concentration was not significantly changed in serum and tissue in samples stored in three kinds of conditions. There appeared to be less loss in samples stored for long times compared to the other two conditions; the reason may be that the enzyme in the biological samples played a role at room temperature.

3.3 Pharmacokinetics of α-Mangostin in Rats

3.3.1 Pharmacokinetics of α-Mangostin after I.V. Administration

The mean serum concentration-time profiles of α-mangostin after intravenous administration (1.025, 4.100 and 16.400 mg/kg) are shown in Fig. 1a, with pharmacokinetic parameters listed in Table 3. The concentration-time data after intravenous administration in rats were best fitted to the two-compartment model. α-Mangostin could be detected until 24 h post dosing using the above-described analytical method. Rapid absorption followed by a slow

Table 1 Precision, accuracy and recovery of the method used for determination of α-mangostin concentrations in rat serum and tissue samples ($n = 6$)

Biosamples	Concentration (μg/ml)	Intra-day		Intra-day		Recovery
		Precision (RSD%)	Accuracy (RE%)	Precision (RSD%)	Accuracy (RE%)	Mean ± SD (%)
Serum	0.04	6.52 ± 0.35	7.82 ± 0.43	7.23 ± 0.31	8.55 ± 0.68	98.42 ± 4.75
	2.5	7.41 ± 0.43	−8.24 ± 0.28	6.90 ± 0.24	−9.26 ± 0.38	99.54 ± 3.98
	40	5.43 ± 0.25	6.95 ± 0.51	5.37 ± 0.34	6.85 ± 0.55	97.32 ± 5.45
Liver	0.04	7.31 ± 0.38	8.53 ± 0.52	5.58 ± 0.24	7.23 ± 0.46	89.61 ± 8.56
	2.5	8.81 ± 0.69	9.15 ± 0.42	10.11 ± 0.72	−10.24 ± 0.62	92.33 ± 9.44
	40	9.32 ± 0.52	−10.03 ± 0.65	8.37 ± 0.47	−9.34 ± 0.51	96.24 ± 9.22
Lung	0.04	8.23 ± 0.53	−10.03 ± 0.38	9.03 ± 0.69	−11.1 ± 0.75	91.23 ± 9.42
	2.5	10.52 ± 0.61	−11.01 ± 0.72	6.48 ± 0.52	8.93 ± 0.64	95.37 ± 8.76
	40	6.75 ± 0.38	9.82 ± 0.29	9.23 ± 0.35	10.1 ± 0.51	98.94 ± 8.45
Pancreas	0.04	7.99 ± 0.32	10.21 ± 0.48	7.59 ± 0.66	8.37 ± 0.57	88.82 ± 8.75
	2.5	9.89 ± 0.67	11.02 ± 0.75	4.74 ± 0.27	−6.43 ± 0.36	91.45 ± 9.77
	40	4.96 ± 0.31	9.78 ± 0.77	5.32 ± 0.35	8.14 ± 0.72	97.55 ± 9.54
Small intestine	0.04	10.02 ± 0.53	10.14 ± 0.54	9.41 ± 0.63	10.34 ± 0.76	89.77 ± 8.79
	2.5	9.23 ± 0.75	9.84 ± 0.62	8.57 ± 0.48	−9.36 ± 0.26	94.93 ± 8.87
	40	9.42 ± 0.77	−10.30 ± 0.53	7.49 ± 0.38	−8.38 ± 0.57	98.77 ± 8.54
Fat	0.04	8.92 ± 0.47	9.34 ± 0.58	6.54 ± 0.54	10.81 ± 0.74	90.23 ± 9.45
	2.5	8.21 ± 0.66	−8.43 ± 0.55	8.27 ± 0.53	−9.62 ± 0.44	94.66 ± 9.45
	40	8.48 ± 0.54	9.12 ± 0.88	8.48 ± 0.36	8.73 ± 0.41	99.64 ± 9.72

RE% stands for relative error

Table 2 Stability of α-mangostin in rat serum and tissue samples ($n = 6$)

Biosamples	Concentration (μg/ml)	Accuracy (RE%)		
		Short-term stability	Freeze-thaw stability	Long-term stability
Serum	0.04	−9.33 ± 0.36	6.32 ± 0.44	11.08 ± 0.62
	2.5	−10.14 ± 0.44	8.42 ± 0.46	−9.43 ± 0.66
	40	8.11 ± 0.55	−9.73 ± 0.36	7.78 ± 0.33
Liver	0.04	−10.23 ± 0.62	−10.30 ± 0.46	7.78 ± 0.38
	2.5	9.13 ± 0.47	9.54 ± 0.55	−6.23 ± 0.33
	40	−7.55 ± 0.46	−8.83 ± 0.37	−5.98 ± 0.65
Lung	0.04	5.46 ± 0.23	7.61 ± 0.35	−7.9 ± 0.32
	2.5	7.67 ± 0.22	−10.53 ± 0.64	9.45 ± 0.53
	40	9.21 ± 0.43	−9.44 ± 0.32	11.21 ± 0.53
Pancreas	0.04	5.67 ± 0.21	−4.90 ± 0.23	6.75 ± 0.44
	2.5	8.56 ± 0.43	−8.45 ± 0.53	7.87 ± 0.34
	40	−6.55 ± 0.38	10.33 ± 0.54	9.22 ± 0.34
Small intestine	0.04	9.20 ± 0.57	8.51 ± 0.65	8.98 ± 0.41
	2.5	−6.88 ± 0.34	8.25 ± 0.34	7.40 ± 0.25
	40	−7.52 ± 0.46	−7.21 ± 0.44	−7.41 ± 0.45
Fat	0.04	9.63 ± 0.52	−5.78 ± 0.37	6.25 ± 0.29
	2.5	−11.13 ± 0.67	9.19 ± 0.77	8.15 ± 0.53
	40	−7.40 ± 0.36	7.58 ± 0.33	−6.83 ± 0.28

RE% stands for relative error

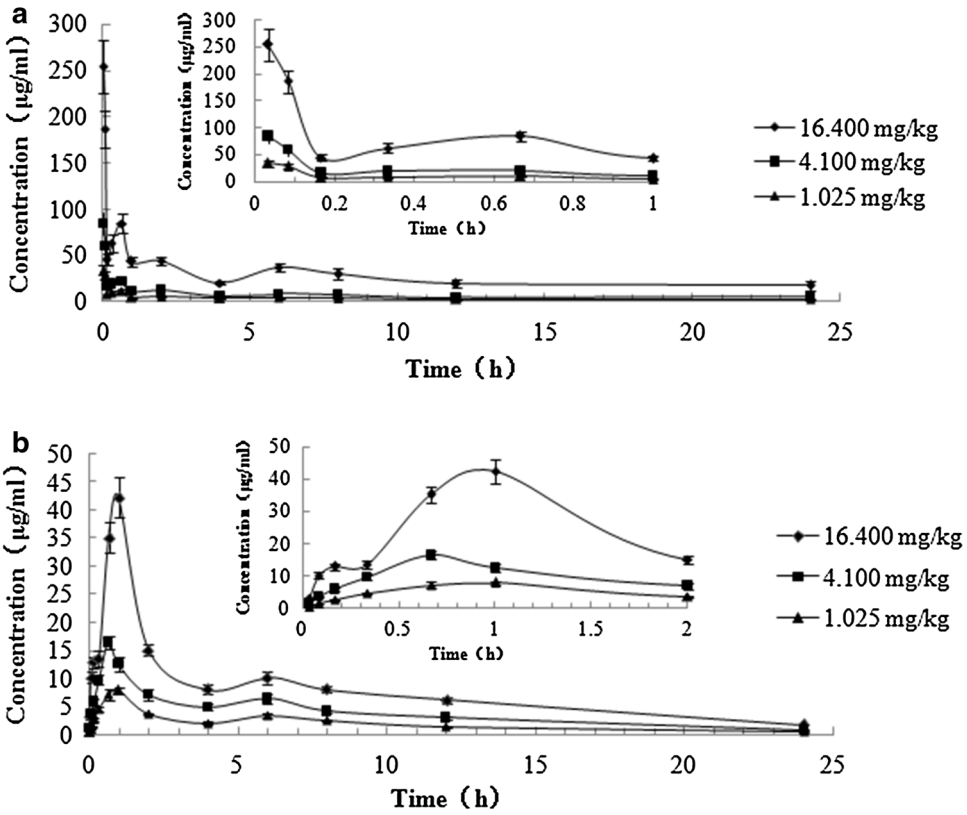

Fig. 1 **a** Mean serum concentration-time curves of α-mangostin after i.v. ($n = 6$) administration at doses of 1.025, 4.100 and 16.400 mg/kg. **b** Mean serum concentration-time curves of α-mangostin after intravenous oral ($n = 6$) administration at doses of 1.025, 4.100 and 16.400 mg/kg

Table 3 Pharmacokinetic parameters of α-mangostin in rat serum after single i.v. administration of α-mangostin at 1.025, 4.100 and 16.400 mg/kg ($n = 6$)

Parameter	Low dose	Medium dose	High dose
Atrioventricular number	2	2	2
A (μg/ml)	59.515	148.675	471.083
α (1/h)	17.021	17.862	20.184
B (μg/ml)	5.627	10.123	37.134
β (1/h)	0.057	0.048	0.041
V_c (mg/kg)/(μg/ml)	0.015	0.025	0.032
$t_{1/2\alpha}$ (h)	0.040	0.038	0.034
$t_{1/2\beta}$ (h)	11.982	14.370	16.734
K21 (1/h)	1.523	1.183	1.513
K10 (1/h)	0.646	0.727	0.552
K12 (1/h)	14.909	15.999	18.160
AUC (μg/ml)*h	100.780	218.212	919.889
CL(s) (mg/kg)/h*(μg/ml)	0.010	0.018	0.017

elimination phase in plasma was observed from the compartment model parameters $t_{1/2\alpha}$ and $t_{1/2\beta}$. The steady apparent volume of distribution was $V_{ss} = V_c \times (1 + K12/K21)$; the data of the low, medium and high-dose group were 0.162, 0.363 and 0.416 l/kg. V_c, the apparent volume of distribution of the central compartment, was 0.040, 0.038 and 0.034 l/kg, which was 24.76, 10.46 and 8.17 % of the total volume of distribution, respectively. These showed that the drugs were mainly distributed in the major circulation and tissues rich in blood flow. The parameters of the elimination rate constant of the central compartment (K10) as well as the transport rate constant from the central to peripheral compartment (K12) and from the peripheral to central compartment (K21) reflected that α-mangostin was eliminated largely from the central compartment of the body. A dose proportionality study indicated that there was good correlation between the AUC and dose. A significant difference in the AUC among the three doses was observed ($p < 0.05$). On the other hand, there was no significant difference in systemic clearances (CLs) at the three dose levels, suggesting that α-mangostin may have linear pharmacokinetic characteristics in rats within the dose ranges tested.

3.3.2 *Pharmacokinetics of α-Mangostin after Oral Administration*

Furthermore, we also conducted a pharmacokinetic study of α-mangostin given to rats by the oral route. The mean serum concentration-time profiles of α-mangostin after oral administration (1.025, 4.100 and 16.400 mg/kg) are shown in Fig. 1b, with pharmacokinetic parameters listed in Table 4. The data were found to fit best with a three-

Table 4 Pharmacokinetic parameters of α-mangostin in rat serum after single oral administration of α-mangostin at 1.025, 4.100 and 16.400 mg/kg ($n = 6$)

Parameter	Low dose	Medium dose	High dose
Atrioventricular number	3	3	3
P (μg/ml)	2.428	6.671	14193.053
γ (1/h)	1.447	1.177	2.092
A (μg/ml)	5.252	8.627	16556.740
α (1/h)	0.979	1.034	2.090
B (μg/ml)	4.244	7.177	14.059
β (1/h)	0.107	0.081	0.080
Ka (1/h)	2.869	3.158	2.094
Lag time (h)	0.017	0.011	0.002
V_c (mg/kg)/(μg/ml)	0.080	0.205	0.160
$t_{1/2pi}$ (h)	0.478	0.588	0.331
$t_{1/2\alpha}$ (h)	0.708	0.669	0.332
$t_{1/2\beta}$ (h)	6.476	8.554	8.712
K12 (1/h)	−0.874	−0.880	−0.914
K21 (1/h)	1.351	1.121	2.092
K13 (1/h)	1.304	1.379	2.186
K31 (1/h)	0.545	0.495	0.636
K10 (1/h)	0.205	0.177	0.262
AUC (μg/ml)*h	61.565	112.426	390.982
CL(s) (mg/kg)/h*(μg/ml)	0.016	0.036	0.042
$t_{1/2Ka}$(h)	0.241	0.219	0.331

compartment model. α-Mangostin had a fast distribution phase ($t_{1/2\alpha}$, 0.332–0.708 h) followed by a relatively slow elimination phase ($t_{1/2\beta}$, 6.476–8.712 h) and could be detected until 24 h post dosing using the above-described analytical method. A dose proportionality study indicated that there was good correlation between the AUC and dose. A significant difference in the AUC among the three doses was observed ($p < 0.05$). On the other hand, there was no significant difference in systemic clearances (CLs) at the three dose levels, suggesting that α-mangostin might have linear pharmacokinetic characteristics in rats within the dose ranges tested.

3.3.3 Bioavailability of α-Mangostin

Mouse experiments by Li and Kondo [32, 39] showed a different T_{max} and half-life suggesting a possible benefit of giving a mixture of xanthones compared to pure α-mangostin. In this regard, Bumrungpert et al. [40] came to a similar conclusion. The authors used Caco-2 cells to investigate the digestive stability, bioaccessibility and intestinal cell transport of α- and β-mangostin from digested mangosteen pericarp and fruit pulp. Transfer of α- and β-mangostin to the aqueous fraction during simulated digestion was efficient (65–74 %). In our research, when α-mangostin dispersed in oil was administered intragastrically, the absolute bioavailability was calculated by the formula of $F = \text{AUCpo/AUCiv}$, and the absolute bioavailabilities of the three dose groups were 61.1, 51.5 and 42.5 %. On the basis of the obtained results, it was apparent that the soft capsule preparation achieved a higher rate and extent of absorption than shown by Li. Moreover, changing the pharmaceutical dosage forms was an effective method to improve the bioavailability of α-mangostin. At the same time, the achieved pharmacokinetic and tissue distribution results may be useful for further study, providing an impetus to use mangosteen products as nutraceuticals, in functional foods and dietary supplements. Furthermore, the results were also useful for researching the bioactive mechanism of other xanthones.

3.4 Tissue Distribution Study

The tissue distribution of α-mangostin was investigated following a single oral dose of 16.400 mg/kg α-mangostin

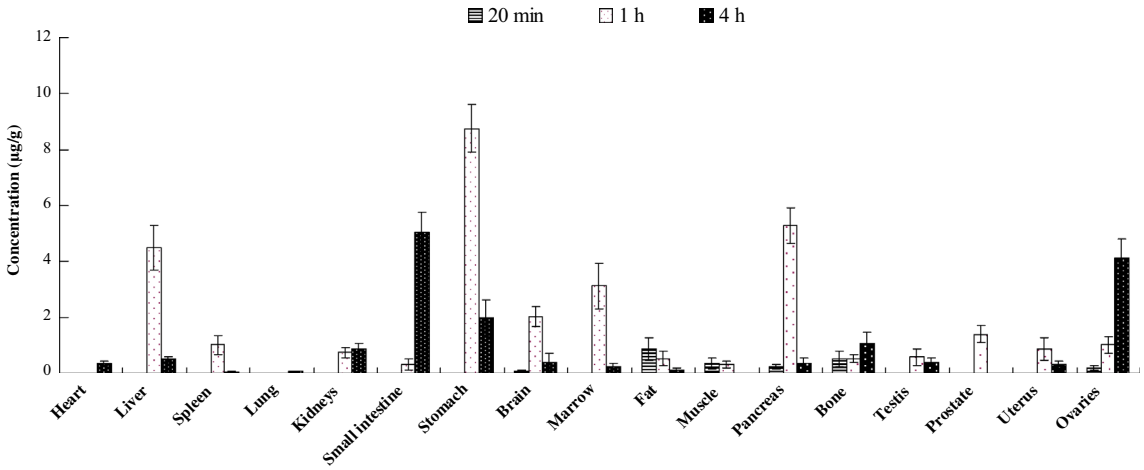

Fig. 2 Tissue distribution of α-mangostin at 20 min, 1.0 and 4.0 h after oral administration of 16.400 mg/kg in rats ($n = 6$)

to rats at 20 min, 1.0 and 4.0 h (Fig. 2). After dosing, only a few kinds of tissue, such as fat, bone, muscle, ovaries and brain, contained a detectable level of α-mangostin after 20 min. After 1 h, except in the heart and lung, α-mangostin was observed in all the other collected tissues, and the highest concentration was observed in the stomach, followed by the pancreas, liver, marrow, brain, prostate, ovaries, uterus and kidneys. Except in the heart, kidneys, small intestine, bone and ovaries, the α-mangostin level gradually decreased after 4 h, and the highest concentration was observed in the small intestine, followed by the ovaries, stomach and bone. In conclusion, α-mangostin could also be detected in the brain homogenate, which suggested that α-mangostin could efficiently cross the blood-brain barrier. The above data indicated that α-mangostin was absorbed quickly and had a wide distribution in tissues throughout the entire body.

4 Conclusions

Research on α-mangostin has been reported in the following areas: the anti-inflammatory response, bioaccessibility, biotransformation and method of metabolization [40–42]. However, in fact, α-mangostin has limited bioavailability. It is one of the most abundant prenylated xanthones present in the mangosteen pericap and has been reported to possess numerous bioactivities, providing the impetus to use mangosteen products as nutraceuticals, functional foods and dietary supplements. Therefore, an effective method to improve the bioavailability of α-mangostin is necessary for its further development and utilization. Here, using α-mangostin in a soft capsule preparation, with vegetable oil as a dispersion matrix, greatly improved the bioavailability. The tissue distribution was studied for the first time, and α-mangostin could be detected in the brain, which indicated that it can cross the blood-brain barrier after oral administration. However, research on cultured cells [40, 42] and rodents [31, 32, 34, 43] and in a human study [41, 44] has demonstrated that α-mangostin (as well as other mangosteen xanthones) is readily conjugated to glucuronide and sulfated metabolites, and some information indicates that the xanthone can be biotransformed to other xanthone compounds. Our team only quantified the administrated compound, i.e., there was no information on extracts from sera and organs also being treated with deconjugating enzymes to determine the total concentrations of α-mangostin and potential biotransformed metabolites. In order to improve the research content, our group will carry out further research to determine the total concentrations of α-mangostin and potential biotransformed metabolites.

Acknowledgments This research was supported by the Natural Science Foundation of China under grant no. 31000154; the National Science and Technology Support Program of China under grant no. 2011BAI03B01; the National Nonprofit Industry Research Special Program of China under grant no. 201303111; the Jilin Province Science and Technology Development Program under grant no. 20126046; 20140204013YY; the Special Program in the Pharmaceutical Industry in Jilin Province under grant no. YYZX201258.

References

1. Ji X, Avula B, Khan IA. Quantitative and qualitative determination of six xanthones in *Garcinia mangostana* L. by LC-PDA and LC-ESI-MS. J Pharm Biomed Anal. 2007;43:1270–6.
2. Jung HA, Su BN, Keller WJ, et al. Antioxidant xanthones from the pericarp of *Garcinia mangostana* (Mangosteen). J Agr Food Chem. 2006;54:2077–82.
3. Gustafson KR, Blunt JW, Munro MHG, et al. The guttiferones, HIV-inhibitory benzophenones from *Symphonia globulifera*, *Garcinia livingstonei*, *Garcinia ovalifolia* and *Clusia rosea*. Tetrahedron. 1992;48:10093–102.
4. Peres V, Nagem TJ. Tryoxigenated naturally occurring xanthones. Phytochemistry. 1997;44:191–214.
5. Peres V, Nagem TJ. Tryoxigenated naturally occurring xanthones. Phytochemistry. 1997;44:191–214.
6. Govindachari TRKP, Muthukumaraswamy N. Xanthones of *Garcinia mangostana* Linn. Tetrahedron. 1971;27:3919–26.
7. Nguyen LH, Venkatraman G, Sim KY, et al. Xanthones and benzophenones from *Garcinia griffithii* and *Garcinia mangostana*. Phytochemistry. 2005;66:1718–23.
8. Suksamrarn S, Suwannapoch N, Ratananukul P, et al. Xanthones from the green fruit hulls of *Garcinia mangostana*. J Nat Prod. 2002;65:761–3.
9. Suksamrarn S, Suwannapoch N, Phakhodee W, et al. Anti-mycobacterial activity of prenylated xanthones from the fruits of *Garcinia mangostana*. Chem Pharm Bull. 2003;51:857–9.
10. Williams RB, Hoch J, Glass TE, Miller JS, et al. A novel cytotoxic guttiferone analogue from *Garcinia macrophylla* from the Surinam rainforest. Planta Med. 2003;69:864–6.
11. Kurose H, Shibata MA, Iinuma M, et al. Alterations in cell cycle and induction of apoptotic cell death in breast cancer cells treated with amangostin extracted from mangosteen pericarp. J Biomed Biotechnol. 2012;2012:672428.
12. Jang HY, Kwon OK, Oh SR, et al. Mangosteen xanthones mitigate ovalbumin-induced airway inflammation in a mouse model of asthma. Food Chem Toxicol. 2012;50:4042–50.
13. José P, Laura MR, Eva GA, et al. ROS scavenging capacity and neuroprotective effect of α-mangostin against 3-nitropropionic acid in cerebellar granule neurons. Exp Toxicol Pathol. 2009;61:491–501.
14. Koh J, Qiu S, Zou H, et al. Rapid bactericidal action of alpha-mangostin against MRSA as an outcome of membrane targeting. Biochim Biophys Acta. 1828;2013:834–44.
15. Krajarng A, Nilwarankoon S, Suksamrarn S, et al. Antiproliferative effect of α-mangostin on canine osteosarcoma cells. Res Vet Sci. 2012;93:788–94.
16. Mizushina Y, Kuriyama I, Nkahara Y, et al. Inhibitory effects of α-mangostin on mammalian DNA polymerase, topoisomerase, and human cancer cell proliferation. Food Chem Toxicol. 2013;59:793–800.
17. Yoo JH, Kang K, Jho EH, et al. Chin, α- and γ- Mangostin inhibit the proliferation of colon cancer cells via β-catenin gene

regulation in Wnt/cGMP signalling. Food Chem. 2011;129:1559–66.
18. Cui J, Hu W, Cai Z, et al. New medicinal properties of mangostins: analgesic activity and pharmacological characterization of active ingredients from the fruit hull of *Garcinia mangostana* L. Pharmacol Biochem Behav. 2010;95:166–72.
19. Yu LM, Zhao MM, Yang B, et al. Phenolics from hull of *Garcinia mangostana* fruit and their antioxidant activities. Food Chem. 2007;104:176–81.
20. Chen LG, Yang LL, Wang CC. Anti-inflammatory activity of mangostins from *Garcinia mangostana*. Food Chem Toxicol. 2008;46:688–93.
21. Nakatani K, Atsumi M, Arakawa T, et al. Inhibitions of histamine release and prostaglandin E2 synthesis by mangosteen, a Thai medicinal plant. Biol Pharm Bull. 2002;25:1137–41.
22. Gopalakrishnan G, Banumathi B, Suresh G. Evaluation of the antifungal activity of natural xanthones from *Garcinia mangostana* and their synthetic derivatives. J Nat Prod. 1997;60:519–24.
23. Kaomongkolgit R, Jamdee K, Chaisomboon N. Antifungal activity of alpha-mangostin against *Candida albicans*. J Oral Sci. 2009;51:401–6.
24. Chen S, Wan M, Loh B. Active constituents against HIV-1 protease from *Garcinia mangostana*. Planta Med. 1996;62:381–2.
25. Ho CK, Huang YL, Chen CC, et al. Garcinone E, a xanthone derivative, has potent cytotoxic effect against hepatocellular carcinoma cell lines. Planta Med. 2002;68:975–9.
26. Devi Sampath P, Vijayaraghavan K. Cardioprotective effect of alpha-mangostin, a xanthone derivative from mangosteen on tissue defense system against isoproterenol-induced myocardial infarction in rats. J Biochem Mol Toxicol. 2007;21:336–9.
27. Weecharangsan W, Opanasopit P, Sukma M, et al. Antioxidative and neuroprotective activities of extracts from the fruit hull of mangosteen (*Garcinia mangostana* Linn.). Med Princ Pract. 2006;15:281–7.
28. Tang YP, Li PG, Kondo M, et al. Effect of a mangosteen dietary supplement on human immune function: a randomized, double blind, placebo-controlled trial. J Med Food. 2009;12:755–63.
29. Sun XH, Niu LJ, Li XQ, et al. Characterization of metabolic profile of mosapride citrate in rat identification of two new metabolites: mosapride N-oxide and morpholine ring-opened mosapride by UPLC-ESI-MS/MS. J Pharmaceut Biomed. 2009;50:27–34.
30. Lv GY, Lou ZH, Chen SH, et al. Pharmacokinetics and tissue distribution of 2,3,5,4′-tetrahydroxystilbene-2-*O*-β-D-glucoside from traditional Chinese medicine *Polygonum multiflorum* following oral administration to rats. J Ethnopharmacol. 2011;137:449–56.
31. Li G, Petiwala SM, Nonn L, et al. Inhibition of CHOP accentuates the apoptotic effect of α-mangostin from the mangosteen fruit (*Garcinia mangostana*) in 22Rv1 prostate cancer cells. Biochem Biophys Res Commun. 2014;453:75–80.
32. Li L, Brunner I, Han A, et al. Pharmacokinetics of a-mangostin in rats after intravenous and oral application. Mol Nutr Food Res. 2011;55:S67–74.
33. Petiwala SM, Li G, Ramaiya A, et al. Pharmacokinetic characterization of mangosteen (*Garcinia mangostana*) fruit extract standardized to α-mangostin in C57BL/6 mice. Nutr. Res. 2014;34:336–45.
34. Ramaiya A, Li G, Petiwala SM, et al. Single dose oral pharmacokinetic profile of α-mangostin in mice. Curr Drug Targets. 2012;13:1698–704.
35. Watanapokasin R, Jarinthanan F, Jerusalmi A, et al. Potential of xanthones from tropical fruit mangosteen as anti-cancer agents: caspase-dependent apoptosis induction in vitro and in mice. Appl Biochem Biotech. 2010;162:1080–94.
36. Zhao Y, Liu Y, Liu J et al. Effects of extract from pericarps of *Garcinia mangostana* on blood lipid in rats with hyperlipidemia. Spec Wild Econ Anim Plant Res. 2007; 2:55–61.
37. Chen CL, Rawwas J, Sorrell A, et al. Bioavailability and pharmacokinetic features of etoposide in childhood acute lymphoblastic leukemia patients. Leuk Lymphoma. 2001;42:317–27.
38. Chen CL, Sangiah S, Bourne DW, et al. Metabolism and metabolite pharmacokinetics of BRB-I-28, a class Ib antiarrhythmic agent. Eur J Drug Metab Pharmacokinet. 1995;20:151–61.
39. Kondo M, Zhang L, Ji H, et al. Bioavailability and antioxidant effects of a xanthone-rich Mangosteen (*Garcinia mangostana*) product in humans. J Agric Food Chem. 2009;57:8788–92.
40. Bumrungpert A, Kalpravidh RW, Suksamrarn S, et al. Bioaccessibility, biotransformation, and transport of α-mangostin from *Garcinia mangostana* (Mangosteen) using simulated digestion and Caco-2 human intestinal cells J Mol Nutr Food Res. 2009; 53(S1):S54–S61.
41. Chitchumroonchokchai C, Thomas-Ahner JM, Li J, et al. Antitumorigenicity of dietary α-mangostin in an HT-29 colon cell xenograft model and the tissue distribution of xanthones and their phase II metabolites[J]. Mol Nutr Food Res. 2013;57:203–11.
42. Gutierrez-Orozco F, Chitchumroonchokchai C, Lesinski GB, et al. α-mangostin: anti-inflammatory activity and metabolism by human cells [J]. J Agr Food Chem. 2013;61:3891–900.
43. Li L, Han A, Kinghorn AD, et al. Pharmacokinetic properties of pure xanthones in comparison to a mangosteen fruit extract in rats. Planta Med. 2013;79:646–53.
44. Chitchumroonchokchai C, Riedl KM, Suksumrarn S, et al. Xanthones in mangosteen juice are absorbed and partially conjugated by healthy adults. J Nutr. 2012;142:675–80.