BILINGUAL PUBLISHING CO.

Journal of Advances in Medicine Science
http://ojs.bilpublishing.com/index.php/jams

ARTICLE

Effects of Exercise on the Pharmacokinetics of Icariin II in Rats

Xiliang Kong*
College of Physical Education, Qufu Normal University, Qufu, Shadong, 273165, China

ARTICLE INFO

Article history:
Received: 25th October 2018
Revised: 15th November 2018
Accepted: 24th December 2018
Published Online: 2nd January 2019

Keywords:
Icariin II
Exercise state
High performance liquid chromatography

ABSTRACT

High performance liquid chromatography (HPLC) was established to determine the concentration of Icariin II in rats. The time-dependent changes in rats after taking Icariin II were studied, and the main pharmacokinetic parameters were obtained. The main pharmacokinetic parameters of taking the medicine in a quiet state, taking the medicine after half an hour of exercise and taking the medicine after long-term exercise were compared to verify whether the absorption and utilization of the Icariin II were more favorable in the exercise state; The paper has important guiding significance for guiding athletes to safe medication and further development of this drug.

1. Introduction

Epimedium is a perennial medicinal plant of Epimedium. It is a nourishing traditional Chinese medicine and is one of the most widely used and longest-established traditional Chinese medicines in China.[1] There are 43 species of Epimedium in China, which is the modern geographical distribution center of this genus. There are five species of Epimedium collected by the Chinese Pharmacopoeia, which are widely cultivated in China.[2] It is mainly distributed in the provinces of Shaanxi, Shanxi, southern Gansu, Henan, Gansu, and Qinghai, Sichuan, Ningxia and other provinces. It grows in shrubs at 650~2100m above sea level or in the backlit wet areas.[3] Icariin (ICA) is a flavonoid contained in it and is also the main active ingredient of Epimedium.[4] Epimedium is a commonly used traditional Chinese medicine for improving exercise capacity. It has the effects of tonifying kidney, strengthening bones and strengthening rheumatism. In recent years, the special effects of using Chinese herbal medicine in competitive sports at home and abroad have been reported and applied in improving athletes' performance, promoting athletes' recovery, and healing and

*Corresponding Author:
Xiliang Kong,
College of Physical Education, Qufu Normal University,
No.57 Jingxuan West Road, Qufu, Shadong, 273165*, China;*
*E-mail: lijuncheng*1971@163*.com.*

Fund Project:
Key Laboratory Construction Project of the"Twelfth Five-Year Plan" of Shandong Province: Construction Project of Key Laboratory of Physical Fitness Monitoring and Regulation in Qufu Normal University.

DOI: https://doi.org/10.30564/jams.v2i1.161

rehabilitation of sports injuries.[5]

In the practice of sports and exercise, Herba Epimedii is a representative aphrodisiac traditional Chinese medicine to achieve good pharmacological effects against athletes' sports fatigue, but there is no pharmacokinetic study of Epimedium aqueous extract. This paper aims to study the pharmacokinetics of Icariin II, the main active ingredient of Epimedium, and find out its mechanism of action, revealing its absorption and metabolism in the body, according to its differences in pharmacokinetic parameters in the exercise state and resting state, to guide the reasonable dosage regimen of athletes under normal and exercise conditions. It also provides theoretical and practical basis for the use of such Chinese medicine in the science of sports and exercise.

2. Experimental Instruments and Materials

2.1 Experimental Instruments

Japan Shimadzu liquid chromatograph, rapid homogenizer (SK-1 type, Jiangsu Jintan Zhengji Instrument Co., Ltd.), high-speed refrigerated centrifuge (GL-20G-II type, made in Shanghai), CNC ultrasonic cleaner (KH-500DB type, Kunshan Hechuang Ultrasonic Instrument Co., Ltd.), vacuum dryer, high speed centrifuge tubes, beakers, pipettes.

2.2 Experimental Reagents

Rhizoma curculiginis aqueous extract solution (homemade, 0.9g/ml), orcinol glucoside standard substance (purity: 99%, purchased from Shanghai Abbott Technology Co., Ltd.), mobile phase methanol (chromatographically pure, made in Tianjin), experimental water (double distilled water) and filter membranes.

2.3 Experimental Animals

Male Wistar rats, weighed 220-240g (Jining Lukang Group Pharmaceutical Co., Ltd.).

3. Experimental Methods

3.1 Determination of the Concentration of Icariin II in Rat Plasma by HPLC

3.1.1 Chromatographic Condition

The mobile phase was acetonitrile: 0.1% formic acid (70:30, v/v), which was filtered through a 0.22 μm microporous filter. On-line degassing; flow rate lml/min; column temperature 30 °C; injection volume 20 μl, detection wavelength is 270 nm, external standard method for the concentration of Icariin II.

3.1.2 Plasma Sample Treatment

Accurately draw 0.5 ml of plasma, add 1 ml of methanol, vortex and mix, centrifuge at 4800 rpm for 15 min to take the supernatant, and filter the supernatant through a 0.45 μm micropore filter.

3.1.3 Preparation of Standard Curve

The Icariin II reference methanol solution was diluted with blank rat plasma to obtain standard samples with mass concentrations of 100, 200, 400, 600, 800 and 1000 $ng•mL^{-1}$, respectively. The concentration of Icariin II in plasma ρ is plotted on the abscissa, and the ratio of reference to internal standard peak height Y is plotted on the ordinate. The regression operation is performed by weighted (l/c2) least square method to calculate the regression equation.

3.2 Plasma Protein Binding Rate

Four rats were administered quietly, taking 4 ml of rat plasma sample for 2 hours, placed in a heparin-coated centrifuge tube, 2 ml was centrifuged at 4800 r/min for 15 min at normal speed, and then 0.5 ml of plasma was taken and stored in a refrigerator at -24 °C to be tested. Then, 2 ml of plasma was centrifuged (16000 r/min) in an ultracentrifuge tube. After 10 minutes, 0.5 ml of plasma was taken and a 0.45 μm micropore filter was used for high-performance liquid phase analysis. The degree of binding of the drug to the plasma protein was calculated using the following equation.

Binding Rate% = 100-(100* Filtrate Drug Concentration/Plasma Drug Concentration)

3.3 Administration Method

Sixty male Wistar rats were randomly divided into a quiet medication group (40), a half-hour exercise group (10), and a long-term exercise group (10). The dosage was 0.9g/kg, and the water was fasted for 12 hours. The eyeballs were taken at 5, 15, 30, 60, 120, 180, 240, 360, 480, and 600 min, and centrifuged at 4000 rpm for 10 min. The concentration was measured after the treatment.

3.4 Pharmacokinetic Parameter Calculation

The DAS2.0 software package of the Chinese Pharmacological Society was used to fit the chamber model and calculate the pharmacokinetic parameters (Statistical Moment Method).

4. Experimental Results

4.1 Concentrations of Rats in different groups

The area under the curve of the quiet medication group, the exercise half-hour medication group and the long-term exercise medication group were substituted into the standard curve equation Y=0.0159X-49.769 (R2=0.9972), and the average blood concentration of the three groups was obtained (Table 1).

 DOI: https://doi.org/10.30564/jams.v2i1.161

Table 1. Mean Plasma Concentration of Icariin II under Different Exercise States

	Quiet Medication Group		Half-hour Exercise Group		Long-term Exercise Group	
T(min)	Mean area $\bar{x}$	Mean concentration	Mean area $\bar{x}$	Mean concentration	Mean area	$\bar{x}$ Mean concentration
	（ug/L*min）	$\bar{x}$(ng/ml)	（ug/L*min）	$\bar{x}$(ng/ml)	（ug/L*min）	$\bar{x}$(ng/ml)
5	5778.03	316.25	9547.9	102.04	29114.4	413.15
15	7676.23	437.31	13354.5	162.57	34592.4	500.25
30	9380.30	440.63	14257.5	176.93	36129.5	524.69
60	8460.98	487.45	17056.3	221.43	5140.6	767.58
120	15968.7	966.68	34010.9	491.00	56173.5	843.39
180	13242.6	792.76	31151.7	445.54	70185.5	1066.18
240	6063.125	334.55	27551.7	388.30	41720.1	613.58
360	4600.025	240.91	19338.5	257.71	35893.7	520.94
480	4050.675	205.44	13122.6	158.88	14113.8	174.64
600	3798.65	189.32	4988.7	29.55	13579.8	166.15

4.2 Main Pharmacokinetic Parameters of Different Groups of Rats

The one-compartment model was selected as the best compartment model, and the pharmacokinetic parameters of rats in different groups were obtained by the pharmacokinetic software DAS2.0 (Table 2).

5. Analysis and Discussion

5.1 Analysis of Pharmacokinetic Parameters of Icariin II in Different States

Pharmacokinetic parameters are some constants that reflect the dynamic changes of drugs in the body, such as absorption, transport and elimination of transport constants, elimination of half-life, etc., is one of the main bases for the development of rationalized drug delivery programs. Depending on the nature of the pharmacokinetic parameters, a safe and effective dosing regimen can be designed and developed, including the dosage administered, the time of administration, and the optimal route of administration. In this paper, the pharmacokinetic characteristics and dynamic changes of Icariin II were revealed by the analysis of the pharmacokinetic parameters of Icariin II, to clarify the regularity of the effect of Icariin II, to understand its main action site in vivo and the material basis of toxicity, and the pharmacokinetic parameters of Icariin II are important data evaluation indicators for its preparation quality. The half-life of the three groups of pharmacokinetic parameters was t1/2=69.315, indicating that there was no difference in the elimination rate of the three groups of experiments; half-hour exercise medication and long-term exercise medication did not change the elimination rate of Icariin II, and the elimination rate constant ke was 0.01, confirming that the exercise did not change their

Table 2. Pharmacokinetic parameters of Icariin II under different exercise conditions

	Quiet Medication Group	Half-hour Exercise Group	Long-term Exercise Group
t1/2 min	69.32	69.32	69.32
Ke 1/min	0.01	0.01	0.01
V1/F L/kg	2.49	6.96	2.76
CL/F L/min/kg	0.06	0.07	0.028
AUC(0-t) ug/L*min	207965.16	118424.28	296970.37
Ka 1/min	0.03	0.01	0.01
t1/2Ka min	27.88	69.32	69.32
Tmax min	120	120	180
Cmax ng/L	966.00	491.00	10660.18

DOI: https://doi.org/10.30564/jams.v2i1.161

elimination rate of the drug; the absorption rate constant was quiet in the drug group, ka=0.025, half-hour exercise group, ka=0.01, and long-term exercise group, ka=0.01, indicating that the quiet group took the drug faster than the exercise group; the AUC(0-t) quiet medication group value was 207,965 ug/L*min, the half-hour exercise medication group AUC(0-t)=118424, and the long-term exercise medication group AUC(0-t)= 296970.

From the three sets of data, it can be seen that the area under the curve of their medicine time is:

Long-term exercise group > quiet medication group > half-hour exercise group. This indicates that the long-term exercise group is larger than the quiet medication group; the quiet medication group is larger than the exercise half-hour medication group, and the peak time (Tmax) peak concentration (Cmax):

The peak time of the quiet medication group and the half-hour exercise medication group was 120 min, and the peak time of the long-term exercise medication group was 180 min. Although the long-term exercise peak time was longer, the peak concentration of the long-term exercise medication group was 1066 ug/L, which is larger than 966 ng/L in the quiet medication group and 491 ug /L in the half-hour exercise group. The longer the peak time, the longer the drug stays in the body, which is more conducive to the efficacy of the drug.

5.2 Factors Influencing the Pharmacokinetic Parameters of Icariin II

After exercise, a large amount of lactic acid is produced in the body, which causes the pH of the internal environment to change, thereby affecting the polarity of the drug and affecting the absorption degree of Icariin II; the blood flow of exercise blood is distributed to muscles and other sports organs, and the decrease of visceral blood flow during exercise leads to the decrease of absorption of most drugs after oral administration, which affects the absorption of Icariin II; exercise may affect the binding rate of the drug to the plasma protein, thereby affecting the concentration of the free drug, and ultimately affecting the absorption of the drug; after exercise, the temperature in the body increases, the activity of the enzyme in the body also changes, the decomposition of the drug is accelerated, or the temperature affects the plasma protein binding rate of the drug. All of the above may be factors affecting the degree of absorption of Icariin II. Specifically, a certain factor or the influence of the above-mentioned comprehensive factors still needs to be verified by subsequent research.

6. Conclusion

(1) There were differences in pharmacokinetic parameters between the quiet medication group, the half-hour exercise group, and the Long-term exercise group.

(2) In the long-term exercise group, the area under the curve (AUC) was larger than that in the quiet medication group, and the area under the curve (AUC) in the quiet medication group was larger than the half-hour exercise group. It indicated that the long-term exercise group absorbed more drugs than the quiet group, and the quiet medication group absorbed more drugs than the half-hour exercise group.

(3) In the long-term exercise group, the plasma concentration peak time (Tmax) was larger than that in the quiet medication group, and the peak time (Tmax) of quiet medication group was the same as the half-hour exercise group.

(4) The peak concentration (Cmax) of the long-term exercise group was larger than that of the quiet medication group, and the peak concentration (Cmax) of the quiet medication group was larger than the half-hour exercise group.

References

[1] Zuozhou Li, Yanqin Xu, Ying Wang, etc. Research status and prospects of Epimedium medicinal plants[J].Chinese Herbal Medicine, 2005, 36 (2). (in Chinese)

[2] Xin Ji. Research progress on immunomodulatory effects of Icariin[J].Hebei Medical Journal, 2016 (5): 753-756. (in Chinese)

[3] GUO B L XIAO PG. Comment on main species of Herb Epimedium[J]. China Journal of Chinese Matera Medico, 2003, 28(4). (in Chinese)

[4] Jinxiao Shi, Cheng Dian. Study on leaf anatomical structure of Epimedium sagittatum in different populations[J]. Life Science Research, 2005, 9 (4). (in Chinese)

[5] Hongming Nie, Rong Wang, et al. Experimental study on liver-protecting and choleretic treatment of Epimedium decoction and Icariin II[J].Journal of Practical Chinese Medicine, 2006, 20(4). (in Chinese)

[6] Erxin Shang, Xinsheng Fan, Yuping Tang, et al. Study on the variation of prescription and dosage of"Golden Chamber"[J].Journal of Nanjing University of Traditional Chinese Medicine,2009,25(1). (in Chinese)

[7] Yifan Zhou, Changjiang Hu, et al. Pharmacokinetics of Epimedium in rats before and after processing[J].Chinese Patent Medicine, 2013, 12. (in Chinese)

[8] Xiwei Zhang, Yingping Chen,et al. LC-MS/MS method for the detection of plasma Icariin in healthy humans and its pharmacokinetic study[J].Liaoning Journal of Traditional Chinese Medicine, 2014, 1. (in Chinese)

[9] P Xue, X Wu, L Zhou, H Ma. IGF1 promotes osteogenic differentiation of mesenchymal stem cells derived from rat bone marrow by increasing TAZ expression[J]. Biochemical & Biophysical Research Communications, 2013, 433 (2): 226-231. (in Chinese)

 DOI: https://doi.org/10.30564/jams.v2i1.161