Determination of Bergenin in Yuchuangling Capsules by High Performance Capillary Electrophoresis

Yunchen Zhang, Xiaoping Zhang, Xue Wang, Haixing Liu[a,*]

Chemistry & Chemical and Environmental Engineering College, Weifang University, Weifang 261061, P.R. China

[a,*]haixingliu@tom.com

Abstract. This paper presents the determination of bergenin content of Yuchuangling capsules by high performance capillary electrophoresis (HPCE) method. Elastic quartz capillary electrophoresis separation control at the temperature of 20°C without coating capillary (75μm× 44/52 cm), the sample height was 7.5cm, we choose the borax solution as buffer solution, and its concentration was 20 mmol/L at a constant voltage of 20kV and injecting time of 10s. Linearity was kept in the concent ration range of 0.085~1.35mg /mL of bergenin with correlation coefficient of 0.998. Measured bergenin content in Yuchuangling capsules was 15.93 mg /g (RSD = 4.78%) (n = 5). The recovery was in the range of 90.7% - 115.1% (n=5). This method is specific, simple and rapid and accurate, which is suitable for the detection of the content of bergenin in Yuchuangling capsules.

Keywords: capillary electrophoresis, bergenin, Yuchuangling capsules.

1. Introduction

Yuchuangling capsules is consists of sanqi, safflower, cucumber seeds (fried), ground beeltle, chinese angelica, the natural copper (calcined), borneol, himalayan teasel root, chinese astilbe, etc. It has the effect of promoting blood circulation and scattered stasis and diminishing swelling pain relief. It is used for the treatment of knocks falls contusion, bones and muscles hemostasis swelling pain disease. It is also used for the adjuvant therapy of fracture [1]. Qu et al [2] established an HPLC-ELSD method to determine the content of saponins in Yushangling capsules. The contents of Notoginsenoside R1, ginsenoside Rg1, ginsenoside Re, ginsenoside Rb1 and ginsenoside Rd in Yushangling capsules were determined by HPLC-ELSD. The separation was carried out on C18 column with acetonitrile-water as mobile phase by gradient elution: 0-20 min, 25% acetonitrile; 20-30 min, 25%-30% acetonitrile; 30-45 min, 30%-50% acetonitrile; 45-48 min, 50%-90% acetonitrile. The flow rate was 1 mL·min^{-1}. The column temperature was 25°C . Zhang et al [3] established a method for determination of aflatoxin G2, G1, B2, B1 in Yushangling capsules by HPLC. The samples were extracted with 70% methanol and purified with immunoaffinity column, then analyzed by HPLC with fluorescence detection. The positive samples were identified by ultra performance liquid chromatography - tandem mass spectrometry. Chen et al [4] developed an HPLC method for determination of hydroxysafflor yellow A in Yushangling capsules. The Zorbax Eclipse XDB-C18 (150 mm×4.6 mm, 5 μm) column was used. The mobile phase composed of methanol-0.5% phosphoric acid (25:75) with flow rate of 1.0 mL·min^{-1}.The detection wavelength was 403 nm. The column temperature was 35°C. Huang et al [5] established a method of the determining bergenin in Yushangling Capsula by HPLC. A HPLC was used for the determining the contents of bergenin in yushanglin gcapsula with bergenin as the marker. The chromatographic column was Kromasil C18 column (250 mm×4.6 mm, 5 μm), with a mobile phase of methanol-water(25:75) and the flow rate of 1.0 ml·min^{-1}. The content of ginsenoside Rg1 in Yushangling Capsules was measured by Zeng et al [6]. A RP-HPLC method was developed using C18 column, with acetonitrile-0.05 % phosphoric acid as a mobile phase and the flow rate of 1.0mL ·min^{-1}. The detection wavelength was 203nm. Shi et al [7] established an HPLC - ELSD method for determination of ginsenoside Rg1 and Rb1 in Yushangling capsule. The separation was studied in a Hypersil ODS2 column with a mobile phase of acetonitrile -water in gradient elution, ELSD nebulization at 110°C and flow rate of nitrogen at 3.02–3.05 SLPM. In this paper, the bergenin content in Yuchuangling capsules was determined by High Performance Capillary Electrophoresis.

2. Experimental Section

2.1 Instruments and Reagents

Experimental instruments: CL-1030-type high performance capillary electrophoresis (Beijing Cailu Scientific Instrument Co., Ltd.); HW2000-type chromatography workstation (Nanjing Qianpu Software Ltd.); Capillary (75 μm inner diameter, 52 cm overall length, 44 cm effective length) from Hebei Yongnian Ruifeng Chromatographic Devices Co., Ltd.).

Bergenin (Chinese Drugs and Biological Products); Yuchuangling capsules (Shanxi baiyun pharmaceutical Co., Ltd.); Other reagents used in the experiments were all analytical grade; Double-distilled water was used.

2.2 Experimental Methods

Before the start of the experiment, capillary was successively washed with 1 $mol \cdot L^{-1}$ hydrochloric acid solution, double-distilled water, 1 $mol \cdot L^{-1}$ sodium hydroxide solution, double-distilled water, buffer solution, each for 8 min. After three times running, capillary was cleaned again using the above method.

Measurements were carded out at 20 kV voltage and experimental temperature at 20°C. UV detection wavelength was 275 nm. Injection time was 10s (7.5 cm height difference).

2.3 Sample Preparation

Yuchuangling capsules sample solution: Yuchuangling capsules powder was accurately weighed 1.44 g, added 40 mL water with 30% methanol, extracted time of 3h at 60°C, filtered, washed and set the volume to 50 mL that was the Yuchuangling capsules sample solution.

Bergenin standard solution: Bergenin was accurately weighed 5.4 mg, added 4 mL water with 30% methanol.

3. Results and Discussion

3.1 Selection Electrophoresis Conditions

Based on past experiment experience, we chose 20 mmol/L borax solution as a running buffer solution.

According to the literature, bergenin maximum absorption wavelength was at 275 nm, so we chose the 275 nm detection wavelength.

3.2 Quantitative Analysis

3.2.1 Standard Curve

First, bergenin standard solution that the concentration were 1.350, 0.675, 0.338, 0.169, 0.085 mg/mL was prepared. Each standard solution was run for three times under the above electrophoresis conditions and the results averaged. The chromatogram of bergenin standard solution was showed in Figure 1. Taking concentration as the abscissa and peak area as the ordinate, the standard curve was drew. Linear regression equation of bergenin (peak area: y μV•s, density: x mg/mL) and the linear range was as follows: Y=-6404.58+129218.88X (r=0.998), 0.085~1.350 mg/mL.

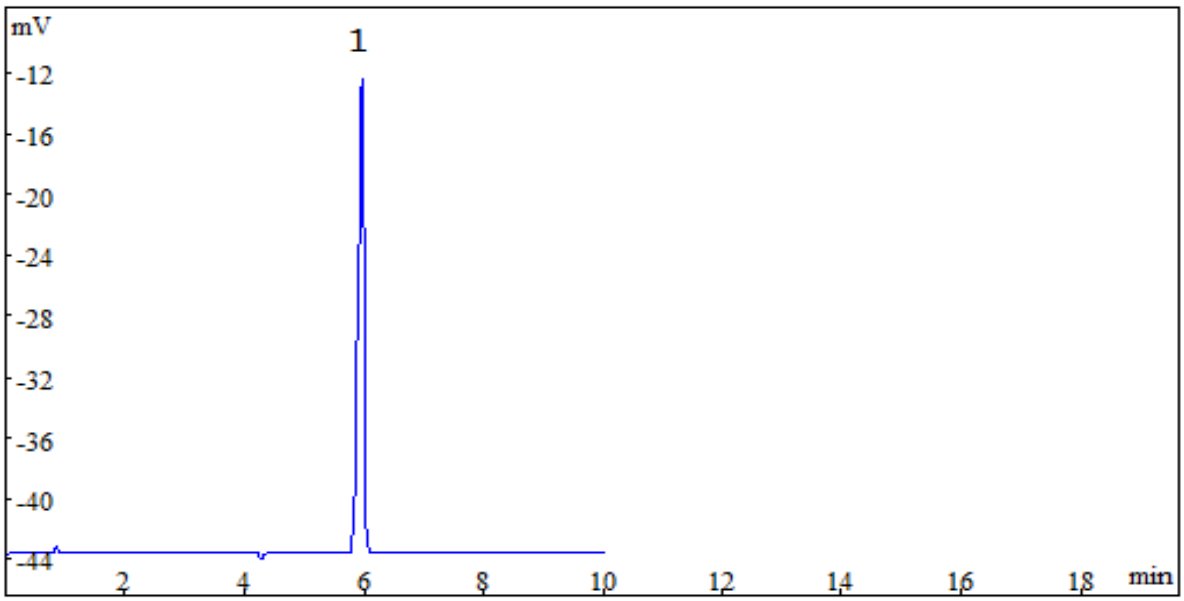

Fig 1. Electrophorogram of bergenin standard solution 1- bergenin

3.2.2 Precision Test

A bergenin standard solution precisely drew and continuously injected for sixt times under electrophoretic separation conditions, the RSD of bergenin peak area were 3.92%, indicating good precision.

3.2.3 Determination of Sample Content

Under selected electrophoresis conditions, Yuchuangling capsule sample solution was run. Separation chromatogram of the Yuchuangling capsules sample solution was showed in Figure 2. Measured bergenin content in Yuchuangling capsules was 15.93 mg/g (RSD = 4.78%) (n = 5).

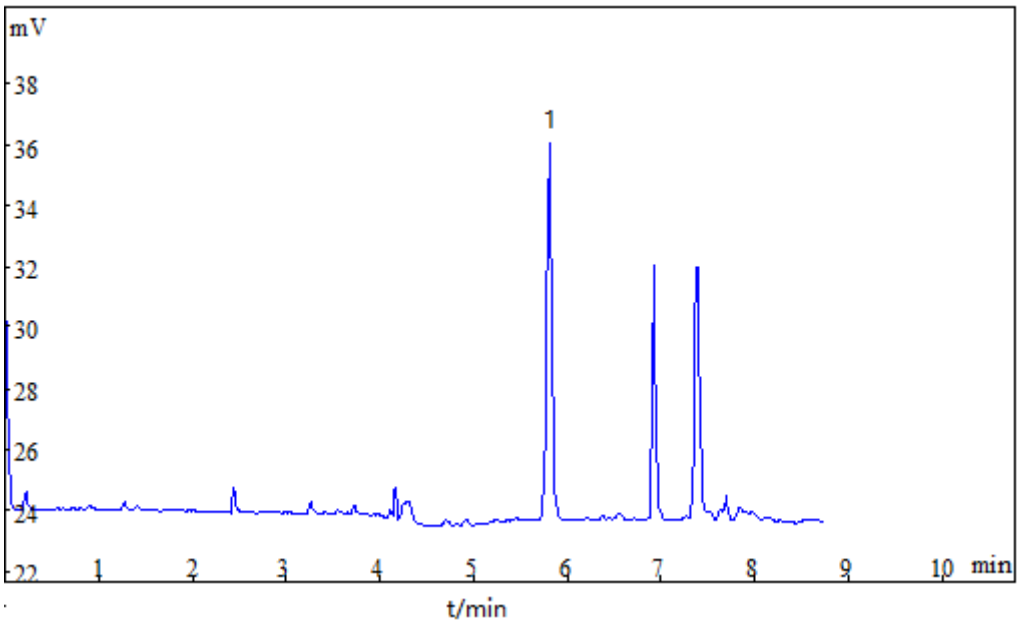

Fig 2.Electrophorogram of Yuchuangling capsules sample solution 1- bergenin

3.2.4 Recovery

After determination for six times, the recovery of bergenin in Yuchuangling capsules sample was in the range of 90.7% - 115.1% (n=5).

Acknowledgments

This study were supported by the Natural Science Foundation of Shandong Province (No. ZR2010BL025), Open Project of State Key Laboratory of Supramolecular Structure and Materials (No. sklssm201323)(Jilin University), State Key Laboratory of Inorganic Synthesis and Preparative Chemistry (No. 2011-13)(Jilin University).

References

[1]. Jinhong Wang, Yunong Hong, China Pharmaceuticals, 2009, 18(22), 39-40.

[2]. Lihua Qu, Ke Tan, Central South Pharmacy, 2014, 12(5), 481-484.

[3]. Zhengfeng Zhang, Yi Zhang, China Pharmaceuticals, 2015, 24(15), 37-39.

[4]. Zongliang Chen, Ke Zhu, Lingna Zhou, Jiufeng Dou, Feng Wu, Chin J Pharm Anal, 2010, 30(2), 297=299.

[5]. Junrong Huang, Anhui Medical and Pharmaceutical Journal, 2009, 13(1), 37-38.

[6]. Sanping Zeng and Ye Ding, Chinese Pharmaceutical Affairs, 2008, 22(8), 697-698.

[7]. Yue Shi, Lan Sun, Yang Cao, Xingye Cheng, Shunliang Zheng, Lishizhen Medicine and Materia Medica Research, 2007, 18(2), 368-369.