Journal of Ayurvedic and Herbal Medicine

Research Article

ISSN: 2454-5023
J. Ayu. Herb. Med.
2022; 8(2): 85-87
Received: 30-04-2022
Accepted: 07-06-2022

www.ayurvedjournal.com
DOI: 10.31254/jahm.2022.8205

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The identification of (−)-epigallocatechin-3 gallate (EGCG) and (−)-epicatechin (EC) content in Trungdutim tea (*Camellia sinensis* var. macrophilla) at Vietnam

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ABSTRACT

The identification of (−)-epigallocatechin-3 gallate (EGCG) and (−)-epicatechin (EC) content in Trungdutim tea (*Camellia sinensis* var. macrophilla) was analysed by high performance liquid chromatography. The results are 11.50±0.20 % and 15.51±0.40 % dry weight, respectively.

Keywords: EGCG, EC, Trungdutim Tea, *Camellia sinensis* var. macrophilla.

INTRODUCTION

Tea (*Camellia sinensis*) was detected very early, around 2700 BC [1-3]. Trungdu varieties including Trungduxanh and Trungdutim have been considered the origin of Vietnam Tea. Trungdutim tea belonging Theaceae family has science name of *Camellia sinensis* var. macrophilla [2,3]. It is claimed to have originated in China [4]. but it is now grown in many countries having tropical and temperate climates around the world. The composition of the green tea product is very similar to that of fresh tea leaves except for some changes due to enzymatic hydrolysis which occurs extremely rapidly after the tea is removed from the plant. Because during the green tea production process, efforts have been made to limit the oxidation of polyphenols in tea leaves [5]. Compounds (-)-epigallocatechin gallate (EGCG), (-)-epigallocatechin (EGC), (-)-epicatechin gallate (ECG), (-)-epicatechin (EC) and (+)-catechin are the main catechins in fresh tea leaves as well as in green tea products [2]. These catechin compounds, which can account for up to 30% of the dry matter of brewed tea, are colorless, water-soluble chemical compounds that give brewed tea a bitter and acrid taste [2,3].

The main biological effect of catechins in green tea leaf extract is antioxidant. Free radicals are generated and accumulated in the process of life, which is the main cause of disease and accelerates the aging process of the human body. Today, it has been found that the catechins in green tea leaves have different effects on cancer, cardiovascular disease, high blood pressure, intestinal disease, dental disease, and have the effect of slowing down the aging process, increasing life expectancy.

In Vietnam, there are many tea growing areas, such as Thai Nguyen, Tuyen Quang, Yen Bai and Phu Tho. However, the content of catechins is still little attention. While these components vary greatly according to cultivar as well as climatic and soil conditions.This study will contribute to elucidating the content of compounds (-)-epigallocatechin gallate (EGCG) and (-)- epicatechin (EC) from Trungdutim tea in Thai Nguyen.

MATERIALS AND METHODS

Chemicals and reagents

Methanol (95%) was purchased from Echo Chemical Co., LTD. (Taiwan). Dichloromethan was purchased from J.T. Baker (Avantor Performance Inc, USA). EGCG and EGC were procured from Sigma-Aldrich (St. Louis, MO, USA), and other chemicals and reagents.

Preparation of samples

The *Trungdutim* tea bud (*Camellia sinensis* var. macrophilla) was harvested at Tan Cuong commune,

Thai Nguyen city. After that samples were washed by distilled water and dried in oven at 45℃ for 48 h. 50 grams of 50-mesh sieved dry powder were separately extracted three times with MeOH:H_2O (80:20,v/v) at 40-50°C by ultrasonic for 30 min and then was filtered. The filtrates were combined and concentrated in a vacuum evaporator at 45℃. The dehydrated fractionation was processed in dichloromethan to remove impurities. Aqueous solution is supplemented with etyl axetat. The total solid tanin was concentrated in a vacuum evaporator and the activities were measured.

High performance liquid chromatography (HPLC) analysis

The analysis for phenolic compounds was determined quantitatively by HPLC. The HPLC system (Hitachi, Tokyo, Japan) consisted of a chromaster 5110 pump, a chromaster 5210 autosampler, a chromaster 5430 diode array detector and a Mightysil RP-18 GP column (4.6 x 250 mm) (Kanto Chemical Co. Inc., Tokyo, Japan). Samples were centrifuged at 3000 x g for 10 min and filtered by 22 µm membrane prior to HPLC injection. The mobile phase for HPLC consisted of solvent (A)-distilled water adjusted at pH 2.8; solvent (B)-CH_3CN was adjusted respectively at pH 2.8 by phosphoric acid. The following gradients were used: 0-10 min, 15% B; 10-16 min, 15 to 70% B; 16-20 min, 70% B; 20-25 min, 70 to 85% B. Operating conditions were as follows: flow rate, 0,6 mL/min; injection volume, 5 µL and UV detector at 254 nm. The column was operated at temperature of 25℃. The results were obtained by interpolation using the linear regression plot from the standard component solution.

RESULTS AND DISCUSSION

Calibration curve construction

From the stock solution of EGCG and EC with a concentration of 1 mg/ml, dilute to create solutions with solvents. The stock solution of EGCG and EC with a concentration of 1 mg/ml was diluted to obtain concentrations of 0.02; 0.04; 0.06; 0.1 mg/ml. Calibration curve was constructed by running the sequence with the automatic pump and the running programm with the help of chemstation software.

The UV chromatograms of all EGCG samples showed stable signals at a retention time of 12.2 min, of EC 17.767 min. From the data on peak area and concentration, the regression equation and correlation coefficients of the calibration lines were as follows: y = 51977.75320x - 86.68356, with high reliability (r = 0.99390).

EGCG and EC identification by high performance liquid chromatography

Five grams of dry extract were dissolved in 10 ml MeOH to obtain concentration of 0.5 mg/ml and were filtered through a 0.45 µm Millipore filter prior to HPLC analysis. Through the results of comparison of retention times, combining EGCG, EC calibration curve and integrating peaks at retention time 12.2; 17.764 minutes on the character colors of the parsing sample. The quantitative EGCG results were 11.50 ± 0.20% and the EC was 15.51 ± 0.40% dry weight. The result is in agreement with previous reported works that the buds and young leaves of *Trungdutim* tea contain EGCG (Hoang *et al*., 2019). In the present work, one catechin of EC in *Trungdutim* tea cultivar was newly quantitatively detected by high-performance liquid chromatography. HPLC chromatograms of EGCG and EC components were showed in figure 1 and figure 2.

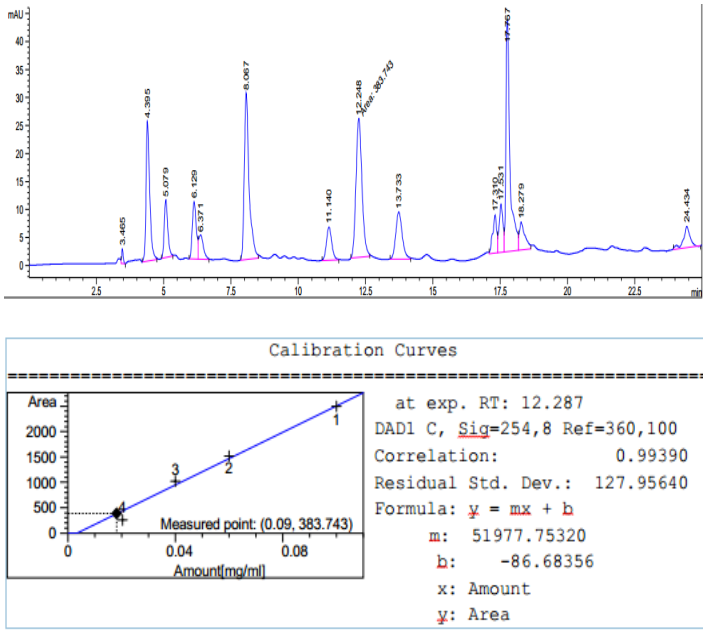

Figure 1: HPLC chromatograms of EGCG component

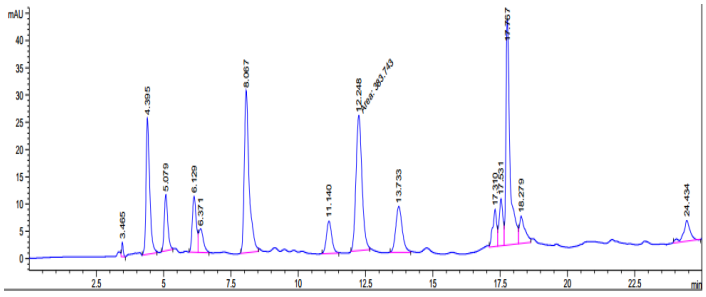

Figure 2: HPLC chromatograms of EC component

CONCLUSION

The present work is the first report about the content of (-)-epicatechin (EC) in the *Trungdutim* tea cultivar (*Camellia sinensis* var. macrophilla) at Thai Nguyen in Vietnam. Therefore, the findings obtained in the present work could enhance knowledge the catechin components of this tea cultivar.

Acknowledgments

This research was funded by Thai Nguyen University of Agriculture and Forestry, Thai Nguyen, Vietnam.

Conflict of Interest

None declared.

REFERENCE

1. Hoang TT, Luu HL, Nguyen TL, Duong TD, Nguyen HD, Huynh TTH, *et al*. A comparison of the production of polyphenol contents and the expression of genes involved in Vietnamese tea cultivars. International Food Research Journal. 2019;26(6):1781-88.
2. Owuor PO, Reeves SG. Optimising fermentation time in black tea manufacture. Food Chem. 1986;21:195–03.
3. Weisburger JH. Tea and health: a historical perspective. Cancer Lett. 1997; 114:315–17.
4. Wang J, Rossow WB, Zhang YC. Cloud vertical structure and its variations from a 20-year global rawinsonde dataset. J. Climate. 2000;13:3041-56.
5. Vinson JA, Hao Y, Su X, Zubic L. Phenol antioxidant quantity and quality in foods: vegetables. J. Agric. Food Chem. 1998;46:3630-34.

HOW TO CITE THIS ARTICLE

Lanh TN, Quang-Ung Le, Pham TT. The identification of (−)-epigallocatechin-3 gallate (EGCG) and (−)-epicatechin (EC) content in Trungdutim tea (*Camellia sinensis* var. macrophilla) at Vietnam . J Ayu Herb Med 2022;8(2):85-87. DOI: 10.31254/jahm.2022.8205