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Centella asiatica (L.) Urb. EXTRACTS: EFFECT OF EXTRACTION CONDITIONS ON BIOACTIVITY AND PHYTOCHEMICAL COMPOSITION

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ABSTRACT

This study aimed to preliminarily analyze the phytochemical composition and evaluate factors affecting the bioactivity of *Centella asiatica* (L.) Urb. extracts. *C. asiatica* sample was collected in a local garden in Tien Giang province under traditional local cultivation conditions; the VietGAP *C. asiatica* sample was also collected for comparison. The results showed a strong effect of solvents on the phytochemical content and bioactivity of the *C. asiatica* extract when using the ultrasonic-assisted extraction technique. The ethanol/water (3:2 v/v) solvent provided the highest antioxidant extract, while the ethanol solvent supported the extraction of antibacterial compounds. The appropriate conditions for antioxidant activity-guided extraction were determined including ethanol/water (3:2 v/v) solvent, extraction time of 45 min, and temperature of 40 ^{o}C. Under these conditions, the total saponin content, total phenolic content, and total flavonoid content of the extract were 24.15 ± 0.04 mg oleanolic/g, 22.17 ± 0.63 mg GAE/g, and 21.25 ± 0.46 mg RE/g, respectively. The *C. asiatica* sample under VietGAP had a higher amount of total saponins than the *C. asiatica* sample collected in the local garden, while the latter contained higher amounts of phenolics and flavonoids.

Keywords: Centella asiatica (L.) Urb., antioxidant activity, antibacterial activity, phytochemical content.

1. INTRODUCTION

Centella asiatica (L.) Urb. is a popular herbal plant [1] of the family Apiaceae distributed in tropical and subtropical regions. *C. asiatica* has been known for remarkable biological activities and holds great potential for applications in personal-care products, medicines, and other health-related products. Vietnamese communities widely plant and use this herb as a fresh vegetable. In some local areas, it has been applied to improve memory and treat mental fatigue, anxiety, eczema, and kin-related diseases [2]. Clinical studies showed that *C. asiatica* extract could be used to stimulate skin regeneration at burns, preventing scar tissue formation by inhibiting collagen production at the wound [3]. The *C. asiatica* extract contained isothankuniside and thankuniside glycosides, which had anticoagulant effects on white mice [3]. Effects against diseases, such as psoriasis, convulsions, cancer, diabetes, sore throat, asthma, colitis, and mouth sores were also reported [4]. The therapeutic effect of *C. asiatica* is mainly attributed to secondary metabolites associated with antioxidant, antibacterial, and anti-inflammatory activities such as tannins, flavonoids, and alkaloids [1-4]. Triterpenes are the major active components found in *C. asiatica*. For example, asiatic acid, madecassic acid, asiaticoside,

and madecassoside have been supposed to be responsible for anti-ulceration and anti-inflammatory properties [1]. On the other hand, anti-inflammatory, anti-lipid peroxidative, and free radical scavenging activities are contributed by flavonoids and phenolic derivatives, such as quercetin, kaempferol, rutin, and chlorogenic acid [1].

The bioactivity of *C. asiatica* extract should vary depending on the plant origin, geographical location, and extraction procedure. Except for genetic and environmental factors, building an appropriate extraction procedure with the right choice of extraction method and extraction conditions (solvent type, concentration, time, etc.) is crucially important in deciding the activities of received extracts. Many attempts have been made to achieve the desired quality of *C. asiatica* extract. Various extraction techniques have been investigated including maceration, distillation, Soxhlet, ultrasound, and microwave application. Duval et al. [5] performed the extraction with 30 wt.% of an alcoholic solvent to obtain a mixture of madecassoside and terminoloside from C. asiatica. In another work, Loiseau et al. [6] established an extraction process to obtain the extract with more than 75 wt.% of the madecassoside, asiaticoside, and terminoloside mixture. To receive the required activities and prevent the loss of the target compounds, the effect of extraction conditions is always a topic of interest, particularly for C. asiatica plants cultivated in different conditions and geographical areas.

This study focused on analyzing phytochemical composition and evaluating the bioactivity of *C. asiatica* extracts from Tien Giang province. The extraction was conducted under ultrasonic. The effect of extraction conditions on antioxidant activity and phytochemical content was investigated. The antioxidant activity was determined through DPPH radical scavenging assay and the antibacterial activity was tested by using the agar well diffusion method. The phytochemical content and bioactivity of the VietGAP *C. asiatica* sample were also examined for comparison.

2. MATERIALS AND METHODS

2.1. Chemicals and materials

C. asiatica samples were collected in the *C. asiatica* garden in the June-July period in Tan Hiep ward, Chau Thanh district, Tien Giang province, Vietnam. The voucher specimens (Figure 1) were taxonomically identified at the Southern Institute of Ecology, Vietnam. The VietGAP *C. asiatica* sample was collected in the same region for comparison study. Ethanol (>99.5%), methanol (>99.5%), sodium hydroxide (>96.0%), sodium nitrite (>99.0%), phosphoric acid (≥85.0%), acetic acid (>99.5%), sodium carbonate anhydrous, aluminum chloride (>97.0%) and Folinol-Ciocalteu were purchased from Xilong Ltd. (China). Gallic acid, vitamin C, oleanolic acid, and rutin were supplied by Merck (Germany). 1,1-Diphenyl-2-picrylhydrazyl (>97.0%) was purchased from TCI (Japan).

Figure 1. C. asiatica specimens collected in the garden for identification

2.2. Extraction of *C. asiatica* (L.) Urb.

C. asiatica was washed with water to remove impurities and dried at 50 ^{o}C for 48 h. After drying, the plant sample was tested for moisture, ground, and sieved through a 0.5 mm mesh, and stored in PE bags at -40 ^{o}C. The moisture content of the garden-collected *C. asiatica* was

determined to be 3.23 ± 0.39% and not significantly different from that of the VietGAP sample (4.03 ± 0.11%), these values are within the range (<13%) permitted by the Vietnamese pharmacopeia. In the extraction process, the plant powder (0.5 g) was dispersed in an extraction solvent at a solid/liquid ratio of 1/40 (w/v) under ultrasonic (Elmasonic S120H, Elma Schmidbauer GmbH, Germany) at a frequency of 37 kHz. Then, the mixture was filtered and centrifuged at 5,000 rpm for 20 min (Hermle Z 206 A, Germany), then the clear solution was collected for bioactivity tests. Double distilled water, methanol (MeOH), and ethanol (EtOH) were tested as extraction solvents. The effects of extraction time (30−60 min) and extraction temperature (30−50 ^{o}C) were investigated.

2.3. Determination of antioxidant activity

The antioxidant activity was assessed using the DPPH assay [10]. One milliliter of the extract was added to 1.5 mL of EtOH solution containing 50 μg/mL DPPH$^{\bullet}$ free radicals. The mixture was shaken vigorously and incubated in the dark for 30 min. The absorbance was measured at 517 nm. The results were expressed as % DPPH free radical inhibition as:

$$H\% = \frac{OD_{control} - OD_{sample}}{OD_{control}} \times 100$$

In which, $OD_{control}$ is the absorbance of the mixture of pure solvent and DPPH$^{\bullet}$, OD_{sample} is the absorbance of the mixture of extract sample and DPPH$^{\bullet}$. The IC_{50} value was determined from the curve of lnC versus DPPH$^{\bullet}$ free radical scavenging efficiency. IC_{50} is the sample concentration value at which 50% of free radicals are inhibited.

2.4. Determination of total saponin content (TSC), total flavonoid content (TFC), and total phenolic content (TPC)

TSC was determined using the spectrophotometric method following the procedure described by Miao et al. [7] with a slight modification. A small amount of the extract was evaporated, then 0.2 mL of 5% (w/w) vanillin reagent and 0.8 mL of perchloric acid (72%) were added and incubated at 70 °C for 20 min. Then, the mixture was cooled rapidly with ice for 2 min and mixed with the acetic acid. The light absorption of the solution was measured at 560 nm on a UV-Vis spectrometer (JENWAY's 6705 UV/Vis series UV-Vis, England). TSC was calculated by using an oleanolic acid standard curve (5−30 μg/mL), expressed as milligram oleanolic acid per gram dry sample weight (mg oleanolic/g)

TFC was determined by the aluminum chloride method following the report by Siddhuraju et al. [8] with a slight modification. A volume of 0.5 mL extract was placed in a 10 mL volumetric flask containing 4 mL distilled water. Next, 0.3 mL of $NaNO_2$ 5% (w/w) was added, followed by adding 0.3 mL of $AlCl_3$ 10% (w/w) after 5 min and NaOH 1M after 6 min. The mixture was incubated in darkness for 10 min and the absorption was measured at 510 nm. TFC was expressed as milligram rutin per gram dry weight (mg RE/g) based on the rutin standard curve (50−1,000 μg/g).

TPC was determined by the Folin-Ciocalteu method [9] with a minor modification. The extract (0.5 mL) was placed in a 10 mL beaker, added with 2.5 mL of 10% Folin (w/w), and incubated in the dark for 3 min followed by adding 2 mL Na_2CO_3. The mixture was incubated in the dark for 30 min and the absorption was measured at 756 nm. A standard curve of gallic acid was constructed in the range of 5−60 μg/g. TPC was expressed in milligrams of gallic acid per gram dry weight (mg GAE/g). All the experiments were done in triplicates, the data are shown as mean values ± SD.

2.5. Antibacterial activity

The antibacterial inhibition capacity of *C. asiatica* extract was tested against *Staphylococcus aureus* (Gram-positive bacteria) and *Escherichia coli* (Gram-negative bacteria). The agar well diffusion method as reported by Idris et al. [11] was applied with a minor modification. The bacterial strains were grown in MHB medium (autoclaved at 121 ^{o}C, 1 atm for 15 min), incubated for 24 h in a shaking cabinet at 200 rpm at 37 ^{o}C, and reached a cell count density of about 1 x 10^8 CFU/mL (0.5 McFarland standard). The bacterial concentration was adjusted by adding more medium or increasing the incubation time to obtain the OD value at 625 nm of about 0.2 ± 0.02.

To prepare samples for the antibacterial test, methanolic and ethanolic extracts were prepared under ultrasonic (45 min, 40 °C). The extracts were filtered and centrifuged at 5,000 rpm for 20 min. The clear solution was then collected and evaporated at 50 ^{o}C to obtain a viscous paste. The concentrated extract was diluted with a suitable solvent for the tests. The antibiotic chloramphenicol dissolved in 99.5% ethanol at a concentration of 5 ppm was used as a positive control. The bacterial suspension of each strain (0.1 mL) was spread onto the surface of MHA agar with a spreader. A sterile cotton swab was used to spread the bacteria over the agar surface 3 times with each turn of the plate 60 ^{o} to ensure even distribution. Three wells of 6 mm sizes were created and injected each with 40 uL of the negative control (solvent), the positive control (antibiotic-chloramphenicol 5 ppm), and the extract sample, respectively. Then they were incubated at 37 ^{o}C for 24 h and the data were collected after the incubation period. The experiments were carried out in triplicates and the data are presented as means ± SD.

3. RESULTS AND DISCUSSION

3.1. Effect of extraction conditions on antioxidant activity of *C. asiatica* extract

The DPPH free radical assay is ordinarily used to evaluate the antioxidant activity through an IC_{50} value (a sample concentration required to inhibit 50% of DPPH radicals). The more active antioxidants in the extract, the lower the IC_{50} value. The effect of extracting solvent composition on the antioxidant of *C. asiatica* extract was first investigated. Six composition recipes were tested, including water, methanol, ethanol, ethanol/water (4:1 v/v), ethanol/water (3:2 v/v) ethanol/water (2:3 v/v). We found that only the extract prepared in ethanol/water (3:2 v/v) gave an IC_{50} below 1,000 μg/mL, thus this mixture was used for the following tests. With the ethanol/water (3:2 v/v) extraction solvent, the ultrasonic extraction time was examined at 30, 45, and 60 min. The IC_{50}, and thus the antioxidant activity of the extracts, changed considerably according to the extraction time as seen in Figure 2A. When the extraction time increased from 30 min to 45 min, the IC_{50} decreased from 1,155.47 μg/mL to 910.63 μg/mL and raised slightly to 1,149.31 μg/mL with 60 min. These results indicated that 45 min and 40 ^{o}C are suitable for the antioxidant activity-targeted extraction of *C. asiatica* when using ethanol/water (3:2 v/v) as the extracting solvent. With a shorter or longer extraction time, the extract had a lower DPPH radical scavenging activity.

Temperature is an important factor governing the solid-liquid mass transfer rate. The increase in temperature may decrease the viscosity of solvents and reduce the cell barrier due to the weakening of cell walls and cell membranes; thus, making a solvent easily penetrate, diffuse inwards, and contact with active substances. However, a temperature higher than a certain threshold can result in the deterioration of active components. This study shows that 40 ^{o}C is the most appropriate in the examined range (30−50 ^{o}C). The antioxidant activity decreased gradually as the order: 40 ^{o}C > 30 ^{o}C > 50 ^{o}C (Figure 2B). According to the results, the appropriate extraction conditions for this kind of *C. asiatica* are proposed: ethanol/water (3:2 v/v), an extraction time of 45 min, and an extraction temperature of 40 ^{o}C.

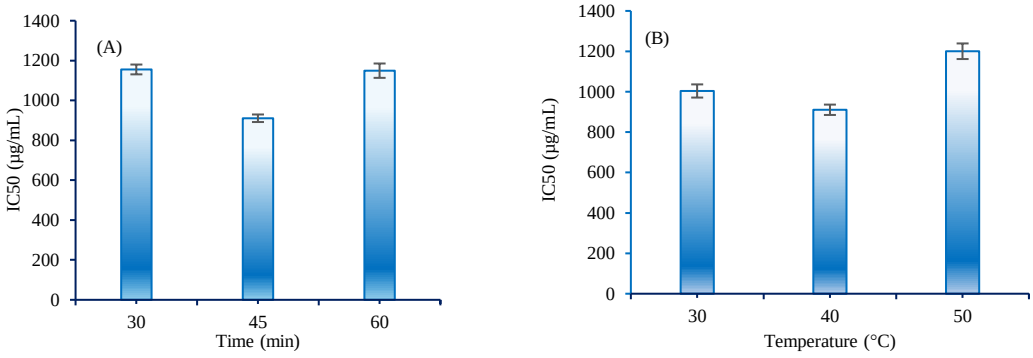

Figure 2. Effect of extraction time (A) and temperature (B) on IC50 values of the *C. asiatica* extract using ethanol/water (3:2 v/v) as the solvent. Each value is presented as mean ± SD.

With the as-determined conditions, the VietGAP *C. asiatica* sample was extracted and tested for antioxidant activity. The IC_{50} of the VietGAP *C. asiatica* extract was 2,545.55 ± 100.56 µg/mL, indicating that the antioxidant activity of the extract of *C. asiatica* from the local garden was significantly higher than that of the VietGAP *C. asiatica* sample used in this study, and also higher than that of the extract of *C. asiatica* studied by Quyen N.T.C. et al. [12]. However, the extraction of *C. asiatica* collected from Can Tho province [13] using 96% ethanol under reflux conditions had a higher antioxidant activity (Table 1).

Table 1. Comparison of antioxidant activity and active compounds of *C. asiatica* extracts

Sample	Sampling area	IC_{50} (µg/mL)	TSC (mg oleanolic/g)	TFC (mg RE/g)	TPC (mg GAE/g)	Conditions	Reference
C. asiatica	Tien Giang	1,057.10	24.15 ± 0.04	21.25 ± 0.46	22.17 ± 0.63	EtOH/water (60:40, v/v), ultrasonic, 45 min, 40 °C	This study
VietGAP *C. asiatica*	Tien Giang	2,545.55	26.89 ± 0.18	13.02 ± 0.38	14.77 ± 0.55	EtOH/water (60:40, v/v), ultrasonic, 45 min, 40 °C	This study
C. asiatica	Ho Chi Minh City	1,744.77	-	-	2.14 ± 0.29	EtOH, 1 h at 70 °C	[12]
C. asiatica	Can Tho	754.61	-	-	-	96% EtOH, reflux extraction.	[13]

**The data obtained from this study are presented as means ± SD values.*

3.2. Determination of TSC, TFC, and TPC in the *C. asiatica* extracts

Figure 3 illustrates the effect of extraction conditions on the extracted contents of active groups: saponins, flavonoids, and phenolics. A mixture of ethanol/water (3:2 v/v) provided the highest TSC (32.53 ± 2.93 mg/g). The temperature at 40 °C and extraction time at 45 min provided the highest saponin content. The highest TPC (26.47 ± 1.85 mg/g) was obtained in ethanol/water (3:2 v/v) at 60 min and 40 °C, while the highest TFC (34.27 ± 0.71 mg/g) was obtained in methanol solvent at 40 °C and 45 min. It is worth mentioning that TSC changed synchronously with the antioxidant activity but until now there has been unclear evidence linking antioxidant activity and saponins. Meanwhile, both TPC and TFC have been widely reported to be responsible for antioxidant activity.

Under the same extraction conditions, the TFC and TPC of the VietGAP sample were about 13.02 ± 0.38 mg/g and 14.77 ± 0.55 mg/g, respectively, and lower than those found for the sample from the garden. However, the TSC of the VietGAP sample was higher (26.89 ± 0.18 vs. 24.15 ± 0.04 mg/g). As mentioned above, the antioxidant activity of the VietGAP sample was much lower when compared to the one cultivated under normal local conditions. This result strongly suggests the considerable influence of TFC and TPC but not TSC in the antioxidant activity of the *C. asiatica* extracts.

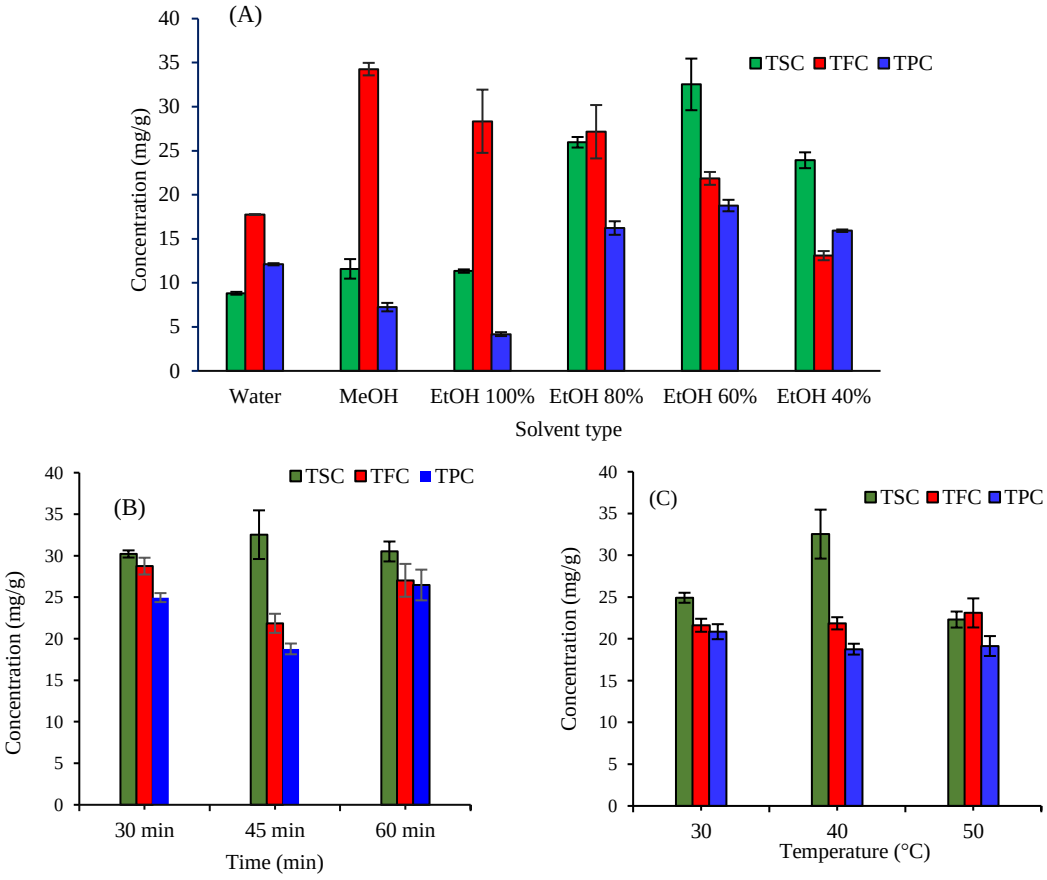

Figure 3. Effect of extraction conditions on the content of active compounds: solvent type (40 ºC, 45 min) (A); extraction time (ethanol/water (3:2 v/v), 40 ºC) (B); extraction temperature (ethanol/water (3:2 v/v), 45 min) (C). Each value is expressed as mean ± SD.

3.3. Antibacterial activity

Antibacterial tests were performed against *Staphylococcus aureus* (Gram-positive bacteria) and *Escherichia coli* (Gram-negative bacteria). To prepare samples for antibacterial tests, water-free solvent systems were applied to achieve highly concentrated extracts to obtain sufficient antibacterial activity. The results showed that concentrated ethanol extract (CEE) exhibited a stronger antibacterial activity than concentrated methanol extract (CME). The inhibition diameters of CEE against *Escherichia coli* and *Staphylococcus aureus* were around 9.0 mm and 7.4 mm, respectively (Table 2).

Table 2. Antibacterial activity of *C. asiatica* extracts against *Staphylococcus aureus* and *Escherichia coli*

Extracting solvent	Diameter* (mm)		Reference
	Staphylococcus aureus	*Escherichia coli*	
Methanol	–	7.27 ± 0.25	This study
Ethanol	7.43 ± 0.35	9.03 ± 0.15	This study
Ethyl acetate	10.07 ± 0.11	8.53 ± 0.15	Sellathoroe et al. [14]

*(–) No growth inhibition zone; *Values are presented as means* ± SD.

The inhibitory effect of CME was found on *Escherichia coli* but not *Staphylococcus aureus*. This result of the CEE is quite comparable to that reported previously by Sellathoroe et al. [14], in which the concentrated ethyl acetate extract (70 ^{o}C, 5 h) exhibited antibacterial activity against *Staphylococcus aureus* and *Escherichia coli*. According to the previous report of Idris and Nadzir [1], components such as asiaticoside and asiatic acids in *C. asiatica* are more likely responsible for the inhibitory effect against *Staphylococcus aureus* and *Escherichia coli.*

4. CONCLUSIONS

This study provided valuable supportive data to demonstrate the effect of extraction conditions and the importance of cultivation conditions on the composition and bioactivity of *Centella asiatica* (L.) Urb. extract. In the ultrasonic-assisted extraction of *Centella asiatica*, the appropriate conditions were determined including ethanol/water (3:2 v/v) solvent for antioxidant activity, methanol solvent for antibacterial activity, temperature of 40 ^{o}C, and extraction time of 40 min. The inhibition effect of the concentrated ethanolic extract was found against *Staphylococcus aureus* and *Escherichia coli*. The phytochemical content of the extracts was also varied depending on the extraction conditions. The highest contents of saponins and polyphenols were obtained in the ethanol/water (3:2 v/v) solvent, while the highest content of flavonoids was achieved in methanol extraction. In the same region, *Centella asiatica* samples collected from the local garden and VietGAP exhibited remarkable differences in phytochemical content and antioxidant activity. Therefore, further studies on the effects of cultivation and extraction conditions on antibacterial and pharmaceutical activities are suggested.

REFERENCES

1. Idris F.N., Mohd Nadzir M. - Comparative studies on different extraction methods of *Centella asiatica* and extracts bioactive compounds effects on antimicrobial activities. Antibiotics **10** (2021) 457. https:// doi.org/10.3390/antibiotics10040457
2. Rupa D.B., Komal M.P., Prakash R.I., Satyendra K.P. - Phytochemical and pharmacological evaluation of organic and non-organic cultivated nutritional *Centella asiatica* collected after different time intervals of harvesting. South African Journal of Botany **112** (2017) 237-245. https://doi.org/10.1016/j.sajb.2017.06.003
3. Kashmira J.G., Jagruti A.P., Anuradha K.G. - Pharmacological review on *Centella asiatica*: a potential herbal cure-all. Indian Journal of Pharmaceutical Sciences **72** (5) (2010) 546–556. https://doi.org/10.4103/0250-474X.78519
4. Minarti, Megawati, Sofa F. - Total phenolic, total flavonoid content, and α-glucosidase inhibitory activity of *Centella asiatica* (L.) Urb. Advances in Health Sciences Research **40** (2021) 222–226. https://doi.org/10.2991/ahsr.k.211105.033
5. Duval, C. - Method for preparing extract of *Centella asiatica*. U.S. Patent 948,755,2B2 (2011). https://patents.google.com/patent/US9487552B2/en
6. Loiseau, A.; Sene, G.; Theron, E. - Method for preparing *Centella asiatica* extract rich in madecassoside and in terminoloside. U.S. Patent 848,690,0B2 (2013). https://patents.google.com/patent/US7402669B2/en
7. Jing Miao, Chengcheng Zhao, Xia Li, Xuetao Chen, Xinhui Mao, Hanhan Huang, Tingting Wang, Wenyuan Gao - Chemical composition and bioactivities of two common *Chaenomeles* fruits in china: *Chaenomeles speciosa* and *Chaenomeles sinensis*. Journal of Food Science **81** (8) (2016) H2049-H2058. https://doi.org/10.1111/1750-3841.13377
8. Perumal S., Klaus B. - Antioxidant properties of various solvent extracts of total phenolic constituents from three different agroclimatic origins of drumstick tree (*Moringa oleifera* Lam.) leaves. Journal of Agricultural and Food Chemistry **51** (8) (2003) 2144–2155. https://doi.org/10.1021/jf020444

9. Luciana L. M., Fábio S. M., Gilda G. L., Alexandre S. R., Tereza C. S., Cintia S. C., Suzana G. L. - Screening of Brazilian plant extracts for antioxidant activity by the use of DPPH free radical method. Phytotherapy Research **15** (2) (2001) 127–130. https://doi.org/10.1002/ptr.687
10. Singleton V. L., Joseph A. R. - Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. American Journal of Enology and Viticulture **16** (1965) 144-158. https://doi.org/10.5344/ajev.1965.16.3.144
11. Idris F. N., Nadzir M. M. - Antimicrobial activity of *Centella asiatica* on *Aspergillus niger* and *Bacillus subtilis*. Chemical Engineering Transactions **56** (2017) 1381-1386. https://doi.org/10.3303/CET1756231
12. Quyen N. T. C., Quyen N. T. N., Quy N. N., Quan P. M. - Evaluation of total polyphenol content, total flavonoid content, and antioxidant activity of *Centella asiatica*. IOP Conf. Series: Materials Science and Engineering **991** (2020) 012020. https://doi:10.1088/1757-899X/991/1/012020
13. Dung N. T., Ngoc T.K. - Studying on screening antioxidant activity of herbal plants in Can Tho city. Journal of Scientific Research and Economic Development **1** (2017) 143–152. https://tdu.edu.vn/TAPCHIKHOAHOC/so1_2017/13.pdf
14. Sellathoroe S., Marimuthu S., Ramays T. R. - Comparison of different extraction methods to study the antimicrobial activity of *Centella asiatica* leaf extracts. International Journal of Advanced Research **7** (5) (2019) 344–347. http://dx.doi.org/10.21474/IJAR01/9044.

TÓM TẮT

CHIẾT XUẤT RAU MÁ (C*entella asiatica* (L.) Urb.): ẢNH HƯỞNG CỦA ĐIỀU KIỆN CHIẾT ĐẾN HOẠT TÍNH SINH HỌC VÀ THÀNH PHẦN HÓA THỰC VẬT

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Mục tiêu của nghiên cứu là phân tích sơ bộ thành phần hóa thực vật và đánh giá các yếu tố ảnh hưởng đến hoạt tính sinh học của dịch chiết cây rau má (*Centella asiatica* (L.) Urb.). Các mẫu rau má được thu thập tại tỉnh Tiền Giang, mẫu được thu thập từ 2 nguồn là mẫu từ vườn rau ở địa phương và mẫu VietGAP ở cùng khu vực. Kết quả nghiên cứu thể hiện sự ảnh hưởng lớn của dung môi chiết đến hàm lượng các nhóm chất hoạt tính và hoạt tính sinh học của dịch chiết thu được bằng kỹ thuật chiết có hỗ trợ bằng siêu âm. Hệ dung môi chiết ethanol/nước (3:2 v/v) cho dịch chiết với hoạt tính kháng oxy hóa cao nhất, trong khi đó dung môi ethanol cho dịch chiết có tính kháng khuẩn. Các điều kiện chiết phù hợp, định hướng thu dịch chiết có khả năng kháng oxy hóa, được xác định bao gồm ethanol: nước (3:2 v/v), thời gian chiết 45 phút và nhiệt độ 40 ^{o}C. Trong điều kiện này, tổng hàm lượng saponin, tổng hàm lượng phenolic và tổng hàm lượng flavonoid của dịch chiết thu được lần lượt là 24,15 ± 0,04 mg oleanolic/g, 22,17 ± 0,63 mg GAE/g, và 21,25 ± 0,46 mg RE/g. Bên cạnh đó dịch chiết từ mẫu *C. asiatica* VietGAP có hàm lượng saponin cao hơn, trong khi mẫu *C. asiatica* từ vườn rau của địa phương cho hàm lượng phenolic và flavonoid cao hơn.

Từ khóa: Cây rau má, *Centella asiatica*, hoạt tính kháng khuẩn, hoạt tính kháng oxy hóa, thành phần hóa thực vật.