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RESEARCH ARTICLE

Secondary Metabolites from *Usnea* sp. and an Evaluation of Their Cytotoxic and Antibacterial Activities

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ABSTRACT

The genus *Usnea* was reported to synthesize the bioactive secondary metabolites, including cytotoxic and antibacterial activities. To continue our study on bioactive compounds from *Usnea* sp, we studied secondary metabolites and their bioactivities of *Usnea* sp. The aim of this study was to determine the chemical constituents of lichen *Usnea* sp. Methanol extract of *Usnea* sp. was prepared by solid liquid extraction followed by fractionation and purification of the crude extract by several chromatographic techniques using *n*-hexane and ethyl acetate as solvents to obtain two pure compounds (**1** and **2**). The structures of both secondary metabolites were analyzed by 1D and 2D (one and two-dimensional) NMR spectroscopy and identified as 2,6-dihydroxy-4-methylbenzoic acid **(1)** and (-)-placodiolic acid **(2)**. Cytotoxic activity of compounds against MCF-7 cells was evaluated using the MTT assay (IC_{50} = 554.82 and 5.10 μM, respectively). Both compounds were also assayed for their antibacterial activity using disc diffusion method. Inhibition zones of compound **1** against *S. aureus*, *E. coli*, and *S. pyogenes* were 6.37 ± 0.21, 7.57 ± 0.15, and 5.43 ± 0.12 mm, respectively. In addition, compound **2** inhibited the growth of *S. aureus*, *E. coli*, and *S. pyogenes* bacteria (Inhibition zones = 10.43 ± 0.25, 11.63 ± 0.21, and 10.63 ± 0.21). The result of biological activities demonstrated that **2** exhibited stronger cytotoxic activity against MCF-7 cells as well as antibacterial activity than compound **1**. Based on the theses results, compound **2** could be a promising candidate of antibiotics in the near future.

Keywords: Antibacterial, cytotoxicity, MCF-7 cells, Secondary metabolites, Usnea sp

Introduction

Usnea is a lichen genus in family Permeliaceae growing like leafless mini-shrubs or tassels anchored on bark or twigs. Traditionally, *Usnea* sp. has been used as medicine including antimicrobial and anticancer agents.[1,2] Most of the lichen *Usnea* sp. scattered in the tropics and others in the sub-tropics. The morphology of the *Usnea* is a branched thread-like tallus shape, greenish-white color, and vertical growth.[3,4]

Previous phytochemical study reported that *Usnea* sp. produced benzofuran derivatives, depsides, depsidones, terpenoids, steroids, and phenolic compounds.[5–7] Some of the reported compounds exhibited several activities including anti-inflammatory, antibacterial, antifungal as well as cytotoxic activity.[8–10] Two isolated compounds namely usnic acid and diffractic acid from *Usnea* sp. exhibited antioxidant activity with IC_{50} values of 7.67 and 18.51 μg/ml, respectively.[11] In addition, usnic acid obtained from *U. longissimi* Ach. inhibited the growth of *E. coli* (ATCC35218) and *S. aureus* (ATCC25923) with inhibition zones between 12 mm and 17 mm.[12] Diffractic acid isolated from *U. blepharea* Motyka actively inhibited (very strongly) the growth of *S. aureus*

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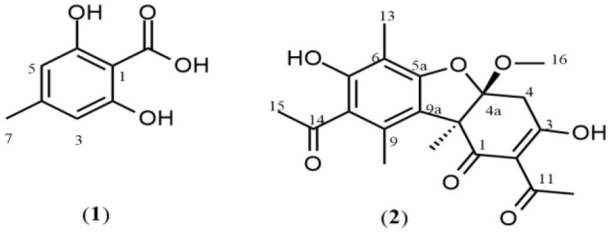

(1) (2)

Fig. 1. Structures of compounds 1-2 isolated from *Usnea* sp.

at concentrations of 750 and 1,000 ppm with inhibition zones of 14.27 and 17.25 mm, respectively. In addition, *E. coli* was also actively inhibited (strongly) at concentrations of 750 and 1,000 ppm at 11.00 and 12.75 mm, respectively.[13]

Furthermore, (-)-usnic acid from *Usnea* sp. showed cytotoxicity against murine leukemia P388 cells with IC_{50} 5.738 ± 0.61 μg/mL.[14] Four new tetrahydroxanthone–chromanone heterodimers, usneaxanthones E–H as well as eleven known compounds were isolated from lichen *Usnea aciculifera* Vain and showed potent cytotoxicity against HCT116 colon cancer with IC_{50} values from 3.37 to 4.53 μM.[15] The previous data of cytotoxic and antibacterial activities by isolated compounds from *Usnea* indicated that *Usnea* is source of cytotoxic and antibacterial compounds.

In continuing study for searching bioactive compounds from lichen *Usnea* sp., we have studied the chemical constituents on lichen *Usnea* sp. In this study, two secondary metabolites as in Fig. 1 have been isolated from the methanol extract of *Usnea* sp. Spectroscopic analysis was used to determine the structures of isolated compounds. Cytotoxic and antibacterial activities of both compounds were also assayed. Interestingly, compound **1** is firstly reported from *Usnea* sp. herein.

Materials and methods

Instrumentation

An Agilent Varian spectrometer (Agilent Technologies, United States) at 500MHz (^{1}H-), and 125 MHz (^{13}C-) using Me_4Si as reference and CD_3OD as solvent was used to obtain the NMR spectra. An Autopol IV polarimeter (Rudolph Research Analytical, United States) was used to determine the optical rotation. Silica gel plates (Silica gel 60 F254, 20×20 cm, Merck, Germany) was used for thin layer chromatography. Silica gel Kieselgel 60, 0.04–0.063 mm (Merck, Germany) was used to vacuum liquid chromatography (VLC) and column chromatography. UV detector (254 and 336 nm) was used to detect the TLC spots.

Lichen material

Lichen *Usnea* sp. was collected on December 2020 from Ciwidey, South Bandung, West Java, Indonesia, latitude: 7.0849° S, longitude: 107.4461° E, elevation: 1169 m.

Extraction and isolation

The dried Lichen *Usnea* sp. (2 kg) was extracted with methanol[6] to obtain crude extract (30.8 g) which was tested by their cytotoxic activity. The crude extract (20 g) was fractionated using VLC technique (solvents: gradient system (*n*-hexane and ethyl acetate = 100:0 – 0:100))[16] to give nine major fractions (A-I). Two largest mass fractions (H and I) were further fractionated. Fraction H (1.33 g) was further fractionated over silica gel vacuum liquid chromatography (solvents: with *n*-hexane and ethyl acetate (100:0 – 0:100)) to result 16 sub-fractions (H.1-H.16). Sub-fractions H.15 and H.16 were subjected to silica gel radial chromatography (solvents: gradient system of *n*-hexane and ethyl acetate) obtaining compound **1** (1.2 mg).

Fraction I (1.98 g) was fractionated over silica gel vacuum liquid chromatography (solvents: gradient system of *n*-hexane and ethyl acetate) to give 25 sub-fractions (I.1–I.25). Sub-fractions I.9, I.10, and H.11 were subjected to silica gel radial chromatography (solvents: *n*-hexane and ethyl acetate (7:3)) obtaining compound **2** (12.8 mg).

Characteristic data of isolated compounds

2, 6-dihydroxy-4-methylbenzoic acid (1)

Yellow solid (1.2 mg). Soluble in acetone. Rf = 0.65 (n-hexane and ethyl acetate = 1:1). ^{1}H-NMR ($(CD_3)_2CO$ at 500 MHz): δ 13.99 (s, 1H, -COOH), 11.46 (s, 2H, 2-OH dan 6-OH), 6.99 (s, 2H, H-3/H-5), and 2.79 (br, 3H, H-7). ^{13}C-NMR ($(CD_3)_2CO$ at 125 MHz): 172.3 (C10), 160.7 (C4/6), 146.3 (C5), 107.9 (C1/3), 98.9 (C2), and 21.6 (C9).

(-)-Placodiolic acid (2)

Yellow solid (1.2 mg). Soluble in acetone. Rf = 0.70 (n-hexane and ethyl acetate = 1:1). ^{1}H-NMR ($(CD_3)_2CO$ at 500 MHz): δ 13. 44 (s, 1H, 7-OH), 11.46 (s, 1H, 9-OH), 3.53 (s, 3H, H-16), 3.14 (s, 1H, 3-OH), 3.04 (s, 2H, H-4), 2.54 (s, 3H, H-15), 2.31 (s, 3H, H-12), 1.92 (s, 3H, H-13), and 1.50 (s, 3H, H-10). ^{13}C-NMR ($(CD_3)_2CO$ at 125 MHz): δ 202.0 (C-14), 198.9 (C-11), 191.8 (C-3), 163.5 (C-7), 163.0 (C-9), 161.2 (C-5), 157.9 (C-1), 114.1 (C-2), 111.8 (C-4a), 108.0 (C-9a), 105.6 (C-6), 101.8 (C-8), 59.9 (C-9b),

51.6 (C-16), 42.4 (C-4), 32.0 (C-12), 31.3 9C-15), 19.3 (C-10), and 7.5 (C-13).

In-vitro cytotoxic assay

Both secondary metabolites (**1** and **2**) were assayed for their cytotoxicity against MCF-7 breast cancer line using MTT method[16–19] with modification. Cells of MCF-7 in 96-well plates with cell density of 3×10^4 cells/cm^3 were incubated for 24 hours. Then, the compounds with several concentrations were added and incubated for 48 h. Every compound was dissolved in dimethyl sulfoxide (DMSO). Negative control wells received only DMSO. Then, 10 μL MTT reagents were added into each well. After 4 h incubation, the MTT-stop solution containing SDS (Sodium Dodecyl Sulfate) were added. After 24 h, a microplate reader ($\lambda = 550$ nm) was used to determine the optical density.

Antibacterial assay

Two secondary metabolites were evaluated for their antibacterial activity with disc diffusion method following the reported procedure[20,21] with modification. *Staphylococcus aureus*, *Escherichia coli*, and *Streptococcus pyogenes* were used for tested bacteria. Amoxicillin was used as a positive control. 15 ml of MHA media were poured into Petri dishes followed by inoculation of tested bacteria. Sterile paper disc (6 mm) was placed on the MHA media. Serial concentrations of isolated compounds were dropped on the paper disc and incubated for 24 hrs. The inhibition zone of isolated compounds was performed three times. Statically, the zone of inhibition was calculated and presented as mean ± standard deviation.[20]

Results and discussion

Compound **1** was isolated as a yellow solid. Spectrum of ^{1}H-NMR in Table 1 of **1** showed signals for a tetrasubstituted benzene ring [$\delta_H = 6.98$ ppm (2H, *brs*), H-3/H-5] where both protons were symmetrical H atoms, a methyl proton ($\delta_H = 2.79$ ppm, 3H, s, H-7), and a COOH proton at $\delta_H = 13.99$ ppm (1H, s). The fragment of compound **1** based on the spectrum of ^{1}H-NMR indicated the compound **1** was the 1, 2, 4, 6-tetrasubstituted benzene where the substituents of it were two hydroxyl groups, a methyl, and a carboxylic group. The result of comparing this data with literature confirmed compound **1** as 2, 6-dihydroxy-4-methylbenzoic acid Fig. 1.

Several phenolic compounds which have similar structure with compound **1** have been isolated from various species of *Usnea*. *Usneaceratina B* which has a different position of the hydroxyl group with compound **1** was isolated from *Usnea ceratina*.[22] Furthermore, divaric acid and divaricatic acid with propyl group at C-6 have been isolated from *Usnea barbata* 2017-KL-10.[23] However, the chemical study of compound **1** from *Usnea* sp. first reported in this study. The isolation of compound **1** in this research completed the phytochemical profile of Lichen *Usnea* sp.

Table 1. ^{1}H-NMR data of compound 1.

Position	^{1}H (δ_H (ppm), i, m, J)
1	–
2/6	–
3/5	6.98 (2H, s)
4	–
7	2.79 (3H, *brs*)
-OH	11.46 (2H, s)
-COOH	13.99 (1H, s)

Compound **2** was isolated as a powder (yellow). Spectrum of ^{1}H-NMR of **2** displayed three hydroxyl signals [$\delta_H = 13.44$ ppm (1H, s), $\delta_H = 11.46$ ppm (1H, s), and $\delta_H = 3.14$ ppm (1H, s)], a methoxy group at $\delta_H = 3.53$ ppm (3H, s), four methyls [$\delta_H = 2.54$ ppm (3H, s), $\delta_H = 2.31$ ppm (3H, s), $\delta_H = 1.92$ ppm (3H, s), and $\delta_H = 1.50$ ppm (3H, s)] and a methylene signal at $\delta_H = 3.04$ ppm (2H, s). Spectra of 1D NMR (^{1}H- and ^{13}C-) as well as HSQC of compound **2** showed signals for three carbonyl carbons (δ_C 202.0, 198.9, and 191.8 ppm), four sp^2 oxy quaternary carbons (δ_C 163.5, 163.0, 161.2, and 157.9 ppm), four sp^2 oxy

Table 2. ^{1}H and ^{13}C-NMR data of compound 2.

Position	^{1}H (δ_H (ppm), i, m, J)	^{13}C (δ_H, ppm)
1	–	157.9 (=C_q)
2	–	114.1 (=C_q)
3	–	191.8 (CO)
4	3.04 (2H, s)	42.4 (CH_2)
4a	–	111.8 (-C_q)
5	–	161.2 (=C_q)
6	–	105.6 (=C_q)
7	–	163.5 (=C_q)
8	–	101.8 (=C_q)
9	–	163.0 (=C_q)
9a	–	108.0 (=C_q)
9b	–	59.9 (-C_q)
10	1.50 (3H, s)	19.3 (CH_3)
11	–	198.9 (CO)
12	2.31 (3H, s)	32.0 (CH_3)
13	1.92 (3H, s)	7.5 (CH_3)
14	–	202.0 (CO)
15	2.54 (3H, s)	31.3 (CH_3)
16	3.53 (3H, s)	51.6 (-OCH_3)
3-OH	3.14 (1H, s)	–
7-OH	13.44 (1H, s)	–
9-OH	11.46 (1H, s)	–

Table 3. The IC_{50} and antibacterial activity (ZI, mm) of compounds 1 and 2.

Compounds	Cytotoxicity (IC_{50}, μM)	Inhibition Zones (mm)		
		S. aureus	*E. coli*	*S. pyogenes*
1	554.82	6.37 ± 0.21	7.57 ± 0.15	5.43 ± 0.12
2	5.10	10.43 ± 0.25	11.63 ± 0.21	10.63 ± 0.21
DMSO (-)	–	0	0	0
Amoxicillin (+)	–	15.57 ± 0.15	16.37 ± 0.21	15.43 ± 0.25

carbons (δ_C 114.1, 108.0, 105.6, and 101.8 ppm), a sp^3 oxy quaternary carbon at δ_C 111.8 ppm, a sp^3 quaternary carbon at δ_C 59.9 ppm, a methoxy carbon (δ_C 51.6 ppm), a sp^3 methylene at δ_C 42.4 ppm, and four methyl carbons (δ_C 32.0, 31.3, 19.3, and 7.5 ppm). Data of 1D-NMR of compound **2** are displayed in Table 2.

Chemical structure of **2** was confirmed by correlation of long correlation of H to C in HMBC spectrum. Methyl group (δ_H 2.54, H-15) was correlated with C-8 (δ_C 101.8), C-14 (δ_C 202.0) indicating the methyl as a part of alkyl group of ester connecting to benzene ring at C-8. Furthermore, a long-range correlation from methyl proton (δ_H 1.50, H-10) to quaternary carbon (C-4a, δ_C 111.8; C-9a, δ_C 108.0 and C-9b, δ_C 59.9) showed C10-C9a connected with a benzene ring and cyclic structure. The result of comparing the NMR data with the literatures[24,25] confirmed compound **2** as (-)-placodiolic acid Fig. 1.

Compound **2** has been reported from the Fungus *Mycosphaerella* sp., isolated from marine sediment. This study also indicated that compound **2** showed antibacterial activity against *Escherichia coli* ATCC 11775, *Klebsiella pneumonia* ATCC 4352, and *Staphylococcus aureus* ATCC 6538[24]. Furthermore, compound **2** was isolated from *Leprocaulon microscopicum* acetone extract. This study was also revised for ^{13}C-assignments of compound **2.**[26]

The pure compounds (**1–2**) was examined for their cytotoxicity against MCF-7 cells with cisplatin as a positive control. The categories of cytotoxic values are active with IC_{50} < 10.00 μM, moderate with IC_{50} 10.00-40.00 μM, and inactive with IC_{50} > 40.00 μM.[27] Based on them, compound **1** was inactive with IC_{50} value of 554.82 μM. In the contrast, compound **2** was active against MCF-7 cells (active, IC_{50} value of 5.10 μM). The strong activity of compound **2** indicated that compound **2** might be useful as anticancer agents.

Both compounds were also evaluated for their antibacterial activity against *S. aureus*, *E. coli*, and *S. pyogenes* using disc diffusion method. Amoxicillin was used as a positive control and DMSO was used as a negative control. Compound **2** showed the greatest inhibition zone at a concentration of 5% against all tested bacteria. Data of biological activities displayed at Table 3.

Conclusion

In the present research, phytochemical study of two secondary metabolites from methanol extract of *Usnea* sp. was described. Both compounds were 2,6-dihydroxy-4-methylbenzoic acid **(1)** and (-)-placodiolic acid **(2)**. Interestingly, compound **1** is isolated from *Usnea* sp. for the first time. Compound **2** exhibited significant cytotoxicity (IC_{50} 5.10 μM) against MCF-7 cells and showed antibacterial activity against three tested bacteria at concentration of 5%. The distinguished result of IC_{50} and inhibition zones by both compounds indicated that compound **2** more potent than compound **1** as cytotoxic and antibacterial activities. In the future, compound **2** could be studied as a promising candidate of anticancer and antibiotic.

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Authors' declaration

- Conflicts of Interest: None.
- We hereby confirm that all the Figures and Tables in the manuscript are ours. Furthermore, any Figures and images, that are not ours, have been included with the necessary permission for republication, which is attached to the manuscript.
- Authors sign on ethical consideration's approval.
- Ethical Clearance: The project was approved by the local ethical committee at University of Universitas Negeri Padang, Padang, Indonesia.
- No animal studies are present in the manuscript.
- No potentially identified images or data are present in the manuscript.

Authors' contribution statement

D.M.A. planned the research work, contributed to the writing of the paper and data collection. R.R. contributed to the writing, data analysis of the paper and publishing the paper as correspondence author. E.H. planned the research work and performed the experiments. B.S. contributed to the writing of the paper and data collection.

References

1. Jannah M, Afifah N, Hariri MR, Rahmawati A, Wulansari TYI. Study of Lichen (Usnea spp.) As A Traditional Medicine in Bogor, West Java. Berkala Penelitian Hayati. 2020;26(1):32–38. https://doi.org/10.23869/bphjbr.26.1.20206.
2. Sepahvand A, Studzińska-Sroka E, Ramak P, Karimian V. *Usnea* sp. Antimicrobial potential, bioactive compounds, ethnopharmacological uses and other pharmacological properties; a review article. J Ethnopharmacol. 2021;268:113656. https://doi.org/10.1016/j.jep.2020.113656.
3. Jannah M, A'yun Q, Afifah N, Prasetya E, Hariri MR. *Usnea* in West Java: a potential source of bioactive secondary metabolites. Berk Penelit Hayati. 2022;28:26–31. https://doi.org/10.23869/bphjbr.28.1.20224.
4. Gerlach A, Clerc P, Lücking R, Moncada B, Nobleza JC, Ohmura Y, *et al.* The genus *Usnea* (Parmeliaceae, Ascomycota) in the Southern Philippines: A First Phylogenetic Approach. The Lichenologist. 2023;55(6):451–480. https://doi.org/10.1017/S0024282923000579.
5. Popovici V, Matei E, Cozaru GC, Aschie M, Bucur L, Rambu D, *et al.* Usnic acid and *Usnea barbata* (L.) F.H. wigg. dry extracts promote apoptosis and DNA damage in human blood cells through enhancing ROS levels. Antioxidants. 2021;10(8):1171. https://doi.org/10.3390/antiox10081171.
6. Salgado F, Albornoz L, Cortéz C, Stashenko E, Urrea-Vallejo K, Nagles E, *et al.* Secondary metabolite profiling of species of the genus *Usnea* by UHPLC-ESI-OT-MS-MS. Molecules. 2018;23(1):54. https://doi.org/10.3390/molecules23010054.
7. Vu TH, Le Lamer AC, Lalli C, Samson JBM, Le Dévéhat FL, Le Seyec J. Depsides: Lichen metabolites active against hepatitis C virus. PLoS One. 2015;10(3):1–14. https://doi.org/10.1371/journal.pone.0120405.
8. Dieu A, Mambu L, Champavier Y, Chaleix V, Sol V, Gloaguen V, *et al.* Antibacterial Activity of the Lichens *Usnea Florida* and *Flavoparmelia caperata*. Parmeliaceae. 2020;34(23):3358–3362. https://doi.org/10.1080/14786419.2018.1561678.
9. Oh JM, Kim YJ, Gang HS, Han J, Ha HH, Kim H. Antimicrobial activity of divaricatic acid isolated from the lichen *Evernia mesomorpha* against methicillin-resistant *Staphylococcus aureus*. Molecules. 2018;23(12):1–9. https://doi.org/10.3390/molecules23123068.
10. Yu X, Guo Q, Su G, Yang A, Hu Z, Qu C, *et al.* Usnic acid derivatives with cytotoxic and antifungal activities from the lichen *Usnea longissimi*. J Nat Prod. 2016;79(5):1373–1380. https://doi.org/10.1021/acs.jnatprod.6b00109.
11. Maulidiyah M, Darmawan A, Ahmad E, Musdalifah A, Wibowo D, Salim LOA, *et al.* Antioxidant activity-guided isolation of usnic acid and diffractaic acid compounds from lichen genus *Usnea* sp. J Appl Pharm Sci. 2021;11(02):075–083. http://dx.doi.org/10.7324/JAPS.2021.110210.
12. Maulidiyah Sabarwati SH, Harjuliarto R, Watoni AH, Nurdin M. Antibacterial activity of usnic acid from *Usnea longissima* Ach. Pak. J Pharm Sci. 2020;33(4):1631. http://dx.doi.org/10.36721/PJPS.2020.33.4.REG.1631-1639.1.
13. Maulidiyah M, Natsir M, Nazila W, Musdalifah A, Salim OA, Nurdin M. Isolation and antibacterial activity of diffractic acid compound from lichen *Usnea blepharea* Motyka. J Appl Pharm Sci. 2021;11(11):121–130. http://dx.doi.org/10.7324/JAPS.2021.1101116.
14. Maulidiyah M, Azis T, Sabarwati SH, Nurdin M. Isolation and Identification of (-)-usnic acid compound from lichen *Usnea* sp. and its cytotoxic activity on murine leukemia P388 Cell. Jurnal Ilmu Kefarmasian Indonesia. 2015;13(1):40–44. http://jifi.farmasi.univpancasila.ac.id/index.php/jifi/article/view/121.
15. Tuong TL, Do LTM, Aree T, Wonganan P, Chavasiri W. Tetrahydroxanthone–chromanone heterodimers from lichen *Usnea aciculifera* and their cytotoxic activity against human cancer cell lines. Fitoterapia. 2020;147:104732. https://doi.org/10.1016/j.fitote.2020.104732.
16. Riga R, Happyana N, Hakim EH. Chemical constituents of *Pestalotiopsis microspora* HF 12440. J Appl Pharm Sci. 2019;9(1):108–124. http://dx.doi.org/10.7324/JAPS.2019.90116.
17. Herlina T, Merlin, AM, Supratman U. Cytotoxic constituents from the bark of *Erythrina poeppigiana* against the MCF-7 breast cancer cell lines. J Nat Prod. 2019;10(3):257–261. http://dx.doi.org/10.2174/2210315509666191115152811.
18. Riga R, Happyana N, Hakim EH. Sesquiterpenes produced by *Pestalotiopsis microspora* HF 12440 isolated from *Artocarpus heterophyllus*. Nat Prod Res. 2020;34(15):2229–2231. https://doi.org/10.1080/14786419.2019.1578764.
19. Roesman DI, Tjong DH, Hanif MM. Cytotoxic Activities, Determining Toxin, and Molecular Docking of Ovary Pufferfish (*Tetraodon leiurus*) in Singkarak Lakeas Cancer Chemoprevention Candidate. Baghdad Sci J. 2023;20(6):2372–2384. https://doi.org/10.21123/bsj.2023.8785.
20. Suryelita S, Riga R, Etika SB, Ikhsan MH, Febria F, Yolanda M, *et al.* Phytochemical Screening and Biological Activities of Fungal *Phyllosticta capitalensis* derived from *Andrographis paniculata*. Mor J Chem. 2023;11(2):553–565. https://doi.org/10.48317/IMIST.PRSM/morjchem-v11i2.33657.
21. Hussein NN, Marzoog TR, Al-Niaame AE. The Antibacterial, Antiheamolytic, and Antioxidant Activities of *Laurus nobilis* and *Alhagi maurorum* Native to Iraq. Baghdad Sci J. 2019;16(3):707–712. https://doi.org/10.21123/bsj.2019.16.3(Suppl.).0707.
22. Bui VM, Huynh BLC, Pham NKT, Nguyen TAT, Nguyen TTT, Nguyen KPP, *et al.* Usneaceratins A and B, two new secondary metabolites from the lichen *Usnea ceratina*. Nat Prod Res. 2022;36(15):3945–3950. https://doi.org/10.1080/14786419.2021.1901288.
23. Chae H-J, Kim G-J, Deshar B, Kim H-J, Shin M-J, Kwon H, *et al.* Anticancer activity of 2-O-caffeoyl alphitolic acid extracted from the lichen, *Usnea barbata* 2017-kl-10. Molecules. 2021;26(13):1–12. https://doi.org/10.3390/molecules26133937.
24. Lee J, Lee J, Kim GJ, Yang I, Wang W, Nam JW, *et al.* Mycousfurans A and B, antibacterial usnic acid congeners from the fungus *Mycosphaerella* sp., isolated from marine sediment. Mar Drugs. 2019;17(7). https://doi.org/10.3390/md17070422.
25. Millot M, Kaouadji M, Champavier Y, Gamond A, Simon A, Chulia AJ. Usnic acid derivatives from *Leprocaulon microscopicum*. Phytochem Lett. 2013;6(1):31–35. https://doi.org/10.1016/j.phytol.2012.10.009.

26. Lee J, Lee J, Kim GJ, Yang I, Wang W, Nam J-W, *et al.* Mycousfurans A and B, antibacterial usnic acid congeners from the fungus *Mycosphaerella* sp., isolated from marine sediment. Marine Drugs. 2019;17(7):422. https://doi.org/10.3390/md17070422.

27. Bahuguna A, Khan I, Bajpai VK, Kang SC. MTT assay to evaluate the cytotoxic potential of a drug. Bangladesh J Pharmacol. 2017;12(2):115–118. http://dx.doi.org/10.3329/bjp.v12i2.30892.

المستقلبات الثانوية من اوسينا اس بي وتقييم أنشطتها السامة للخلايا والمضادة للبكتيريا

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الخلاصة

من المعروف عن جنس اوسينا بأنه يقوم بتجميع المستقلبات الثانوية النشطة بيولوجيًا، بما في ذلك الأنشطة السامة للخلايا والمضادة للبكتيريا. لمواصلة دراستنا حول المركبات النشطة بيولوجيًا من اوسينا اس بي، قمنا بدراسة المستقلبات الثانوية ونشاطها الحيوي في Usnea sp. كان الهدف من هذه الدراسة هو تحديد المكونات الكيميائية لنبات الحزاز اوسينا اس بي مستخلص الميثانول من اوسينا اس بي تم تحضيره عن طريق استخلاص السائل الصلب متبوعاً بتجزئة وتنقية المستخلص الخام بواسطة عدة تقنيات كروماتوغرافية باستخدام الهكسان ن وخلات الإيثيل كمذيبات للحصول على مركبين نقيين (1 و 2). تم تحليل هياكل كل من المستقلبات الثانوية بواسطة التحليل الطيفي للرنين المغناطيسي النووي 1D و2D (أحادي وثنائي الأبعاد) وتم تحديدها على أنها حمض 6،2-ثنائي هيدروكسي -4-ميثيل بنزويك (1) و(-)-حمض بلاكوديوليك (2). تم تقييم النشاط السمي للخلايا للمركبات ضد خلايا MCF-7 باستخدام اختبار MTT (IC50 = 554.82 و5.10 ميكرومتر، على التوالي). تم أيضًا تقييم كلا المركبين لنشاطهما المضاد للبكتيريا باستخدام طريقة الانتشار القرصي. كانت مناطق تثبيط المركب 1 ضد المكورات العنقودية الذهبية والإشريكية القولونية والبكتيريا المقيحة 0.21 ± 6.37 و0.15 ± 7.57 و0.12 ± 5.43 ملم على التوالي. بالإضافة إلى ذلك، يمنع المركب 2 نمو بكتيريا S. aureus وE. coli وS. pyogenes (مناطق التثبيط = 0.25 ± 10.43، 11.63 ± 0.21، و0.21 ± 10.63). أظهرت نتيجة الأنشطة البيولوجية أن 2 أظهر نشاطًا سامًا للخلايا أقوى ضد خلايا MCF-7 بالإضافة إلى نشاط مضاد للبكتيريا مقارنة بالمركب 1. وبناءً على نتائج الأطروحات، يمكن أن يكون المركب 2 مرشحًا واعدًا للمضادات الحيوية في المستقبل القريب.

الكلمات المفتاحية: مضاد للجراثيم، السمية الخلوية، خلايا MCF-7، المستقلبات الثانوية، اوسينا اس بي.