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Compositional Study, Antibacterial And Antioxidant Potential Of *Lepidium draba* L. (Brascicaceae).

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ABSTRACT

The present study was designed to evaluate the phytochemical screening, antimicrobial and antioxidant activities of Lepidium draba The arial parts of *Lepidium draba* showed the presence of alkaloids, terpenoids, tannins,saponins, Leuco anthocyans, triterpenoids and flavonoids. Three known flavonoid glycosides: rhamnocitrin- 3-O-β Dglucoside (1), complanatuside (2), genkwanin- 4'-O-β Dglucoside (3) were isolated from the all parts of Lepidium draba their structures were elucidated by NMR experiments and acid hydrolysis. The antibacterial activity of the extracts was evaluated by disc diffusion assay and the antioxidant activity and phenolic contents of n-butanol extract were evaluated in vitro using spectrophotometer methods.

Keywords: *Lepidium draba*, flavonoids, Brassicaceae, antioxidant activity, antibacterial activity.

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INTRODUCTION

Lepidium draba L. (Brassicaceae) commonly known as whitetop or hoary cress, is a perennial herb that reproduces by seed and by horizontal creeping roots [1]. This plant is native to western Asia, Eastern Europe, North America and Africa including Algeria. Infusion of *L.draba* leaves and seeds have purgative and expectorant effects [2]. It can be found in a variety of soil types where moisture is adequate, grows in a wide range of disturbed habitats including cultivated land, rangeland, pastures, along roadsides, waste areas and is known to particularly thrive in riparian or irrigated areas [3-4]. Previous phytochemical studies of this plant from other continent led to the isolation and characterization of flavonoid glycosides and sulforaphane [5-7]. Only allelopathie[8] and antibacterial activity of *L.draba* were reported [9].But antioxidant activity have never been reported.

The present paper was designed to evaluate the phytochemical screening, antimicrobial and antioxidant activities of *L.draba*. We describe also the identification of four known flavonoids (**1-3**) isolated for the first time from *n*-butanol extract.

MATERIALS AND METHODS

Plant material

Lepidium draba was collected from the area of M'sila in the North Eastern of Algeria in June 2014 and authenticated by Dr. R. Khellaf (Biology Department, University of M'sila, Algeria).

Phytochemical analysis

Preliminary screening of secondary metabolites such as alkaloids, flavonoids, saponins, coumarins, quinons, anthocyans, terpenoids, steroids and sterols were carried out according to the common phytochemical methods described by TREASE, EVANS and HARBORNE [10-11]. (Table1)

Extraction and isolation

The whole dried plant of *L.draba* (300 g) was macerated at room temperature with EtOH/H O_2 (70:30 v/v) for 24 h, three times. After filtration, the filtrates were combined, concentrated and successively re-extracted several times with petroleum ether, chloroform, EtOAc and *n*-butanol to give 0.15, 0.2, 0.6, 4g of the respective residues.

A part of the n-butanol extract (3 g) was chromatographed on a 60-200-mesh silica gel (145 g) column eluted with a gradient of chloroform–MeOH (95:5 to 100% MeOH) to yield 23major fractions (L1 –L23) obtained by combining the eluates on the basis of TLC analysis. Fraction L9 (20 mg) was submitted to preparative TLC on silica gel 60, HF (AcOEt-MeOH-H_2O, 5: 0.5: 0.2 to give rhamnocitrin- 3-O-β Dglucoside (**1**) (5mg)[12], complanatuside (**2**)(6mg) [13]. Fractions L 23 (30 mg) which were submitted to preparative TLC on silica gel 60, HF using AcOEt-MeOH-H_2O 8:1:0.5 to give genkwanin- 4'-O-β Dglucoside (**3**)(4.5mg) [14]. The structures of the isolated compounds were elucidated by UV, ^{1}H and acid hydrolysis experiments. All these results were in good agreement with the literature data [12-14].

Antimicrobial activity

Three bacterial strains with Gram positive were used in this study: *Escherichia coli*, *Staphylococcus aureus* and *Klebsiellapneumoniae*. These test microorganisms were obtained from Pasteur institut (Algiers). Antibacterial potential of chloroform, ethyl acetate and *n*-butanol extracts were assessed in terms of inhibition zone of bacterial growth [15-16].

Total phenolic content assay

The total phenolic content was quantified using a modified version of the assay described by Singleton et al using Folin-Ciocalteu reagent. The concentration of total phenolic compounds was determined as μg of gallic acid equivalents per 1mg of extract [17].

Determination of total flavonoid content

Total flavonoid was estimated according to the method described by Wang et al. Total flavonoid content was calculated as μg of quercetin equivalents per 1mg of extract [18].

DPPH radical-scavenging activity assay

The *n*-butanol extract was subjected to screening for its possible antioxidant activity using DPPH test [19-20]

RESULTS AND DISCUSSION

The results of the screened aerial part from *Lepidium draba* are shown in the table 1.

Table 1: phytochemical screening of *L.draba*

Class of compounds	*Lepidium draba*
Alkaloids	+
Coumarins	-
Saponins	+
Flavonoids	+
Tannins	+
Terpenoids	+
Sterols	+
Quinons	-
Triterpenoids	+
Anthocyans	-
Leucoanthocyans	+

+ = Presence of compounds; - =Absence of compounds

Antimicrobial and antioxidant activities

The evaluation of the antimicrobial activity of extracts against bacterial strains (*Escherichia coli*, *Staphylococcus aureus* and *Klebsiellapneumoniae*) showed that the *n*-butanol extract was the most active, it develop excellent activity against Gram positive strain especially against Escherichia *coli* and *Staphylococcus aureus*.The chloroform and ethyl acetate extracts develop fairly well activity against *Escherichia coli* and *Klebsiellapneumoniae*. Our results are presented in table2

Table 1: Antimicrobial activiy of *Lepidium draba*

Microorganism	Diameter of zones of inhibition (mm)		
	Chloroform extract	Ethyl acetate extract	*n*-Butanol extraxt
Escherichia coli	8	8	18
Staphylococcus aureus	14	14	16
Klebsiellapneumoniae	8	10	14

The *n*-BuOH extract showed fairly well activity in scavenging DPPH (IC_{50} =196.47±2.87 μg/ml) which correlated with their total phenolic content and total flavonoid (70.66±9.23 μg/mg extract of gallic acid equivalents and48.52±33.95μg quercetine equivalents, respectively). The activity of *n*-BuOH was comparable to that of the well-known antioxidant, vitamin C (IC_{50} =5.18±0.12 μg/ml). The other extracts didn't exhibit activity in this assay.

Isolated and identified compounds

Compound1.yellowpowder, UV (MeOH), λmax, nm 347,266; +NaOH; 385,266; +NaOH+5mn 385,266; +$AlCl_3$387,301; $AlCl_3$+HCl 396, 298 ;+NaOAc 375, 266 ;+NaOAc+H_3BO_3 348, 267 after acid hydrolysis of

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compound UV, (MeOH) λmax, nm: 366,268; +NaOH: 415,269; +$AlCl_3$:355,268; +$AlCl_3$/HCl:422,268; +NaOAc: 374,268; +NaOAc/H_3BO_3:365,268

^{1}HNMR (250 MHz, CD_3COD_3) δ (ppm) 8.15(2H,d, *J* = 8.8Hz, H2', H_6'); 6.95(2H, d, *J* = 8.8Hz, H_3', H_5'); 6.65(1H,d, *J* =2.1 Hz, H_8); 6.35(1H,d, *J* = 2.1Hz, H_6); 5.5 (1H, d, *J* = 7.5 Hz, H-1"), 4.1-3.5 (5H, m, H -6", H-5", H-4", H-3", H-2"), (3H, s, 7-OCH_3). This compound was identified as rhamnocitrin- 3-O-β Dglucoside

Compound2.yellowpowder, UV (MeOH) λmax, nm351, 266; +NaOH; 389,270; +NaOH+5mn 389,270; +$AlCl_3$404, 305; $AlCl_3$+HCl 393, 302; +NaOAc 350, 266; +NaOAc+H_3BO_3 347, 267. After acid hydrolysis of compound UV, (MeOH) λmax, 370,270; +NaOH degradation of spectrum; +NaOH+5mn degradation of spectrum; +$AlCl_3$: 370,270; +$AlCl_3$/HCl: 370,270; +NaOAc: 370,268; +NaOAc/H_3BO_3: 350,268

^{1}H NMR (250 MHz, CD_3COD_3) δ (ppm) 8.12(2H,d, *J* = 8.7Hz, H2', H_6'); 6.95(2H, d, *J* = 8.7Hz, H_3', H_5') ; 6.65(1H,d, *J* =2 Hz, H_8) ; 6.37(1H,d, *J* = 2Hz, H_6) ; 5.53 (1H, brs, H-1"), 5.43 (1H, d, *J* = 7.5 Hz, H-1'"),4.1-3.5 (10H, m, H -6", H-5", H-4", H-3", H-2" H -6'", H-5'", H-4'", H-3'", H-2'"), 3.94(3H, s, 7-OCH_3).This compound was identified as complanatuside.

Compound3. Yellow powder, UV (MeOH) λmax, nm349, 266; +NaOH; 386,273; +NaOH+5mn386, 273; +$AlCl_3$398, 307; $AlCl_3$+HCl 395, 303; +NaOAc 351, 267; +NaOAc+H_3BO_3 350, 267. After acid hydrolysis of compound UV, (MeOH) λmax, 370,270; +NaOH degradation of spectrum; +NaOH+5mn degradation of spectrum; +$AlCl_3$: 370,270; +$AlCl_3$/HCl: 370,270; +NaOAc: 370,268; +NaOAc/H_3BO_3: 350,268

^{1}H NMR (250 MHz, CD_3COD_3) δ (ppm) 8.1(2H,d, *J* = 8.8Hz, H2', H_6'); 6.95(2H, d, *J* = 8.8Hz, H_3', H_5'); 6.65(1H,d, *J* =2.2 Hz, H_8); 6.37(1H,d, *J* = 2.2Hz, H_6) ; 6.15(1H,s, H_3); 5.5 (1H, d, *J* = 7.5 Hz, H-1"), 4.1-3.5 (10H, m, H -6", H-5", H-4", H-3", H-2"), 3.92(3H, s, 7-OCH_3).This compound was identified as genkwanin- 4'-O-β Dglucoside.

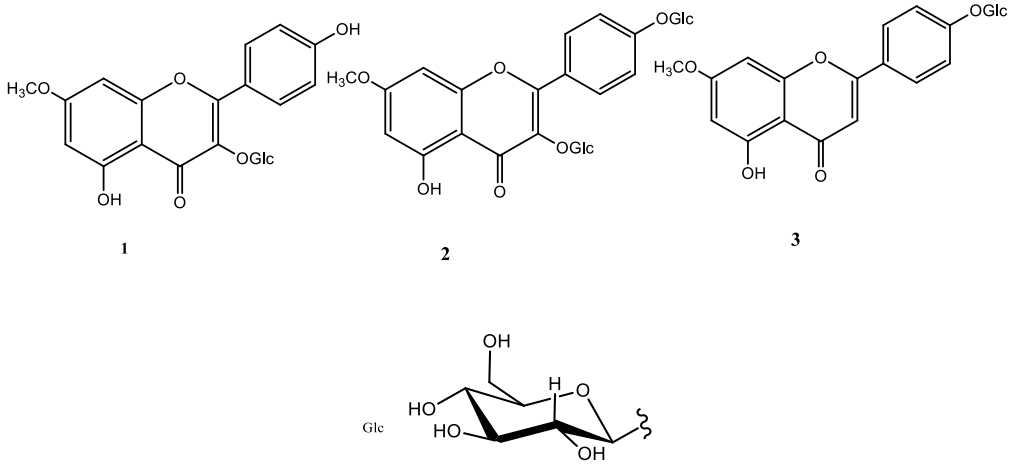

Figure 1: Structures of compounds 1,2 and 3

CONCLUSION

The results of this study demonstrate that active compounds in *Lepidium draba* such as flavonoids, sterols, triterpenoids, saponins exhibited antimicrobial and antioxidant activities. In the aerial parts of *L.draba* three flavonoids were isolated for the first time namely rhamnocitrin- 3-O-β Dglucoside (**1**), complanatuside (**2**), genkwanin- 4'-O-β Dglucoside (**3**)

REFERENCES

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[1] Agriculture Research Service. Cardaria draba (L.) Deav. Selected Weeds of the United States. Agriculture Research Service United States, Department of Agriculture, Washington, DC, (1970), 200.

[2] A. Ghahreman. Basic botany. Tehran: Tehran University Press, (2009), 504.

[3] A. Francis, S.I. Warwick. Can J Plant Sci., 2008; 88:379–401.

[4] G. Scurfield. Cardaria draba (L.) Desv. (Lepidium draba L.). J Ecol., 1962; 50:489–499.

[5] V.A.Kurkin, G.G.Zapesochnaya,P.E.Krivenchuk. Flavonoids of Orobus vernus,Lepidium draba and lepidium ruderale,Khimiya Prirodnykh Soedinenii, 1981;5:661-662.

[6] N.S.Fursa, V.I.Litvinenko, P.E.Krivenchuk. Flavonol glycosides of Lepidium latifolium and Lepidium draba, 1970;6(4):567-571.
[7] Prochazka Z. Isolation of sulforaphane from Lepidium draba.Preliminary commun, Collection of Czechoslovak chemical communications, 1950;24:2429-2430.
[8] A. Miri , J. Sharifi Rad, M. Sharifi Rad and J.A. Teixeira da Silva. Allelopathic activity of medical plant, Cardaria draba (Lepidium draba L.), Annals of Biological Research, 2013;4 (6):76-79.
[9] A.H.AlMarzoqi, N.M.S.Al khafaji and R.A.kadhim. Influence of the crude phenolic alkaloid and terpenoid compounds extracts of cardaria draba (*Lepidium draba* L) on human pathogenic bacteria, 2015;4(6):456-460
[10] G. E.TREASE, I. C. EVANS, (1983), Textbook of Pharmacognosy. 12th Edn., Bailliere Tindall. London; 21-22.
[11] J. B. HARBORN, (1973), Phytochemical Methods: A guide to Modern Techniques of plants Analysis, Chapman & Hall. London, Ltd; pp. 49-188.
[12] L.Jing, Z.Z. Zhen, C.HUbiao. Review of Astragali, *Chinese Herbal Medicines*, 2011;3(2):90-105
[13] L.Wen Tian, Y. Pei, Ying-Jun Zhang, Y.F. Wang, and C.R. Yang, (2009), 7-O-Methylkaempferol and -quercetin Glycosides from the Whole Plant of NerWilia fordii, *J. Nat. Prod*, 72, 1057–1060
[14] T.Iwashina,S.Matsumoto,K.Ozawa,K.Akuzawa (1990),Phytochemistry,Flavone glycosides from Asplenium normale,29(11),3543-3546
[15] S.Abollfazl, N. Hazeri , J.Vlizadeh , Ali Ghasemi and F.Z.Tavallaei,(2012), Phytochemical screening, antimicrobial and antioxidant activities of Anabasis aphylla L. extracts, Kragujevac J. Sci, 34, 71-78.
[16] R.Errakhi, F.Bouteau, A.Lebrihi, M.Barakate,(2007), *World J.Microbiol.Biotechnol*,23,1503.
[17] V.L.Singleton, R.Orthfer,R.M.Lamuela-Raventos(1999), Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin-Ciocalteu reagent.In:PackerL,editor.Methods in enzymol:Oxidant and antioxidants (part A), San Diego,CA:Academic press, 299,152-178.
[18] H.Wang, X.Dong Gao, G.C.Zhou, L.Cai, W.B.Yao. Food Chem, 2008;106:888-895
[19] Soares JR, Dinis TCP, Cunha AP, Almeida LM. Antioxidant activities ofsome extracts of *Thymus zygi*. Free Radical Res., 1997;26:469–478.
[20] Ting Hung-Chih, Yu-Wen Hsu, Chia-Fang Tsai, Fung-Jou Lu, Ming-Chih Chou, Wen-Kang Chen. *in vitro* and *in vivo* antioxidant properties of seabuckthorn (*Hippophae rhamnoides L.*) seed oil. Food Chemistry, 2011;125:652-659.