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Total phenolic and flavonoids content, and *in vitro* antioxidant and antimicrobial activity of ethyl acetate and butanol extract from *Senecio delphinifolius* Vahl.

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ABSTRACT

The present work focuses essentially on the total phenolic and flavonoidscontents, and in vitro antioxidant and antimicrobial activity of two fractions: ethyl acetate and n-butanol extracts of an Algerian endemic plant: Senecio delphinifolius. The total phenolic content was determined using Folin-ciocalteu reagent to yield 7.22 ± 0.84 and 7.86 ± 0.25mg GA/mg respectively. The flavonoids contents were 22.96± 1.41 and 10.25 ± 8.94 mg Q/mg respectively. Antioxidant activity was performed using DPPH reagent. Antioxidant activity ranged from 80.48 % to 73.91 %, using ascorbic acid as a control test. The antimicrobial activity of the two extracts was investigated using a diffusion method against most common pathogenic bacterial strains: E. coli ATCC 2592, Pseudomonas aerogenosa ATCC2783, Staphylococcus aureus ATCC 252923, Selmonella sp., Klebseilla pneumonia and one fungus Candida albicans. The finding show that this extracts have a strong antioxidant activity and can be used as a natural source of scavenging agents.

Key words: *Senecio delphinifolius*Vahl., phenolic and flavonoids content, antioxidant and antimicrobial activity.

INTRODUCTION

The large genus *Senecio* (Tribe Senecionea, Asteraceae family) comprises more than 1500 species [1]. The genus *Senecio* is represented by 18 species in the flora of Algeria [2]. *Senecio* species have been used for treatment of asthma, coughs, bronchitis, eczema and wound healing [3]. Chemical investigations of various *Senecio* species have revealed mainly the presence of sesquiterpenoids, monoterpenoids [4,5], diterpenoids [6], triterpenoids [7], phenolic and flavonoid compounds [8-11], essential oils [12] and pyrrolizidine alkaloids [13] . *Senecio delphinifolius* vahl is an endemic plant found in Sicily (Italy) and in North Africa.

The present work aims to study the:
(i) Total phenolic and flavonoids content
(ii) *in vitro* antioxidant activity,
(iii) the antimicrobial activity of ethyl acetate and butanol extracts.

MATERIALS AND METHODS

Plant material

Senecio delphinifolius Vahl. was collected in April 2013 (flowering stage) in Grareme -Mila, Algeria. The plant was identified by Pr. Kaabache Mohamed, Setif University1. A voucher specimen was deposited at the department of chemistry University of Mentouri Constantine (N° ZA 117).

Extraction

Air-dried powdered material from the aerial parts (100g) of *Senecio delphinifolius* was extracted with 70% MeOH hydro-alcoholic solution for 24 h, three times. The MeOH extract was evaporated to dryness. The residue was dissolved in boiling water. After filtration, the filtrate was concentrated and re-extracted several times with EtOAc and *n*-BuOH resulting in a residue of 0.52g from EtOAc extract and 0.93g from *n*-BuOH extract.

Determination of total polyphenols

The Determination of polyphenols was performed using a modified Folin-Ciocalteau photometric assay [15]. 20ul of each methanol extract were mixed with 250 µl of Folin-Ciocalteu reagent (1N), 1.25 mL sodium carbonate (2%) and 480 µL distilled water. The solutions were homogenized, capped and protected from light and kept at room temperature for 30mn. The absorbance was measured at 750 nm using water as a blank. The test was performed in triplicate. The standard curve was obtained using standard solutions of Gallic acid at the same concentration of the sample. The content of total polyphenols was expressed as microgram of Gallic acid equivalent per milligram of dried plant.

Determination of flavonoids

Aluminum chloride colorimetric method was used for flavonoids [16]. Each plant extract (1 ml) was separately mixed with 1 mL of methanol solution of aluminum chloride (2%). the solutions were homogenized, capped and protected from light and kept at room temperature. The absorbance was measured at 430 nm using the methanol as a blank. The test was performed in triplicate and the standard curve was obtained using a serial concentration of quercetin solutions. The flavonoids content was expressed as microgram of quercetin equivalent per milligram of dried plant.

Antibacterial activity

The Anti-microbial assay was carried out on two extracts using agar diffusion method [17], against five human pathogenic bacteria, including (standart strains) *Staphylococcus aureus ATCC252923* and *Pseudomonas aerugenosa ATCC 27853 E.coli ATCC 2592* and *clinical strains Klebsella pneumonia Selmonella sp* and one fungus *Candida albicans.*

The bacterial strains were first grown on Muller Hinton medium (MHI) at 37 °C for 24 h prior to seeding on to the nutrient agar and the fungal strains at 30 °C for 48 h.

The isolated compounds were mounted on sterile filter paper discs (6 mm in diameter) with the following concentrations in mg/mL: 8, 4, 2, 1 and 0.5. The discs were placed on the inoculated agar media. The treated Petri discs were kept at 4 ◦C for 1 h, and incubated at 37 °C for 24 h. The antibacterial activity was assessed by measuring the zone of growth inhibition surrounding the discs. Each experiment was carried out in triplicate.

DPPH radical-scavenging activity

The capacity of two extracts from *Senecio delphinifolius* to reduce the radical 2,2-diphenyl-1- picrylhydrazyl (DPPH) was assessed using the method of Masuda *et al* [18]. 15 µL of the extract at different concentrations was added to 1.5 mL of a DPPH ethanolic solution. The mixture was shaken vigorously and left standing at room temperature for 30 min in the dark. The absorbance of the resulting solution was then measured at 517 nm. The normal purple color of DPPH will turn into yellow when its singlet electron is paired with a hydrogen atom coming from a potential antioxidant. The scavenging activity of two extracts was evaluated according to the formula:

DPPH scavenging effect (%) = [$(A_0 - A_1)/A_0$]×100, where A_0 is the absorbance of the control at 30 min, and A_1 is the absorbance of the sample at 30 min. All samples were analyzed in three replications.

RESULTS AND DISCUSSION

Determination of total polyphenols and flavonoids

The total phenolic contents of the ethyl acetate and butanol extracts determined by Folin-Ciocalteu method are reported as mass of equivalent of Gallic acid per plant material whereas the flavonoids content is exposed as

quercetin equivalents. The obtained total phenolic contents were of 7.22 ± 0.84 and 7.86 ±0.25mg EAG/mg respectively and the flavonoids contents were 22.96± 1.41 and 10.25±8.94 mgE Q/mg respectively. These results are significantly lower than what was reported by Sevil *et al* [19] who found that The total phenolic contents of *S. pandurifolius, S. trapezuntinus, S. integrifolius* subsp. *aucheri, S. hypochionaeus* var. *argaeus, S. hypochionaeus* var. *ilkasiensis* and *S. lorentii* in methanol extracts are 81.78 ± 1.5, 41.04 ± 1.0, 37.56 ± 0.7, 19.54 ± 0.4, 39.93 ± 0.3 and 22.15 ± 0.1 mg GAE/g dry extract, respectively. In addition, it was found that *Senecio biafare* contains 0.044 % of polyphenols [20] and *Senecio scandens* contains 11.52 mg GAE/g [21].

The flavonoids content is higher than what was reported by Tundis *et al*. [22] for *Senecio stabianus* (11.8 mg GA/g and lower than what was found by Aparna *et al*. [23] in *Senecio tenufolius* (75.42 ug GA /g).

Table 1: polyphenols and Flavonoids contents from *Senecio delphinifolius* Vahl

Species	Extract	polyphenols contents	Flavonoids contents
Senecio delphinifoliusVahl.	AcOEt	7.22 ± 0.84	22.96± 1.41
	n-BuOH	7.86 ±0.25	10.252±8.94

Antimicrobial activity:

The results were summarized in Table 2 which showed that the two extracts of *Senecio delphinifolius* prevented the growth of all the tested microorganisms with an inhibition zone medium diameter increasing proportionally with the concentrations of the tested samples. The obtained inhibition on bacteria strains varied from 6.33 in *Candida albicans* to 12.67 mm and 12.33 with a highest inhibition zone recorded with *Staphylococcus aureus ATCC252923* at 8 mg/mL (butanolic extract and ethyl acetate extract successively). However no activity against *microorganisms* at low concentration (0.5mg/mL and 1mg/mL have been recorded with the ethyl acetate extract except for *Staphylococcus aureus ATCC252923.* also no activity was recorded against *Salmonella sp and Candida albicans* at 0.5, 1, 2 mg/ mL.

S. aureus is the most sensitive microorganism to all extracts examined in this study. All the Gram+ bacteria are sensitive to *Senecio* extracts than all of the tested Gram- bacteria with some exceptions.

The results of the present study support the observations of the antimicrobial properties of some *Senecio* species, which had a similar effect against tested microorganisms. *K. pneumoniae* is the most sensitive to all extracts considered in this study [19]. In addition Yang *et al.* [21] found that *Senecio scandens*a had a large inhibition against *P. aergunosa ATCC6538P* , *S.aureus ATCC6538P* and *E. coli ATCC25922* .The same results was founded by Manjunath *et al*. [24] by testing the anti-staphylococcal effect of ethyl acetate extract from the leaves of *Senecio tenuifolius* .

Aparna *et al*. [23] found that the alcoholic extract of the aerial part of *Senecio tenuifolius* was inactif against *E.coli* MTCC739, *S. aureus* MTCC739, *K. pneumonia* MTCC3384 at a concentration of 150mg/mL

Table 2: antimicrobial activity of ethyl acetate and butanol extract of *S .delphinifolius* Vahl

	8mg/ml		4mg/ml		2mg/ml		1mg/ml		0.5mg/ml	
Microorganis strains	Ethyl acetate	Butanol	Ethyl acetate	butanol	Ethyl acetate	butanol	Ethyl acetate	Butanol	Ethyl acetate	Butanol
E.coli ATCC 2592	10.67+2.31	11±1.73	8.33+0.58	9.67±2.08	-	09±1.53	-	08±0.00	-	7.33±0.58
Pseudomonas aerogenosaATCC27853	12+1.00	10.33±0.58	9.33+0.58	9.33±1.53	7.67+0.58	09±0.58	-	8.67±1.15	-	-
Klebseilla pneumonia	8.33+0.58	8.67±1.15	-	7.66+0.58	-	07±0.00	-	-	-	-
Staphylococcus aureus ATCC252923	12.33+0.58	12.67±2.08	9.67+1.53	12±2.00	9+1.73	11.33±1.00	8.33+1.53	09±1.73	7.68+1.53	7±0.00
Selmonella sp	-	9±1.00	-	-	-	-	-	-	-	-
Candida albicans	7.33+0.58	07±0.00	7+1.00	6.33±0.58	-	-	-	-	-	-

DPPH radical-scavenging activity

Free radical scavenging was performed using DPPH essay. the ethyl acetate and butanolic extracts were tested at concentrations of 10^{-1}M,10^{-2}M,10^{-3}M,10^{-4}M. Vitamin C was used as standard at the final concentration of 1 mg/ml. at the highest concentration ethyl acetate and butanol exhibit 80.48 % and 73.91 % free radical scavenging respectively. However, according To our finding, previous studies on the antioxidant activities of *Senecio* species indicated that *they* have strong antioxidant effects. Also, it was showed that the methanol extract of *S. gibbosus* subsp. *gibbosus* had significant antioxidant effect with the DPPH assay [25]. Sevil *et al*. [19] determined that The radical scavenging activities of six *Senecio* extracts: S. *pandurifolius, S. trapezuntinus, S. integrifoliussubsp. aucheri, S. hypochionaeusvar. argaeus, S. hypochionaeusvar. ilkasiensisand S. lorentii* extracts were very effective. Our findings for *Senecio delphinifolius* extracts are similar to these reports.

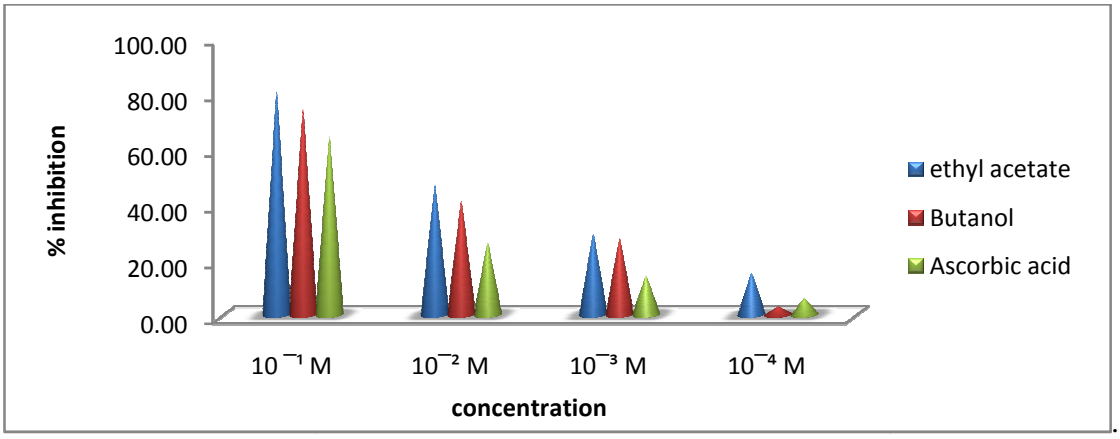

Fig1: Radical scavenging activity of ethyl acetate and butanolic extracts of *Senecio delphinipholius*

REFERENCES

[1]D Stead, *Potato Res*,**1999**,42, 505-509.
[1]S Titdjani, NP Okusa, A Zellagui, LMY Banuls, C Stévigny, P Duez and S Rhouati, *Natural product communications*, **2013** 8(4), 439-440.
[2]P Quezel , S Santa , Nouvelle flore de l'Algérie et des régions désertiques méridionales. Vols II. Ed CNRS, Paris, France; **1963**.
[3]A Zellagui, S Titdjani, N Gherraf and S Rhouati, *Der Pharma Chemica*, **2012**, 4(5), 208-84.
[4]F Bohlmann, C Zdero, J Jakupovic, LN Misra, S Banerjee, P Singh, RN Baruah, MA Metwally, G Schmeda-Hirschmann, PDV Leszek, RM King and H Robinson, *Phytochemistry*, **1985**, 24, 1249-1261.
[5]S Dupre, M Grenz, J Jakupovic, F Bohlmann and HM Niemeyer, *Phytochemistry*, **1991**, 30, 1211-1220.
[6]C Dong-Liang, C Xiao-Ping, C Jie-Kai and E Roeder, *Phytochemistry*, **1992**, 32,151-153.
[7]P Torres, J Ayala, C Grande, MJ Macıas and M Grande. *Phytochemistry*, **1998**, 47, 57-61.
[8]P Torres, CAJ Grande, J Anaya and M Grande, *Phytochemistry*, **1999**, 52, 1507-1513.
[9]EM Suleimenov, R A Jose, SB Rakhmadieva,W Borggraeve and WL Dehaen. *Chemistry of natural compounds*, **2009**, 45(5), 547-972.
[10]HE Zhong-mei, B Bing, H Wang , S Jia-ming, Z Ying, *Chinese Traditional Patent Medicine*, **2010**, 12, 1001-1528.
[11]T Dao-peng, C Gui-xin and W Zheng-tao, *Biochemical Systematic and Ecology*, **2010**, 38(1), 122-124.
[12]Y Yang, L Zhoa, W Yu-Fang, C Man-Li, C H Huo, YC Gu, Qw Shi and H Kiyota, *Chemistry & Biodiversity*, **2011**, 8(1), 3-72.
[13]F Bohlman, CZ dero, J Jakupovic, M Grenz, V Castro, RM King, H Robinson and LPD Vincent, *Phytochemistry*, **1986**, 25, 1151 - 1159.
[14]K R Markham, OM Andersen, flavonoid chemistry biochemistry and application, CRC Press is an imprint of Taylor , the United States of America on acid **2006**.
[15]M Muchuweti, AR Ndhlala and A Kasiamhuru, *Food Chemistry*, **2006**, 94, 415-419.
[16]A Djeridane, M Yousfi, B Nadjemi, D Boutassouna, P Stoker and N Vidal, Food chemistry, **2006**, 79, 654-660.
[17]National Committee for Clinical Laboratory Standards [NCCLS] M7-A6, 6th ed., Wayne.**2003**
[18]T Masuda, S Yonemori, Y Oyama, Y Takeda and T Tanaka. *Food Chem*, **1999**, 47(4), 1749-1754.
[19]A sevil, A Akzoy, H Ergin, E Lutfiye and B Umit, *Acta Bot. Gallica*, **2008**, 155(3), 447-56.
[20]BO Ajiboye, EO Ibukun, G Edobor , AO Ojo and SA Onikanni. *J. Inv. Pharm. Sci*, **2013**, 1(5), 428-432.
[21]CH Yang, HW Chang, HY Lin and LY Chuang. *Journal of Pharmacognosy and Phytochemistry*, **2013**, 2(1), 294-305.
[22]R Tundis, F Menichini, MR Loizzoa, M Bonesi , U Solimene and F Menichini. *Natural Product Research*, **2012**, 26(5), 393-404.
[23]S Aparna , KNV Rao, A Begam and D Banji. *IJPBS*, **2013**, 3(2), 17-23.
[24]M Manjunath, G Lavanya, R Sivajyothi and R vijayasarathi, *Asian Pacific Journal of Tropical Biomedicine*, 2013, 3(3), 191 195.
[25]F Conforti, M Marrelli, G Statti and F Meninchini, *Nat. Prod. Res*, **2006**, 20(9), 805-812.