Int J Pharma Res Health Sci. 2017; 5 (2): 1672-1678
DOI:10.21276/ijprhs.2017.02.14

CODEN (USA)-IJPRUR, e-ISSN: 2348-6465

PHS Scientific House
PHARMAHEALTHSCIENCES

International Journal of Pharma Research and Health Sciences

Available online at www.pharmahealthsciences.net

Original Article

Chemical Composition, Antimicrobial Activity of Essential oil and Chromosome Number of *Senecio jacobaea L.* from Algeria

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ARTICLE INFO

Received: 01 Apr 2017
Accepted: 22 Apr 2017

ABSTRACT

The objective of this work is to identify the chemical composition of the essential oil of *Senecio jacobaea* from Algeria, to test its antimicrobial activities and to identify its chromosomal number. The analysis and identification of the components of the essential oil of this species was performed using the (GC/SM), the antimicrobial activity is tested on five bacterial strains and yeast and the crushing process is used in the karyotype analysis. The samples of *S. jacobaea* were harvested from eastern Algeria: Souk Lethnine (Bejaia) in full bloom stage. The air dried materials were subjected to a hydrodistillation which gave a viscous liquid with pale yellow oil. The analysis by GC-GC/MS allowed the identification of 44 compounds corresponding to 83.56% of the total oil. This oil is characterized by the presence of major components, pentanol-3-methyl (25.70%), followed by cyclopentanedione-3-methyl-1,2 (22.83%) and phytol (3.15%). The hydrocarbons and alcohols represent 56% of the oil of *S. jacobaea*. The essential oil, tested on 5 bacterial strains and yeast, showed a relatively moderate antimicrobial activity against all the bacteria tested. The population of *S. jacobaea* shows a tertraploïde chromosome number, 2n = 4x = 40, with a basic chromosome number x = 10.

Key words: *Senecio jacobaea*, Essential oil, Antimicrobial activity, chromosome number, Algeria

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1. INTRODUCTION

Senecio is one of the most important genera of the *Asteraceae* family. It contains more than 1500 species distributed throughout the world[1]. This genre is cosmopolitan[2]. In Algeria, it is represented by 18 species, 5 of which are endemic[3].

Many species of the genus *Senecio* have been used in traditional medicine. In Chile, Andean populations use *Senecio* species as traditional remedies for altitude sickness[4], and for the treatment of asthma, cough, bronchitis, eczema and wound healing[5-6]. Traditional uses of *S. vulgaris*, in infusion, to calm painful menstruation and *S. cineraria* to relieve ophthalmic problems, have been reported[7]. Some species of the genus *Senecio* have anti-viral activity for hepatitis B[8]. The essential oil of *S. flammeus* is effective in the treatment of acute and chronic inflammations[9].

The essential oils of the genus *Senecio* have been the subject of a large number of phytochemical studies, which made it possible to highlight a significant chemical variability. The α-pinene is the major component of essential oil in *S. angulatus* of France[10], in *S. perralderianus* of Algeria[11] and *S. flammeus* of China[9]. The β-caryophyllene is present in the oil of *S. othonnae* and *S. racemosus* from Turkey[5], of *S. nudicaulis* from India[12] and *S. vulgaris* from France[13]. Several species of *Senecio* possess cytotoxic activities on cancer cell lines[14-16].

The essential oils of the genus *Senecio* have several biological activities; *S. graveolens* inhibits the growth of *Micrococcus luteus*, *Staphylococcus aureus*, and also have antifungal effects on *Candida albicans*[17]. The oils of *S. othonnae* and *S. nemorensis* have antimicrobial activity against *Bacillus cereus*, *Staphylococcus aureus*, *Enterococcus faecalis* and *Candida tropicalis*; those of *S. racemosus* possess antifungal activity against *Candida tropicalis*[5]. The essential oil of *S. aegyptius* and S. *pandurifolius* possesses antifungal activity against *Candida albicans*, and against Gram-positive bacteria[6,18]. The oil of *S. amplexicaulis* exhibits significant antifungal activity against five phytopathogenic fungi[19].

The basic chromosomal number for the genus *Senecio* is traditionally (x = 10), however, Lopez et al.[20], in a study on the number of *Senecio* chromosomes, supported (x = 5) as base number for this genre. Thus, the number 2n = 40 in *S. jacobaea* can be considered as octaploid and 2n = 80 as 16-ploides. The diploids with (2n = 10) were observed in Africa, the most probable centre of origin of *Senecio*[21-22]. According to Lopez *et al.*[20], the secondary polyploidy proposed by Stebbins (1971) is the best explanation for the *Senecio* base number dilemma.

A considerable variation in the number of chromosomes is reported for *Senecio jacobaea* from various regions (2n = 20, 32, 40 and 80)[23]. *S. jacobaea* is one of the species of the genus, most variable at the caryological levels, with several cytotypes[24]. Four different cytotypes have been reported, with the occurrence of tetraploids (2n = 40); In Slovakia, polyploids were identified, one tetraploid and one octaploid at (2n = 80)[23]. The number of diploid chromosomes (2n = 20) was reported in Bulgaria[25], while in Ireland the chromosome number of 2n = 32 was reported[26].

The aim of this work is to identify the chemical composition of the essential oil of *Senecio jacobaea* of Algeria, thus to test its antimicrobial activities and to identify its chromosomal number.

2. MATERIALS AND METHODS

Plant Material

Senecio jacobaea is a glabrous or pubescent plant with a height of 0.50-1.20 m, with striated stems not exceeding 5-7 mm in diameter. The leaves of a dark green are petiolate and deeply erected, the superior are sessile and irregularly laciniated. The inflorescence is composed of yellow, mediocre heads and grouped in corymbs at the top of the stems[3] (Figure 1). It is a plant originating in Europe, Asia and Siberia. In Algeria, it is common in the Tell marshes and known as Debbouz El-Arab[3].

Samples of *S. jacobaea* were collected in the flowering stage in June 2014, in eastern Algeria (Souk Lethnine, Bejaia) (figure 2). The air dried materials were subjected to hydrodistillation for 3h using a Clevenger apparatus type. Voucher specimens were deposited in the herbarium of the Department of Ecology and Biology, Setif 1 University, Algeria. The oil obtained was collected and dried over anhydrous sodium sulphate and stored at 4°C in sealed brown vials until use.

Essential oil analysis

The essential oils were analyzed on a Hewlett-Packard gas chromatograph Model 5890, coupled to a Hewlett-Packard model 5971, equipped with a DB5 MS column (30 m X 0.25 mm; 0.25 μm), programming from 50°C (5 min) to 300°C at 5°C/min, with a 5 min hold. Helium was used as the carrier gas (1.0 mL/min); injection in split mode (1:30); injector and detector temperatures, 250 and 280°C, respectively. The mass spectrometer worked in EI mode at 70 eV; electron multiplier, 2500 V; ion source temperature, 180°C; MS data were acquired in the scan mode in the m/z range 33-450. The identification of the components was based on comparison of their mass spectra with those of NIST mass spectral library[27-28] and those described by Adams, as well as on comparison of their retention indices either with those of authentic compounds or with literature values[30].

Antimicrobial activity

The antimicrobial activities of the essential oil of *S. jacobaea* were evaluated against One Gram positive bacteria (*Staphylococcus aureus* ATCC2592), four Gram negative bacteria (*Pseudomonas aeruginosa* ATCC 27853, *klebsiella pneumoniae ATCC 70060*, *Escherichia coli* ATCC 25922 and *Shigella sp*) and the yeast (*Candida albicans* ATCC 10231). The bacterial inoculums was prepared from overnight broth culture in physiological saline (0.8 % of NaCl) in order to obtain an optical density ranging from 0.08-01 at 625 nm. Muller-Hinton agar (MH agar) and MH agar supplemented with 5 % sheep blood for fastidious bacteria were poured in Petri dishes, solidified and surface dried before inoculation. Sterile discs (6 mm Φ) were placed on inoculated agars, by test bacteria, filled with 10 μl of mother solution and diluted essential oil (1:1, 1:2, 1:4, and

1:8 v/v of DMSO). DMSO was used as negative control. Bacterial growth inhibition was determined as the diameter of the inhibition zones around the discs. All tests were performed in triplicate. Then, Petri dishes were incubated at 37°C during 18 to 24h aerobically (Bacteria). After incubation, inhibition zone diameters were measured and documented. The bactericidal and bacteriostatic test on the five bacterial strains is performed using pure oil of *S. jacobaea*.

Karyology

The crushing process is used in the karyotype analysis. The meristems of the roots, resulting from the germination of seeds, are used for chromosomal preparations. A pretreatment at room temperature for 1 hour 15 minutes was applied before fixation of the root-tips, in a solution of colchicine a 0.05%. After fixation in a mixture of ethanol and acetic acid

3. RESULTS

The hydro-distillation of *Senecio jacobaea* essential oil gave a viscous liquid with yellow oil. The average yield of essential oil of the sample is 0.02%. The analysis and identification of the components of the essential oil of this species was performed using the (GC/SM) (Figure 3).

The chemical analysis of the essential oil of *S. jacobaea* allowed the identification of 44 chemical components corresponding to 83.56% of the total oil. The compound identified in these oils and their relative abundances are presented in order of their appearance (Table 1). The chemical composition of essential oil of this species is dominated by the presence of major compounds, Pentanol-3-methyl (25.70%) and Cyclopentanedione 3-methyl, 1,2 (22.83%), Followed by α-pinene (3.52%) and Phytol (3.15%). The oil of *S. jacobaea* is dominated by the chemical family of hydrocarbons with a percentage of 28.10%, followed by alcohols with 27.94% and monoterpens with 6.92%.

The antimicrobial activity of the essential oil of *S. jacobaea* is tested on five bacterial strains and yeast with three antibiotic controls (gentamicin, cefotaxime and colistinsulfate) according to the disk diffusion method. The essential oil of *S. jacobaea* has shown a bacteriostatic effect and generates diameters of inhibition zones on the microbial strains varying from 7 to 15 mm with the exception of the yeast *Candida albicans* which has resistance to oil of this species (Table 2). The inhibition diameters generated by the essential oil of *S. jacobaea* are considered medium to large in comparison with the diameters generated by the antibiotics

The most important inhibition diameters are observed on the strain *Klebsiella pneumonea* ATCC700603 and *Staphylococcus aureus* ATCC 25923 with a halo of 15 and 14 mm respectively, these two bacterial strains are therefore the most sensitive to this essential oil. The oil is moderately active against *Echerichia coli* ATCC 25922, *Pseudomona aeruginosa* ATCC 27853 and *Shigella* sp. with an inhibition diameter of 11-13 mm. The other dilutions of the essential oil of *S. jacobaea* show moderate to low activity on the bacterial strains studied with diameters of inhibition varied between 7 and 12.5 mm.

The observation of metaphase plates of *S. jacobaea* allowed us to identify a karyotype with a tetraploïde chromosome number 2n = 4x = 40 (Figure 4).

4. DISCUSSION

The yields of plant essential oils are quite low, often less than 1%, and the high levels as in the bud of clove (1.5%) are exceptional[30]. The yield of *S. jacobaea* essential oil of 0.02% is the same in *S. giganteus* of Algeria[31]. This rate is very low compared to that found in other species of the genus *Senecio*, in particular in *S. graveolens* (0.5%)[17], *S. aegyptius* (0.4%)[6], *S. polyanthemoides* (0.23%)[32], *S. perralderianus* (0.1%)[11]. This difference in yield of essential oil can be attributed to several factors, including origin, species, harvest period, drying time and extraction technique of essential oils[33-38].

The chemical profile of the essential oil of *S. jaobaea*, from Bejaia region (Algeria), differs from those reported by Dooren *et al.*[39], whose shows that germacrene-D and undecene-1 are the major components, while these two compounds are completely absent from the oil of the studied population. In general, the chemical composition of the essential oil of *S. jacobaea* is integrated into the overall context of the composition of the genus *Senecio* by the presence of the major compounds, α-pinene, which is present in *S. angulatus* of France, *S. perralderianus* and *S. giganteus* of Algeria and *S. flammeus* of China[9-11,40], while the phytol is present in *S. vulgaris* of France[10].

S. jacobaea is characterized by the prevalence of hydrocarbon components that bring our species closer to the *S. coincyi* species of Spain[41] and *S. giganteus* of Algeria[40].The chemical composition of *S. rowleyanus* species from Egypt[42], *S. royelanus* and *S. belgaumensis* from India[43-44] and *S. vulgaris* and *S. angulatus* from Corsica (France)[10] differs considerably from that of *S. jacobaea*.

The antibacterial and antifungal activities of an essential oil are linked to their chemical composition, to the functional groups of the major compounds (alcohols, phenols, terpene compounds and ketones) and to their synergistic effects[45].

The results of the bacteriological tests of *S. jacobaea* are generally similar to those of the literature. The oils of *S. othonnae* and *S. nemorensis* show activity against *Staphylococcus aureus*[5], as well as the oil of *S. graveolens*[17]. Our results are similar to those of *S. pogonias* and *S. oreophyton* essential oils, which exhibit antibacterial activity against *E. coli* and *K. pneumoniae*[46]. Whereas, *S. aureus*, *E. coli*, *K. pneumoniae* and *P. aeruginosa* are resistant to *S. glaucus* oil from Egypt[47].

The yeast *Candida albicans* is resistant to the essential oil of *S. jacobaea*, the same result is observed by Kenoufi et al.[31]

with the essential oil of *S. giganteus* of Algeria. On the other hand it's sensitive to the essential oil of *S. pedunculatus*[48]. While the oils of *S. glaucus* and *S. aegyptius* show moderate activity against *C. Albicans* [6,47].

A considerable variation in the number of chromosomes is reported for *S. jacobaea* from different regions[23], but the chromosomal number 2n = 40 is the most replied in nature. The caryological study of *S. jacobaea* of Algeria shows a cytotype with a chromosomal number diploid 2n = 4x = 40. The same results were observed in this species[21-23]. *S. jacobaea* is one of the species of the genus, most variable at the caryological levels, with several cytotypes[24]. In Slovakia, a tetraploid and octaploid were identified[23]. The chromosome number (2n = 20) was reported in Bulgaria[25], while in Ireland the chromosome number of 2n = 32 was observed [26].

Table 1: Chemical composition of *Senecio jacobaea* essential oil

Yield v/v	KI	0.02	Yield v/v	KI	0.02
Number of compounds		**44**	**Number of compounds**		**44**
Total		**83.59**	**Total**		**83.59**
Pentanol -3 methyl	833	25.70	β-ionone (E)	1483	0.27
α-pinene	932	3.52	Muurola-4(14),5-diene trans	1493	0.19
Pentyl furan (2)	984	1.68	Viridiflorene	1496	0.99
β-mentha-1 (7), 8 dienne	1003	0.19	α-muurolene	1500	0.28
Cyclopentane dione 3 methyl, 1,2	1017	22.83	Δ-amorphene	1511	0.15
Limonene	1024	0.44	Kessane	1529	0.63
β-ocimene (Z)	1032	2.06	α-calacorene	1544	0.59
Nonanal (n)	1100	2.20	Italicene epoxide	1547	0.47
Ocimene (neo-allo)	1140	0.08	Nerolidol (E)	1561	0.60
Nonen-1-al (2Z)	1144	0.31	Caryophyllene oxide	1582	2.97
Decanal (n)	1201	0.26	Salvial 4-(14) en 1-one	1594	0.11
β-cyclocitral	1217	0.18	Humulene epoxide II	1608	0.59
Decadienal (2E, 4E)	1315	0.34	Citronellyl pentanoate	1624	1.95
β-damascenone (E)	1383	0.95	Pentadecanone (2)	1697	1.18
α-funebrene	1402	1.70	Farnesyl acetone (5E, 9Z)	1886	0.242
Sesquithuijene	1405	0.13	Phytol	1942	3.15
Caryophyllene (E)	1417	0.62	eicosene	1987	2.13
Guaiadiene (6, 9)	1442	0.99	Tricosane	2300	0.17
Geranyl acetone	1453	0.55	Tetracosane	2400	0.10
β-farnesene (E)	1454	0.43	Pentacosane	2500	0.56
Linalool isovalerate	1466	0.29	Hexacosane	2600	0.32
γ-muurolene	1478	0.24	Heptacosane	2700	0.26

Monoterpene hydrocarbons	6.9
Oxygenated monoterpenes	1.5
Sesquiterpene hydrocarbons	6.3
Oxygenated sesquiterpene	5
Diterpenes	3.2
Hydrocarbons	28.1
Alcool	27.9
Aldehyde	3
Others	1.7

Table 2: Inhibition diameter (mm) of *S. jacobaea* essential oil

Microorganisms	Antibiotics*			Dilution			
	CN	CS	CTX	1	1/2	1/5	1/10
Staphylococcus aureus ATCC 25923	0	18	18	14	9	12	12.5
Echerichia coli ATCC 25922	25	15	33.5	13	10	11	10
Klebsiella pneumonea ATCC700603	17	12	19	15	9	9	9.5
Pseudomona aeruginosa ATCC 27853	26	15	18	12	7	9	10.5
Shigella sp	30	15	14	11	10	9	9.5
Candida albicans ATCC 1	0	0	0	0	0	0	0

* CN = gentamicine; CS = colstin sumlfate; CTX = Cefotaxime

Fig 1: *Senecio jacobaea* (Photo Kenoufi 2016)

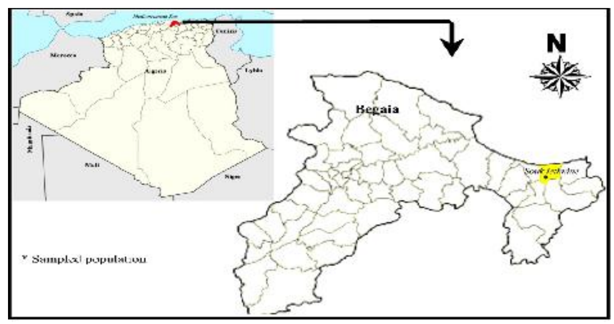

Fig 2: Population of *Senecio jacobaea* sampled

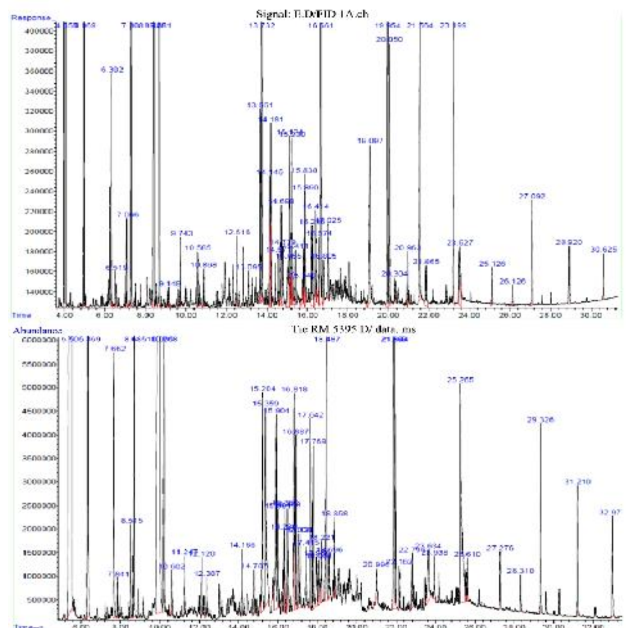

Fig 3: GC/Fid and Masse profiles of *Senecio jacobaea*

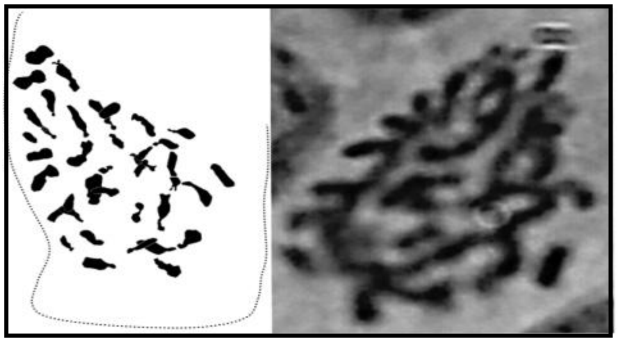

Fig 4: karyotype of *Senecio jacobaea* 2n = 4x = 40 (Magnification = HI 100X)

5. CONCLUSION

Analysis of the chemical composition of essential oils by GC/MS allowed the identification of 44 components in the essential oil of *Senecio jacobaea* collected from Souk lethnine Bejaia region. It is characterized by the main presence of pentanol-3-methyl (25.70%), and cyclopentanedione-3-methyl-1,2 (22.83%). We also note that the chemical composition of the Bejaia sample differs from species of genus *Senecio*. The antibacterial activity of *S. jacobaea* show that the essential oil of this species has moderate inhibitory action on almost all the bacteria tested. The chromosome number of *S jacobaea* is stable and similar to bibliographic results. The caryotype of the Bejaia population is a tetraploid with 2n = 4x = 40, and a chromosomal basic number x = 10.

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Conflict of Interest: None

Source of Funding: This work was supported by MESRS of Algeria (Code Project CNEPRU F01220140022), and by the Laboratory of chemistry and Heterocyclic of Clermont Ferrand, France.