Available online at www.scholarsresearchlibrary.com

Scholars Research Library

Der Pharmacia Lettre, 2013, 5 (5):252-255
(http://scholarsresearchlibrary.com/archive.html)

Der Pharmacia Lettre
Scholars Research Library
ISSN 0975-5071
USA CODEN: DPLEB4

Composition and antibacterial activity of the essential oil of *Ruta chalepensis* subsp. *angustifolia* from Algeria

Chibani S[1], Bouratoua A[1], Kabouche A[1], Laggoune S[1], Semra Z[1, 2], Smati F[2] and Kabouche Z[1*]

[1]University of Constantine, Department of Chemistry, Constantine, Algeria
[2]CHU Benbadis-Constantine, Bacteriology service, Constantine, Algeria

---

ABSTRACT

The essential oil obtained by hydrodistillation of aerial parts of Ruta chalepensis subsp. angustifolia (Rutaceae) was analyzed by GC and GC/MS. 16 compounds were characterized representing 99.99% of the essential oil with 2-acetoxytetradecane (58.44%), 2-acetoxytridecane (19.07%) and 2-tridecanone (6.39%) as major components. The antibacterial activity of the essential oil was tested against 5 gram-positive and 4 gram-negative bacteria by the use of the disc diffusion method.

Keywords: *Ruta chalepensis* subsp. *angustifolia*, Rutaceae, 2-Acetoxytetradecane, 2- Acetoxytridecane, 2-Tridecanone, Antibacterial activity.

INTRODUCTION

The genus *Ruta* (Apiaceae) is very rich in coumarins[1-3]. In some countries of the Mediterranean area, besides its traditional use as abortive, *Ruta* is known for being used against helminths and to treat children fevers. *R. montana* is used in digestive disorders and helminthiasis [4] as well as *R. graveolens,* known for its antiparasitic, stomachic, digestive, vermifuge, emmenagogue and molluscicidal activities [2,5].

There are four *Ruta* species and subspecies in Algeria, *R. montana*, *R. chalepensis* subsp. *angustifolia*, *R. chalepensis* subsp. *latifolia* and the Saharian species *R. tubercula* [6, 7]. In continuation of our works on aromatic plants of Constantine area[3, 8-28], we describe here, the GC and GC/MS analysis of the essential oil of *Ruta chalepensis* subsp. *angustifolia*, in Arabic *"fidjla"*, which is traditionally known for its abortive and anti-fever effects. Reported essential oils of *R. chalepensis* have been found to be rich with undecan-2-one [24,29-43].

MATERIALS AND METHODS

Plant Material

Fresh aerial parts of *R. chalepensis* subsp. *angustifolia* were collected in May 2012 from Grarem (Eastern Algerian). A Voucher specimen was deposited at the herbarium of the Laboratory of Therapeutic Substances, University Constantine 1, Algeria (LOST ZK Rc05/12).

Extraction of the essential oil

The hydrodistillation of the fresh aerial parts (100 g) of *R. chalepensis* subsp. *angustifolia*, for 3 h in a Clevenger-type apparatus, yielded 0.80 % (w/w) of a yellow essential oil which was stored until tested and analyzed.

Gas chromatography

GC analysis was performed on a Shimadzu GC17A gas chromatograph equipped with a split/splitless injector (250°C) and a flame ionization detector (250°C). Retention times for comparison with authentic compounds were

measured using a cross-linked DB5-MS column (40 m × 0.18 mm, film thickness 0.18 µm). The oven temperature was programmed as isothermal at 60°C for 5 min, then raised to 275°C at 5°C/min and held at this temperature for 5 min. Helium was used as the carrier gas at a rate of 1 ml/min. Relative percentage amounts were calculated from peak area without the use of correction factors.

Gas Chromatography-Mass spectrometry

Gas chromatography-mass spectrometry: GC-MS was performed using a Shimadzu QP5050 mass selective detector using a cross-linked DB5-MS column (40 m × 0.18mm, film thickness 0.18 µm). The oven temperature was programmed as isothermal at 60°C for 5 min, then raised to 275°C at 5°C/min and held at this temperature for 5 min. Helium was used as the carrier gas at a rate of 1 ml/min. 0.1 µl oil was introduced directly into the source of the MS via a transfer line (280°C) with a split ratio of 1:50 and a linear velocity of 30.0 cm/sec. Ionization was obtained by electron impact (70 eV, source temperature 200°C, resolution 1000).

Identification of components

Essential oil components were identified based on their retention indices (determined with reference to a homologous series of normal alkanes), and by comparison of their mass spectral fragmentation patterns with those reported in the literature [44,45] and with authentic compounds for major components.

Antibacterial activity

The antibacterial activity of the essential oil was tested against a range of microorganisms, namely *Escherichia coli* ATCC 25922, *Escherichia coli* (HS), *Staphylococcus aureus* ATCC 2913, *Staphylococcus aureus* (HS), *Pseudomanas aeruginosa* ATCC 27853, *Pseudomonas aeruginosa* (HS), *Klebsiella pneumoniae* ATCC, *Streptococcus enterococcus* (HS), *Klebsiella pneumoniae* (HS). The reference strains were obtained from the Pasteur Institute (Algiers). The other strains (HS) were obtained from the laboratory of bacteriology, Benbadis Hospital, Constantine, using conventional methods (clinical isolation) [46].

RESULTS AND DISCUSSION

Chemical composition of the essential oil

The Hydrodistillation of the fresh aerial parts of *Ruta chalepensis* subsp. *angustifolia* collected from Grarem (Constantine area) furnished 0.8% (w/w) of a yellowish essential oil, having an intense and penetrating odor.

16 compounds were identified by GC and GC/MS, representing 99.99% of the total essential oil, characterized by the main presence of 2-acetoxytetradecane (58.44%), 2-acetoxytridecane (19.07%) and 2-tridecanone (6.39%) as major components (Table 1).

The composition of the present essential oil is different from those of the species collected at Boudouaou-City (Central Algerian) and Jijel (North Eastern Algerian). The hydrodistilled essential oil of the plant collected in May 2005 from Boudouaou-City was characterized by the main presence of 2-undecanone (28.20%), 2-nonanol (20.00%), 2-methyloctylacetate (12.70%) and 2-methyldecylacetate (05.80%) [30] while the hydrodistilled essential oil of the same species collected in May 2009 from Jijel (North Eastern Algeria) was predominated by undecan-2-one (83.4 %).

Table 1: Chemical composition, Retention indices and percentage composition of the essential oil of *Ruta chalepensis* subsp. *angustifolia* collected from Grarem (Algeria)

Pic	Compound[a]	RI[b]	%
1	limonene	1028	1.10
2	nonanal	1098	0.45
3	2-decanone	1190	3.44
4	2-nonanol	1196	0.92
5	acetic acid, octyl ester	1211	0.35
6	2-octanol, acetate	1214	0.50
7	**2-acetoxytridecane**	1223	**19.07**
8	carvone	1243	0.59
9	2-undecanone	1292	2.02
10	acetic acid, nonyl ester	1310	0.45
11	5,6-diethenyl-1-methylcyclohexene	1325	0.46
12	2-tridecanol	1381	0.55
13	2-dodecanone	1390	2.00
14	**2-acetoxytetradecane**	1494	**58.44**
15	**2-tridecanone**	1497	**6.39**
16	α-elemol	1551	3.26
		Total	**99.99**

[a] *Compounds listed in order of their RI*

[b] *RI (retention index) measured relative to n-alkanes (C_6-C_{24}) using DB5-MS column*

2-Acetoxytetradecane was mainly found in reported essential oil of *Ruta montana* collected from Constantine (Eastern Algerian) (9.19%) [28] while 2-acetoxytridecane (12.73%) was reported as a major component of the essential oil of *Ruta graveolens* [47]. It's noteworthy that essential oils of *Ruta* genus are generally characterized by the main presence of undecan-2-one [24, 29-43].

Nevertheless, it's the first time that 2-tridecanone is reported as a major component of *Ruta* essential oil.

Antibacterial activity

The essential oil of *R. chalepensis* subsp. *angustifolia* inhibited remarkably the growth of *Klebsiella pneumoniae* ATCC, *Klebsiella pneumoniae* (HS), *Escherichia coli* ATCC, *Escherichia coli* (HS), *Streptococcus enterococcus* (HS) and *Pseudomonas aeruginosa* ATCC strains with 21, 21, 20, 20, 20 and 20 mm (inhibition zone diameters**,** respectively). These results are confirmed by the respective MIC's values (Table 2). This antibacterial activity may be due to the synergic effect of the major components.

Table 2: Antibacterial activity (inhibition zones and MIC's) of the essential oil of *R. chalepensis* subsp. *Angustifolia*

Microrganism	Inhibition zone [a](mm)	MIC (µg/ml)
Escherichia coli ATCC 25922	20	**20**
Escherichia coli (HS) [b]	20	**40**
Pseudomonas aeruginosa ATCC27853	20	**20**
Pseudomonas aeruginosa (HS)	18	**60**
Klebsiella pneumoniae ATCC	21	**20**
Klebsiella pneumoniae (HS)	21	**20**
Staphylococcus aureus ATCC 2913.	17	**60**
Staphylococcus aureus (HS)	14	**80**
Streptococcus enterococcus (HS)	20	**20**

[a] *280 µg/ml*
[b] *Hospital Strain*

CONCLUSION

The essential oil of *Ruta chalepensis* subsp. *angustifolia*, collected from Grarem (Constantine area) is characterized by the main presence of 2-acetoxytetradecane (58.44%), mainly found in almost reported *Ruta* essential oils but 2-tridecanone is reported here for the first time as a major component of *Ruta* oil. This may be explained by the climate and soil differences. Antibacterial assays, showed that the essential oil was more active against, *Klebsiella pneumoniae* ATCC, *Klebsiella pneumoniae* (HS), *Escherichia coli* ATCC and *Escherichia coli* (HS).

Acknowledgments

We are grateful to the ANDRS (Oran) and DGRSDT (MESRS, Algeria) for financial support.

REFERENCES

[1] A.I. Gray, P.G. Waterman, *Phytochemistry*, **1971**. 17, 845-864.
[2] F. Dall'Acqua, A. Capozzi, S. Marciani, G. Caporale, *Z. Naturforsch*, **1972**, 276c, 813-817.
[3] Z. Kabouche, N. Benkiki, C. Bruneau, E. Seguin, *Fitoterapia*, **2003**, 74 (1-2), 194-196.
[4] G. Benitez, M.R. González-Tejero, J. Molero-Mesa, *J. Ethnopharm.*, **2010**, 129, 87-105.
[5] B.E. Ellis, S.A. Brown, *Can. J. Biochem.*, **1974,** 52, 734-738.
[6] P. Quezel, S. Santa, **1963**. Nouvelle Flore de l'Algérie et des Régions Désertiques Méridionales. C.N.R.S., Paris, France.
[7] P. Ozenda, **1958.** Flore du Sahara Septentrional et Central. C.N.R.S., Paris.
[8] N. Kambouche, B. Merah, S. Bellahouel, A. Bouayed, A. Dicko, A. Derdour, C. Younos, R. Soulimani, *J. Med. Food*, **2008**, 11, 593-595.
[9] N. Benkiki, Z. Kabouche, F. Tillequin, P. Vérité, E. Seguin, *Z. Naturforsch*, **2003**, 58c, 665-668.
[10] A. Kabouche, Z. Kabouche, E. Seguin, F. Tillequin, C. Bruneau, *Chem. Nat. Comp.*, **2004**, 40 (2), 188-189.
[11] A. Kabouche, Z. Kabouche, C. Bruneau, *Flav. Fragr. J.*, **2005**, 20, 235-236.
[12] A. Kabouche, Z. Kabouche, E. Seguin, F. Tillequin, C. Bruneau, *Biochem. Syst. Ecol.*, **2005**, 33, 813-816.
[13] Z. Kabouche, N. Boutaghane, S. Laggoune, A. Kabouche, Z. Ait-Kaki, K. Benlabed, *I. J. Aromather.*, **2005**, 15, 129-133.
[14] O. Touafek, A. Kabouche, A. Nacer, Z. Kabouche, C. Bruneau, *Flav. & Fragr. J.*, **2005**, 20**,** 669-670.
[15] A. Kabouche, O. Touafek, A. Nacer, Z. Kabouche, C. Bruneau. *J. Essent. Oil. Res.*, **2006**, 18, 175-177.

[16] J.C. Do Rego, N. Benkiki, E. Chosson, Z. Kabouche, E. Seguin, J. Costentin, *Eur. J. Pharmacol*., **2007**, 569, 197–203.
[17] A. Kabouche, Z. Kabouche, Bioactive diterpenoids of *Salvia* species. *Studies in Natural Products Chemistry*, **2008**, 35C, 753-833, Edited by Atta-ur-Rahman, Elsevier.
[18] S. Laggoune, A. Kabouche, Z. Kabouche, M.A. El-Azzouny, *J. Essent. Oil. Res.*, **2009**, 21, 67-68.
[19] A. Kabouche, A. Ghannadi, Z. Kabouche, *Nat. Prod. Comm.*, **2009**, 4(9), 1251-1252.
[20] D. Berrehal, T. Boudiar, H. Lakhal, A. Khalfallah, A. Kabouche, A. Al-Freihat, A. Ghannadi, E. Sajjadi, M. Mehrabani, J. Safaei-Ghomi, Z. Kabouche, *Nat. Prod. Comm.*, **2010**, **5**, 957-960.
[21] H. Daroui-Mokaddem, A. Kabouche, M. Bouacha, B. Soumati, A. El-Azzouny, C. Bruneau, Z. Kabouche, *Nat. Prod. Comm.*, **2010**, 5(10), 1669-1672.
[22] H. Lakhal, A. Kabouche, Z. Kabouche, *Chem. Nat. Comp*., **2011**, 46, 964-965.
[23] S. Laggoune, A. Zeghib, A. Kabouche, Z. Kabouche, F. Leon, I. Brouard, J. Bermejo, C.A. Calliste, J.L. Duroux, *Rec. Nat. Prod*., **2011**, 3, 238-241.
[24] T. Boudiar, I. Labed, J. Safaei-Ghomi, A. Kabouche, Z. Kabouche, *J. Essent. Oil-Bearing Plants*, **2011**, 14(6), 792-795.
[25] S. Laggoune, I. Brouard, F. Leon, CA. Calliste, JL. Duroux, J. Bermejo, Z. Kabouche, A. Kabouche, *Rec. Nat. Prod.*, **2011**, 5(3), 238-241.
[26] A. Benmerache, D. Berrehal, Khalfallah, A. Kabouche, Z. Semra, Z. Kabouche, *Der Pharm. Lett.*, **2012**, 4(6), 1878-1882.
[27] H. Ghorab, A. Kabouche, Z. Semra, A. Ghannadi, E.B. Sejjadi, R. Touzani, Z. Kabouche, *Der Pharm. Lett.*, **2013,** 5(1), 28-32.
[28] L. Djarri, M. Ferhat, G. Merabet, A. Chelghoum, S. Laggoune, Z. Semra, F. Smati, Z. Kabouche, *Der Pharm. Lett.*, **2013**, 5(4), 70-73.
[29] G.D. Bagchi, P.D. Dwivedi, S. Amrita, A.A. Flora Naqvi, *J. Essent. Oil. Res.*, **2003**, 15(4), 263-264.
[30] R. Abdolhossein, K. Morteza, S. Faramarz, Y. Mohammad, M. Shiva, M. Aazam, *J. Essent. Oil. Res.*, **2002**, 14(5), 378-379.
[31] O. Tzakou, M. Couladis, *J. Essent. Oil. Res.*, **2001**, 13(4), 258-259.
[32] K.H.C. Baser, C.T. Ozek, S.H. Beis, *J. Essent. Oil. Res*., **1996**, 8(4), 413-414.
[33] R.P.A. Inigo, M.E.L. De Viana, C.A.N. Catalan, D. I.A. De Iglesias, *Essent. Deriv. Agrum*., **1981**, 51(4), 349-351.
[34] T. Zunian, Y. Yue, Y. Yang, X, Yajuan, *Zhong. Xia. Ying. Yao*., **2011**, 28(9), 834-838.
[35] S. Mojtaba, A. Parviz Aberoomand, S. Mohammad, R. Abdolhossein, *World Appl. Sci. J.*, **2009**, 7(1), 124-126.
[36] V. De Feo, F. De Simone, F. Senatore, *Phytochemistry*, **2002**, 61(5), 573-578.
[37] K.M. Yaacob, Che M. Abdullah, D. Joulain, *J. Essent. Oil. Res.*, **1989**, 1(5), 203-207.
[38] E.A. Aboutabl, A.A. Elazzouny, F.J. Hammerschmidt, *Sci. Pharma*., **1988**, 56(2), 121-124.
[39] C. Bertrand, N. Fabre, C. Moulis, J.-M. Bessiere, *J. Essent. Oil. Res*., **2003,** 15(2), 98-99.
[40] R.1. Estevez Reyes, A. G. Gonzalez, *Fam. Nueva*, **1972**, 37(430), 903-910.
[41] H. Boutoumi, S. Moulay, M. Khodja, *J. Essent. Oils Bearing Plants*, **2009**, 12(6), 714-721.
[42] A. Belkassam, A. Zellagui, N. Gherraf, M. Lahouel, S. Rhouati, *Adv. Nat. & Appl. Sci*., **2011**, 5(3), 264-268.
[43] N. Kambouche, B. Merah, S. Bellahouel, A. Bouayed, A. Dicko, A. Derdour, C. Younos, R. Soulimani, *J. Med. Food*, **2008**, 11, 593-595.
[44] R.P. Adams, Identification of Essential Oil Components by Gas Chromatography/Mass Spectrometry, Allured Publishing Co. Carol Stream, Illinois, **2007**, 4th Ed.
[45] F.W. Mc Lafferty, D.B. Stauffer, The Important Peak Index of the Registry of New York.
[46] NCCLS. Performance standards for antimicrobial disk susceptibilities tests; Villionova, PA, USA: Approch Standard NCCLS: Publication M2-A5, **1993.**
[47] T. Zunian, Y.Yue, Y.Yang, X. Yajuan, Zhongguo Xiandai Yingyong Yaoxue, **2011**, 28(9), 834-838.