Journal of Materials and Environmental Sciences
ISSN : 2028-2508
CODEN : JMESCN

J. Mater. Environ. Sci., 2018, Volume 9, Issue 7, Page 2017-2022

http://www.jmaterenvironsci.com

Biochemical Studies on Some Egyptian Wild Plants

M. E. Ibrahim[1], S. S. Ahmed[1], S. A. El-Sawi[2], H. M. Motawe[2]

Medicinal and Aromatic Plants Research Dept., 2. Pharmacgnosy Dept., National Research Centre, 12622-Dokki, Egypt

Received 09 Oct 2017,
Revised 18 Nov. 2017,
Accepted 18 Nov. 2017

Keywords
- ✓ Medical plants;
- ✓ Egypt;
- ✓ chemical constituents
- ✓ water extracts;
- ✓ alcoholic extracts;
- ✓ antimicrobial.

melsayed49@yahoo.com ; Phone: +201224392974

Abstract

Medical and aromatic plants in Egypt have been a part of the country's natural and cultural diversity for thousands of years. Most of the landscape in Egypt is a desert, with a few scattered oases. Each geographical area has a variety of flora and fauna all adapted to its own habitat. In recent work we focused on the discovery of this area where these plants grow. The plants were investigated in terms of the chemical constituents of the active chemical groups, as well as study the effect of water and alcoholic extracts on microorganisms. This work includes five plants collected from the eastern Egyptian desert areas parallel to the Red Sea coast. These plants are, *Acacia etbaica* (. Fabaceae), *Aerva lanata* (. Amaranthaceae), *Citrullus colocynthis* (.Cucurbitaceae), *Ochradenus baccatus* (. Resedaceae.) and *Olea europaea,* (. Oleaceae). The phytochemical investigation of the alcoholic and water extracts of the aerial parts of the tested plants showed the different profile of their contents. *A. etbaica*, *A. lanata*, *C. coloeynthis*, *O. baccatus* and *O.europaea* were more or less proportional in their chemical constitution. Flavonoids were common in all of them, and alkaloids dominated them except *Acacia etbaica*. Saponins were detected only in Acacia and *Aerva lanata*. Alcohol and water extract were tested against four ˉ (G -) bacteria, four (G +) bacteria, one yeast and one fungi. There was a variation in activities; however the water extract of some plants was the highest active. It will be better to practice an in vivo experiment to assess the results.

1. Introduction

Because of the strategic location of Egypt at the crossroads of five biogeographical regions (The Nile Valley, the Nile Delta, Western Sahara, the Eastern Desert, and the Sinai Peninsula), it is home to a wide range of plants. Medical and aromatic plants in Egypt have been a part of the country's natural and cultural diversity for thousands of years. Most of the landscape in Egypt is a desert, with a few scattered oases. It has long coastal lines on the Mediterranean Sea, Gulf of Suez, Gulf of Aqaba, the Red Sea and Sinai. Each geographical area has a variety of flora and fauna all adapted to its own habitat [1] In desert countries, such as Egypt, communities live far from each other, so many of them are virtually deprived of the right medical infrastructure, contributing to the main role played by traditional healers so far [2,3]. The Eastern Desert receives the scattered rain, but supports various plants that include *acacia*, a large variety of thorny shrubs and aromatic herbs. So, this study focuses on the classification and evaluation of wild plants in the eastern desert areas parallel to the Red Sea coast in Egypt. As well as to increase Egypt's pharmaceutical raw materials. In some cases, resistance has resulted in human pathogens that have developed drugs against antibiotics in general to search for anti-microbial materials, new from new sources, including plants [4]. Investigation of medicinal plants for antimicrobial activity is important to find possible new compounds for therapeutic use. This work includes plants collected from the eastern Egyptian desert areas parallel to the Red Sea coast. These plants are, *Acacia etbaica* (. Fabaceae) normally a tree (2.5–12 m high), *Aerva lanata* (. Amaranthaceae, perennial herb) *Citrullus colocynthis* (.Cucurbitaceae), it resembles a common melon vine, but it carries small hard fruits with bitter pulp., *Ochradenus baccatus* (. Resedaceae.) Perennial, 0.5-2 m high, with woody bases and many fleshy green smooth branches and *Olea europaea, (.* Oleaceae) [12-13]. The wild olive is regarded as a small-fruited subspecies of the commercial olive. Acacia species has medical and economical importance [14] Variants of its members have health benefits to man. Some of them treat wounds, malaria, sore throat and tooth ache. Cum

Arabic used for kidney ailments. Tannins of many acacia species used for tanning. Many member of Amaranthaceae, e.g. *Aerva lanata* used in folk medicine in many countries as antiplasmodial and antitrypanozomals [15]. The main active compounds of olive oil content of olive tree fruits are oleic acid, phenolic and squalene [16] and they are rich in antioxidant contents which are believed to have the main biological value of olive tree. The oil has a very good effect on vascular system, blood pressure and cholesterol. Also some organs of olive tree have antimicrobial potency. *Citrullus* contained saponins, tannin, terpenes, cardiac glycoside, alkaloids and some other common chemicals. They possessed antioxidant, antidiabetic, antimicrobial, anticancer, analgesic and anti-inflammatory characters [17].
The objective of this study is to evaluate and test the wild plants collected from the Egyptian desert in terms of chemical constituents of active chemical groups, as well as the effect of water and alcohol extracts on microorganisms.

2. Materials and methods

2.1. Plant materials

Plant material was collected from wild shrub populations of some plants species growing in sandy soils on Gebel Elba region approximately 1200 km south of Cairo. Identification of the species was achieved by specialists in the plant classification department at the National Research Centre in Cairo, Egypt, and compared with student flora of Egypt [18]. Voucher specimens are kept in the herbarium of NRC, Cairo, Egypt.

2.2. Preparation of the crude extracts

2.2.1. Alcoholic extract

The 80% ethanolic extracts are prepared following the process described [19]; 100 g of the aerial parts of each plant were collected, dried in the oven at 40 °C and reduced to powder. They were separately macerated with the 80% ethanol and allowed to stand for 72 hrs and then filtered. The filtrates were then evaporated under reduced pressure and dried using a rotary evaporator at 50°C. Dried extracts were stored in labeled sterile screw capped bottles.

2.2.2. Water extract

One hundred gm of the dried powder of each plant were macerated in distilled water at room temperature for 24 hrs. The macerates were filtered and evaporated under vacuum till dryness. The residues were dissolved in ethanol and used for measuring the antimicrobial activity of the water extracts.

2. 3. Phytochemical screening

Plants under study were screened for carbohydrates and/ or glycosides; sterols and/ or triterpenes, flavonoids, tannins, saponins, coumarins and alkaloids, applying the standard procedures.

2.3.1. *Flavonoids*

Half g of the alcoholic extracts was defatted with petroleum ether . The defatted residue was dissolved in 20 mL of 80% ethanol and filtered. The filtrate was used for the following tests: 5 mL of the dilute ammonia solution was added to the portion of the aqueous filtrate of each plant extract followed by the addition of concentrated H_2SO_4. The appearance of the yellow coloration indicated the presence of flavonoids [20].

2.3.2. *Sterols, polyterpenes*

Using Liebermann reagent allows identifying these compounds, Blue-green ring between layers indicates the presence of steroids and pink- purple ring indicates the presence of terpenes[21].

2.3.3. *Polyphenols*

To 1 mL of alcoholic extract, 2 mL of distilled water followed by a few drops of 10% aqueous ferric chloride solution were added. Formation of blue or green color indicated the presence of phenols [20].

2.3.4. *Tannins*

Search for catechin tannins was made using Stiasny reagent. Five mL of alcoholic extract were evaporated to dryness. After adding 15 mL of Stiasny reagent to the residue, the mixture was kept in a water bath at 80°C for 30 min. The observation of a precipitate in large flakes characterized catechin tannins.For gallic tannins, the previous solution was filtered. The filtrate was collected and saturated with sodium acetate. The addition of $Fecl_3$ drops causes the appearance of a blue-black coloration, indicating the presence of gallic tannins[22].

2.3.5. *Alkaloids*
Alkaloids were characterized using Bouchardat reagent (iodo-iodized reagent) and Dragendorff 'reagent (iodobismuthate of potassium reagent). Six mL of the alcoholic extract were evaporated to dryness. The residue is taken up in 6 mL alcohol at 60°C. The addition of 2 drops of Dragendorff reagent on the alcoholic solution caused a precipitate or orange color. Adding 2 drops of Bouchardat reagent on the alcoholic solution caused a reddish brown precipitate which indicated a positive reaction[23].

2.3.6. *Saponins*
Ten mL aqueous total extract in a test tube were shaken for 15 s and allowed to stand for 15 min. A height of persistent foam greater than 1 cm indicated the presence of saponins [24].

2.3.7 *Anthraquinons*
Dilute ammonia was added to the chloroform extract of the plant. The presence of anthraquinones drevatiives was changing the color of the ammonical layer to red. (Borntranger's test) [25]

.

2.4. Biological Activity

2.4.1 Extract preparation
The dry residue of the tested plants was dissolved in alcohol to give concentration of 100 µg/mL .

2.4.2. Microorganism strains
The alcoholic and water extracts antimicrobial activities were tested against four gram negative bacterial strains (*Escherichia coli, Proteus vulgaris, Pseudomonas fluroscens and Salmonella typhi*), Four gram positive strains: (*Bacillus subtilis* -NRRL–B543)*, Lactobacillus breveis, Staphilococcus aureus and chromobacter sp*), one fungal strain (*Aspergillus niger*) and one yeast strain (*Candida albicans*). Test organisms used were obtained from Faculty of Agriculture, Cairo University.

2.4.3 Agar diffusion method
Nutrient agar was used for the cultivation of bacteria and yeast, and Czapek-Dox's medium for cultivation of fungal species. In this method, pre-sterilized Whatman no.1 filter paper discs (5 mm in diameter) (Whatman International Ltd., Maidstone, England) were impregnated with 100 µl of the extract (100 µg/mL), allowed to dry (to get rid of the alcohol) and applied on the surface of agar plates freshly seeded with standard inocula of young cultures, 24-hrs-old bacteria and yeast, and 7-days-old fungi. The plates of test organisms were then incubated at 27° C for 24 hrs for bacteria and yeast and for 48 hrs for fungi. At the end of the incubation period, the inhibition zones were measured (results are the average of triplicate measurements) [26].

3. Results and discussion
The most of selected wild plants were collected from different areas during the growing seasons 2015- 2016, according to the place and date listed in the Table 1., *Acacia etbaica* (Fabaceae) normally a tree (2.5–12 m high), *Aerva lanata* (Amaranthaceae, perennial herb)*,Citrullus colocynthis* (.Cucurbitaceae), it resembles a common melon vine, but it carries small hard fruits with bitter pulp., *Ochradenus baccatus* (Resedaceae) perennial, 0.5-2 m high, with woody bases and many fleshy green smooth branches and *Olea europaea, (.* Oleaceae)*,* the wild olive is regarded as a small-fruited subspecies of the commercial olive, which were collected from wild shrub populations growing in sandy soils in the Egyptian eastern desert, approximately 1200 km south of Cairo. The plants were investigated in terms of the chemical content of the active chemical groups and tested against ten strains of microorganism.
Our target in the present program for scientific research is the discovery of new drug crude material from plants in the Egyptian eastern desert. The studied plants have potent active phytochemical compounds which may be a promised source for therapeutic purposes or intermediate active compounds for synthesis of useful drugs.

Chemical constituents: table (2)
All the extracts (alcohol and aqueous) contained carbohydrate and / or glycoside, flavonoids. *Acacia etbaica* showed high tannin content while *Aerva lanata* was very rich in flavonoids and coumarins while citrullus has huge sterols and terpenes. Anthraquinos were absent in all the plants. All the plant extracts contained saponins except citrullus, Ochradenus and Olea. Alkaloids were absent in citrullus and with average amounts in rest plants. Tannins were absent in *Aerva lanata* and *Ochradenus baccatus*

Antimicrobial activity

Tables 3 and 4 show the result of antimicrobial activity of extracts under test against four G-⁻ , four G+ , one yeast and one fungi using amoxicillin as antibiotc for bacteria and canestine as standard for yeast and fungi. It is clear that 80% alcohol extract and water extract in general showed more or less similar activities. However the highest activities were given by water extracts of *Ochradenus baccatus*, *Aerva lanata* and *Citrullus colocynthis* on *P. vulgaris* (inhibition zones 15,12,12mm, respectively) and both water and alcohol extract of *Aerva* on (G+) *L. breveis*, with inhibition zone 15mm. It is interested that alcoholic extract of olive tree was active on all tested organisms except *A. niger*, while its water extract was either not active or has low effect. It is also interested thatl alcoholic extracts of *Citrullus colocynthis* was inactive while water extract showed effect on *E. coli*, *P. vulgaris*, *B. subtilis*, *S. typhi, L. brevis* and *S aureus.* Remaining results were more or less comparative.
Many Investigators have studied the inventory and evaluation of wild plants from the standpoint of chemical and biological effects, such as, Kubmarawa *et al*. 2007 [27], who studied antimicrobial activity from ethanol extracts of fifty species of wild plants and studied antimicrobial activity against test strains of *Escherichia coli, Bacillus subtilis, Staphylococcus aureus, Pseudomonas aeruginosa* and *Candida albicans.* They recorded that twenty eight extracts prevents growth of one or more of test pathogens. Some plant extracts showed a wide ring of anti-microbial activity. These results showed that the selected plant extracts have antimicrobial properties and can be used to control biological bacterial cultures. It is recorded that the plant extracts differed in their effectiveness to prevent the growth of pathogens that have been tested.

Table 1. ilies, locations and dates of collection of the investigated plants.

Plant Name	ily	Location	Collection Date
Acacia etbaica	Fabaceae	Wadi Kansisrob (GA)	Oct.- 2016
Aerva lanata	Amaranthaceae	Wadi Yahmib (GA)	Feb-2016
Citrullus colocynthis	Cucurbitaceae	Coastal road Wadi kraaf	Dec-2015
Ochradenus baccatus	Resedaceae	Wadi Sarmatay (GA)	Dec 2015
Olea europaea	Oleaceae	Wadi Yahmib (GA)	Dec-2015

Table 2. Phytochemical screening of 80 % alcoholic extract of some plants growing wild in Egypt

Plant Name	Carbohydrates and /or glycoside	Sterol and terpenes	Flavonoids	Tannins	Alkaloids	Saponins	Coumarins	Anthraquinones
Acacia etbaica	M	M	M	H	A	M	M	A
Aerva lanata	H	M	H	A	L	L	H	A
Citrullus colocynthis	M	H	M	L	M	A	A	A
Ochradenus baccatus	M	M	M	A	M	A	M	A
Olea europaea	M	M	M	M	L	A	M	A

M= moderate H= High L= Low A= Absent

Table 3. Antimicrobial activities of the 80 % alcoholic extract of some plants growing wild in the Egyptian eastern desert.

Test	Inhibition zone (mm in diameter) ± SE					
	Plant					
Bacteria (G -ve)	*A.etbaica*	*A. anata*	*C.colocynthi*	*O. baccatus*	*O.europae*	Standard 100µg/
Escherichia coli	LA	NA	NA	LA	9.00± 0.13	16 ± 0.6
P. vulgaris	LA	12.00 ± 0.29	NA	7.00 ± 0.14	8.00 ± 0.12	21 ± 0.90
P.fluroscens	NA	NA	NA	NA	LA	26± 0.39
S. typhi	LA	NA	NA	NA	8.00 ± 0.14	19 ± 0.83
Bacteria(G +ve)						
B. subtilis-NRRL–B543)	LA	20.00 ± 0.24	NA	LA	9.00 ± 0.11	24± 0.51
C. sp	NA	NA	NA	7.00 ± 0.11	LA	N-T
L.breveis	LA	15.00 ± 0.21	NA	NA	LA	N-T
S.Aureus	NA	12.00 ± 0.11	NA	NA	7.00 ± 0. 12	22 ± 0.80
Yeast						
C. albicans	7.0 ± 0.14	NA	NA	7.00 ± 0.15	9.00 ± 0.14	12 ± 0.53
Fungi						
A. niger	NA	NA	NA	NA	NA	9 ± 0.30

LA = Low activity NA= Not active N-T= not tested
Standard for bacteria= amoxicillin, standard for yeast and fungi=canestin.

Table 4. Antimicrobial activities of water extract of some plants growing wild in the Egyptian eastern desert .

Test	Inhibition zone (mm in diameter) ± SE					
	Plant					
Bacteria (G -ive)	*A. etbaica*	*A lanata*	*C. colocynthi*	*O. baccatus*	*O. europaea*	Standard 100µg/
E. coli	12.00 ± 0.11	12.00 ± 014	12.00± 0.13	LA	NA	16 ± 0.6
P. vulgaris	LA	12.00 ± 0.6	12.00 ± 0.14	15.00	LA	21 ± 0.90
P.fluroscens	LA	NA	NA	NA	LA	26 ±0.39
S. typhi	LA	NA	LA	LA	NA	19 ± 0.83
Bacteria(G +ive)						
B. subtilis-NRRL–B543)	12.00 ± 0.22	NA	12.00± 0.18	LA	LA	24± 0.51
C. sp	LA	NA	NA	LA	LA	N-T
L.breveis	LA	15± 0.22	LA	13.00 ± 0.15	LA	N-T
S.aureus	LA	12 ± 0.19	LA	LA	NA	22 ± 0.80
Yeast						
C. albicans	13.00 ± 0.19	NA	NA	LA	NA	12 ± 0.53
Fungi						
A. niger	LA	NA	NA	LA	LA	9± 0.30

LA = Low activity NA= Not active N-T= not tested
Standard for bacteria= amoxicillin, standard for yeast and fungi=canestin.

Conclusion

The human knowledge and custom assessed the fact that plants possess many medicinal, traditional and pharmacological uses make them very useful and their extracts could be beneficial in treatment of man ailments. The plants under test in this work have potent active phytochemical compounds that may be a promised source for therapeutic purposes or intermediate materials for synthesis of useful drugs.

References

1. M.A. Zahran, AJ. Willis, *The Vegetation of Egypt. Springer Science & Business Media. ISBN 978-94-015-8066-3* (2013). 7–8.
2. M. P. Gupta, Solis, P. N., Calderon, A. J., Guinneau – Sinclair, F., Correa, M., Gladames, C., Guerra, C., Espinosa, A., Alvenda, G. L., Robles G & Olampo, R., *J Ethnopharmacol* 96 (2005) 389.
3. D. S. Sandhu, M. Heinrich, *Phytother Res* 19 (2005) 633.
4. O. T. Erdogrul, *Pharmaceutical Biol.* 40 (2002) 269.
5. V. Bolzani, M. Valli, M. Pivatto, C. Viegas, *Pure Appl. Chem*. 84(2012) 1837–1846.
6. G. Yaseen, M. Ahmad, S. Sultana, A. S. Alharrasi, J. Hussain, M. Zafar, S. Ur-Rehman. *J Ethnopharmacol*. 163(2015) 43–59.
7. G. Mustafa,S. Ahmed, N Ahmed, A. Jamil, *Pak. J. Bot*, 48(2016) 2057-2062.
8. S. Khan, M. Imran, M. Imran, N. Pindari, *Bioinformation* 13 (2017) 67-72
9. H. Khalid, W.E. Abdalla, H. Abdelgadir, T. Opatz, T. Efferth.. *Nat. Prod. Bioprospect.* 2(2012) 92–103.
10. A. F. Ibrahim, A. A. Bellail , A. M. Hamad, *Ijsrm.Human,* 5 (2017): 79-94.
11. .S. Khayyat, M. O. AL-Kattan3, *Biomed Res- India* 28 (2017) 389-393
12. V. Tackholm, *Student Flora of Egypt, Second edition. Cairo University* (1974).
13. L. Boulos, *"Flora of Egypt Checklist," Revised Annotated Edition, Al-Hadara Publishing, Cairo*, 2009 *ISBN*:9048185955.
14. AJ. Ayman, A E. Esam, M M. Adam, A . Omer, HY.Abdul *, E-Journal of Chemistry* , 9 (2012)851.
15. AM . Ramzi, M.A . Nawal, FA. Mohamed, C. Paul, M. Louis, *Evid Based Complement Alternat Med* (2014) 7
16. W. Emily,L. Brian, *Altern Med Rev* 12 (2007) 4 .
17. A. E. Al-Snafi,. *A review, IOSR PHR* , 6, (2016) 57-67.
18. H. Wagner, . *Drogenanalys*e, *Springer Verlag Berlin Heidelberg New York ISBN* :0849319617 (1983) 522.
19. Y. A.Békro, J. A. M. Békro, B. B., Boua, , B. F. H. Tra, , E. E. Ehilé, *Rev. Sci. Nat*.4 (2007) 217.
20. J.W. Fairbrain, *J. Pharm. Pharmacol.* 1 (1949) 683.
21. C. T.Collins, P. M.L. yne, *Microbiological Methods (5th Edn),* Butterworth *and Co Pub Ltd, London and Toronto ISBN* 0 340 80896 9 (1985) 167.
22. M. Hossain, M. R. Nagooru, *Phcog J.* 3 (24) (2011) 25.
23. S.A. El-Toumy, F.S. El-Sharabasy, H. Z. Ghanem, M. U. El-Kady, Asmaa F. Kassem, *Aust J Basic Appl Sci5* (8) (2011): 1362.
24. F. El-Shrabasy, Naima Zayed Mahamed, *Intrnational J Pharm SCI)* 5 (1) (2013) 422.
25. P. Cos, A. J. Vlietinck, D.V. Berghe, L. Maes, *J Ethnopharmacol* 106 (3) (2006) 290.
26. M. M. Cowan, *Clin. Microbiol. Rev.* 12 (4) (1999) 564.
27. D. Kubmarawa, GA. Ajoku,NM Enwerem, DA. Okorie, *Afr. J. Biotechnol.*6(2007) 1696.

(2018) ; http://www.jmaterenvironsci.com