The Biological Activity of Alcoholic Extracts of *Cordia myxa* Plant Against *Klebsiella.* Isolated from Infected Patients

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Abstract

Plants have been used as drugs by humans since thousands of years ago . The study aimed to investigate the effectiveness of alcoholic extracts of the fruits , leaves and seeds of ***Cordia myxa*** plant against pathogenic microorganisms (i.e. Klebsiella) . This was done in the laboratories of the Department of Life Sciences / College of Education in Nasiriya southern of Iraq . The results showed that the alcohol extract of the leaf was significantly superior to that of the fruit and seed extract. The results also showed that the growth inhibition and the effectiveness against *Klebsiella* bacteria was at the average concentration of 300mg / mL 32.66 ± 2.08 , representing the highest inhibitory concentration. It was concluded that leaves of the *Cordia myx*a is one of the best parts of the plant that have inhibitory effects on Klebsiella bacteria compared to the fruit and seed .

Keywords: *alcoholic extracts, inhibiton, Cordia myxa, Klebsiella, infections*

Introduction

Medicinal plants are the Nature's gift to human beings to help them pursue a disease-free healthy life. Plants have been used as drugs by humans since thousands of years ago. As a result of accumulated experience from the past generations, today, all the world's cultures have an extensive knowledge of herbal medicine. Two thirds of the new chemicals identified yearly were extracted from higher plants. 75% of the world's population used plants for therapy and prevention. In the US, where chemical synthesis dominates the pharmaceutical industry, 25% of the pharmaceuticals are based on plant-derived chemicals(1). The emergence of herbal medicine goes back to about 6000 years with the discovery of a tomb in the cave in northern Iraq in the year (1960). The analyzes conducted on the soil surrounding the skeleton resulted in the presence of pollen grains for eight plants, seven of which are medicinal, which are still used all over the world (2). *Cordia.myxa L* belongs to the family Ehretiaceae, which contains more than 300 genus and that are distributed in tropical, subtropical and warmer regions around the world. It is a medium-sized deciduous tree, the circumference of the whole bearing tree of the trunk is 75.5 cm, with pruned tree branches, soft wood, light gray, no heartwood(3) . *Cordia.myxa* Tree is a perennial evergreen tree, medium in size if it reaches a height AD 5-7 and lived to 60 years (4) . The fruit is almond with a stone core and is yellow in color and sweetish when ripe. The seed is oval in shape Several chemicals have been identified. Leaves are simple in shape , green in color (5) .

The genus Cordia (containing more than 200 species) is one of the largest genera of this family . The fruits are used to make pickles and have many common anti-bacterial and anti-worm uses (6).It is also widely used in the treatment of diseases of the urinary tract and thoracic tract (7). A botanical study conducted in northwestern Ethiopia reported that *Cordia* africana was traditionally used to treat liver disease, dysentery, stomach pain, and

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diarrhea [8]. The dried leaf powder is used to treat malaria and its associated symptoms by traditional healers[9] . Klebsiella species are routinely found in the human nose, mouth, and gastrointestinal tract as normal flora; however, they can also behave as opportunistic human pathogens , it can lead to a wide range of disease states, notably pneumonia, urinary tract infections, sepsis, meningitis, diarrhea, peritonitis and soft tissue infections it may be treated by leaves of *Cordia leucocephala*, popularly known as "Maria Brita", are used as an infusion to treat dysmenorrhea[10,14] . The genus Cordia belong to the Kingdom : Plantae; Subkingdom: Tracheobionta; Superdivision: Spermatophyta; Division: Magnoliophyta; Class: Magnoliopsida; Subclass: Asteridae; Order: Lamiales, Family: Eretiaceae, Genus: *Cordia* L., Species: *myxa* L[1].

Materials and Method

The parts of the *Cordia myxa* tree collected all of the fruits, leaves and seeds from some scattered areas in the city of Nasiriyah, where they were cleaned and washed well with running water to get rid of impurities and dried, and left in the shade to dry, exposed to the air, after which they were ground by an electric grinder and placed in opaque bottles. The alcoholic extract (ethanol) extract was prepared according to the method of Harborne[11]. In this study, *Klebsiella* bacteria were used that were previously diagnosed from patient samples and were taken from the Microbiology Laboratory in the Department of Life Sciences at the College of Education for Pure Sciences at. Dhi Qar University, in order to evaluate the effectiveness of the and alcoholic extract towards the pathological bacterial isolate used during the study, the agar well diffusion method was used as it was mentioned in Hammer [12] . Muller Huntington agar medium was prepared and the bacterial isolates used were activated on this medium and then placed in the incubator at a temperature of 37 ° C for a period of 24 hours after which a bacterial suspension was prepared from the developing colonies at a concentration (6 x 10 ^{6}) Using the physiological solution, and comparing the number of bacteria to the previously prepared McFarland. Take 100 ml of the bacterial suspension by means of a sterile micropipette and pour it over the culture medium, then spread the suspension with a glass diffuser spreader sterilized with alcohol and flame, then leave the dishes for one hour to dry the suspension. Three pits with a diameter of 7 mm were made in each dish using a sterile metal cork borer, then 100 micrometers of each concentration of the extract were placed in each hole using a small volume pipette micropipette, as one dish contains three concentrations (100 micrograms / ml and 200 micrograms / ml. And 300 µg . The dishes were incubated in the incubator at 37 ° C for 24 hours, after which a transparent corona was observed around each hole, which represented the diameter of the inhibition zone. The diameter of the inhibition zone was measured using a ruler by taking the average of two perpendicular diameters measured in mm. were measured.

Results and Discussion

Klebsiella organisms can lead to a wide range of disease states, notably pneumonia, urinary tract infections, sepsis, meningitis, diarrhea, peritonitis and soft tissue infections . Klebsiella species have also been implicated in the pathogenesis of ankylosing spondylitis and other spondyloarthropathies[14]. The majority of human Klebsiella infections are caused by K. pneumoniae, followed by K. oxytoca. Infections are more common in the very young, very old, and those with other underlying diseases, such as cancer , and most infections involve contamination of an invasive medical device . It was noted through Table (1) that all the concentrations of the ethanol solvent used were effective and with all the concentrations used, and the concentration of 300 mg was one of the best alcohol concentrations used, and through the statistical analysis of the fruit, it was noticed that there were no significant differences between the average concentration of 100 mg / mL of 25.66 ± 1.52^{b} and the average The average concentration of 200mg / mL was 26.66 ± 1.52^{b}, but there was a significant difference in the concentration of 300mg / mL 29.66 ± 0.57^{a}, as a significant difference was found between it and the concentration of 100mg / mL and the concentration of 200 mg / mL.in the paper, through the statistical analysis, it was noticed that

there are significant differences between the average mean concentration of 100mg / mL 24.33 ± 1.15^{c}, the average concentration 200mg / mL 28.33 ± 1.52b, and the average concentration rate 300mg / mL 32.66 ± 2.08^{a}. As for the seeds, it was observed that there are significant differences between the average mean concentration 100mg / mL 23.55 ± 1.50^{c}, the average concentration rate 200mg / mL 27.55 ± 1.66^{b}, and the average concentration 300mg / mL 31.11 ± 1.76^{a} . and the results of the current study are consistent with the results of the study he did[13] . The inhibitory action of the alcoholic extract of the ***Cordia myxa*** plant may be attributed to the presence of alkaloids and phenols, in addition to triple terpenes, saponins, glycosides, tannins, flavonoids and carotenoids. These substances possess positive and negative bacterial activity as they cause inhibition of the formation of the cell wall of the microorganism or inhibition of the synthesis of essential proteins in it, and the formation of complexes with the cell wall that impede the regularity of permeability, and inhibition of some enzymes that have an important metabolic role in growth and reproduction, and the rupture of cell membranes or change their function. These reasons made the extract an antibiotic and anti-bacterial[15].

Table (1) the effect of the alcohol solvent of the *Cordia myxa* plant extract against *Klebsiella* bacteria that cause infections in human

Extraction	Concentration	Mean ± SD	N
Fruit	100mg	25.66 ± 1.52b	3
	200mg	26.66 ± 1.52b	3
	300mg	29.66 ± 0.57a	3
	Total	27.33 ± 2.21	9
Leaf	100mg	24.33 ± 1.15c	3
	200mg	28.33 ± 1.52b	3
	300mg	32.66 ± 2.08a	3
	Total	28.44 ± 3.87	9
Seed	100mg	23.66 ± 1.52c	3
	200mg	27.66 ± 2.08b	3
	300mg	31.0 ± 1.0a	3
	Total	27.44 ± 3.46	9
Total	100mg	24.55 ± 1.50c	9
	200mg	27.55 ± 1.66b	9
	300mg	31.11 ± 1.76a	9
	Total	27.74 ± 3.15	27
L.S.D	2.05		

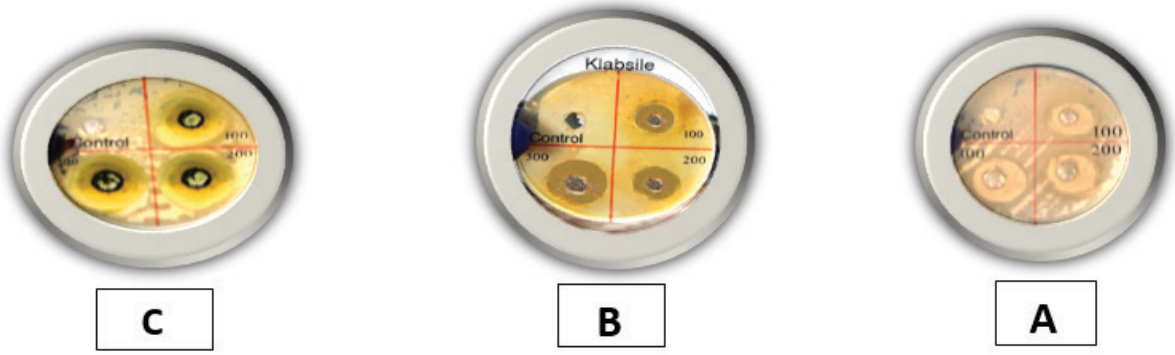

Figure (1) inhibitory effecrs of the alcoholic extract for the (A) leaves, (B) fruits, (C) seeds of the *Cordia myxa* plant, against *Klebsiella* bacteria that cause infections in human .

Conclusion

The possibility of adopting the *Cordia myx*a as an influential medicinal plant, as an important and essential source of effective compounds and nutrients, and this confirms the importance in terms of nutritional and therapeutic . The results of the study showed that the *Cordia myx*a contains high concentrations of total phenols that act as antioxidants and have proven their efficiency in scavenging free radicals, in addition to resistance to dangerous diseases related to free radicals . The leaf of the *Cordia myx*a is one of the best parts of the plant used in the current study in terms of the inhibitory effectiveness of bacteria (Klebsiella) compared to the fruit and seed used in the study.

Ethical Clearance : Taken from University of Thi-Qar ethical committee

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Conflict of Interest : Nil

References

(1) Al-Snafi, A. E. The Pharmacological and therapeutic importance of Cordia myxa-A review. IOSR Journal of Pharmacy. 2016 ; 6(6):47-57.

(2) Solecki, R. ,and Shanidar, I.V. A Neanderthal flower burial in northern Iraq . Science, 1975 : 190 : 80-881.

(3) Gupta, R., and Gupta, G. D. A review on plant Cordia obliqua Willd.(Clammy cherry). Pharmacognosy Reviews, 2015 ; 9(18): 127.

(4) Townsend, C. C.and Guest E. Flora of Iraq-Part two Ministry Vol. four of Agriculture and Agrarian reform, Iraq, 1980 ; l : 627- 1197 .

(5) AbdEl-Aleem, E. R., Seddik, F. E. F., Samy, M. N., & Desoukey, S. Y. Botanical studies of the leaf of Cordia myxa L. J of Pharmacognosy and Phytochemistry, 2017 ; 6(6) : 2086-91.

(6) Silva S.A. Souto, A.L. and Agra, M.F. A new arylnaphtalene type ligan from Cordia rufescens A.DC.(Boraginaceae). Arkivoc, 2004 ; 6:54-58 .

(7) Al-Awadi, F.M. Srikumar, T.S.Anum ,J.T. and Khan, J. Anti- inflamation effects of Cordia myxa fruit on experimentally induced colitis in rats. Nutration . 2001 ; 17:391-396.

(8) Asrie, A. B., Abdelwuhab, M., Shewamene, Z., Gelayee, D. A., Adinew, G. M., & Birru, E. M. Antidiarrheal activity of methanolic extract of the root bark of Cordia africana. Journal of experimental pharmacology, 2016 ; 8: 53.

(9) Wondafrash, D. Z., Bhoumik, D., Altaye, B. M., Tareke, H. B., & Assefa, B T. Antimalarial Activity of Cordia africana Lam. Boraginaceae Leaf Extracts and Solvent Fractions in Plasmodium berghei-Infected Mice. Evidence-Based Complementary and Alternative Medicine, 2019 ; 2019 : 8324596.

(10) De Sousa Nogueira, B. D. S., Antas e Silva, D., Tavares, J. F., De Oliveira Lima, E., De Oliveira Pereira, F., De Souza Fernandes, M. M. M., ... & De Fátima Vanderlei de Souza, M. First chemical constituents from Cordia exaltata Lam and antimicrobial activity of two neolignans.

Molecules, 2013 ;18(9): 11086-11099.

(11) Harborne, J. B. Methods of plant analysis. In Phytochemical methods . Springer, Dordrecht, pp: 1-36 , 1984 .

(12) Hammer, K. A., Carson, C. F., and Riley, T. V. In vitro activity of Melaleuca alternifolia (tea tree) oil against dermatophytes and other filamentous fungi. Journal of Antimicrobial Chemotherapy, 2002 ; 50(2): 195-199.

(13) Sahibt, A. H. A., Al-Shareefi, E., Hameed, I. H. Fruit Extract Using Fourier-Transform Infrared Spectroscopic Technique. Indian Journal of Public Health Research & Development 2019 ; 10 (1), 981-987

(14) Rodríguez-Medina N, Barrios-Camacho H, Duran-Bedolla J, Garza-Ramos U. Klebsiella variicola: an emerging pathogen in humans. Emerg Microbes Infect. 2019;8(1):973-988.

(15) Mahran, G. H., Glombitzam K. w.; Mirhom, Y. w.; Hartmann, R. and Michel, C. G. Novel saponins from Zizipus spina- Christi growing in Egypt. Planta Med., 1996 ; 62:63-165.