



The antibacterial efficacy of the secondary metabolites extracted from (*Melia azedarach* L.) leaves against pathogenic microorganisms isolated from burns and gingivitis infections

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Abstract

The current investigation was accompanied to assess the antibacterial efficacy of secondary metabolites compounds, such as: phenolic, terpenoids, and alkaloids extracted from leaves of Chinaberry tree (*Melia azedarach* L., Family: *Meliaceae*) counter to pathogenic microorganisms isolated from burns and gingivitis cases. by using agar well diffusion method. The aimed of this investigation was to assess the antibacterial efficacy of Phenolic, Terpenoids, and Alkaloids extracted from leaves of Chinaberry tree (*Melia azedarach* L.) against some pathogenic microorganisms isolated from burns and gingivitis cases. The data collected from the study revealed that, secondary metabolites compounds, such as: phenolic, terpenoids, and alkaloids extracted from leaves of Chinaberry tree had a range of antibacterial efficacy against pathogenic microorganisms isolated from burns and gingivitis cases. *Streptococcus mitis* and *Staphylococcus aureus* were exhibited fully unaffected to phenolic and terpenoids secondary metabolites extracted from leaves of Chinaberry tree (*Melia azedarach* L.). *Escherichia coli* were fully resistant to terpenoids compounds. *Staphylococcus aureus* were fully resistant to alkaloids compounds. In conclusion, secondary metabolites compounds extracted leaves of Chinaberry tree (*Melia azedarach* L.) considered a good source for make drugs against microorganisms isolated from burns and gingivitis cases especially alkaloids compounds.

Keywords: antibacterial, secondary metabolites, melia azedarach, pathogenic microorganisms

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INTRODUCTION

Secondary metabolites of plants can be defined as organic compounds produced as by-product as a result of primary metabolic activity such as the manufacture of proteins, fats, carbohydrates and nucleic acids. It has many important functions of the plant body such as sources of storage and transport of nitrogen; protect the plant from ultraviolet rays, factors to attract insects for the purpose of completing the pollination process as well as the shed and distribution of seeds by animals and the key role of secondary metabolites is believed to serve as chemical defenses mechanisms counter to herbivores and pathogens (Chen 2008). Alkaloids are one of the largest secondary chemical compounds groups in plant; generally include those basic substances which contain one or more nitrogen atoms (Harborne 1984). Alkaloids compounds are mainly derived from amino acid by process called Protein metabolism (Raffauf 1996). Alkaloids are mainly crystalline and colourless in appearance except few like Nicotine is liquid at room

temperature (Cowan 1999, Harborne 1984). Terpenoids also can be defined as aromatic compounds that have ability to dissolve into fat. Their locations are either in the cytoplasm or in special glands such as volatile oils (Harborne 1993). The essential oil considered key terpenoids that give to plants special odor and smell (Stipanovic 1983). Terpenes have some properties such as food preservatives, antimicrobial agents, feeding stimulants and antiseptic activities (Tyler *et al.* 1988). Phenolic compounds are secondary metabolic compounds produced in plants, which possess in common an aromatic ring bearing one or more hydroxyl substituents C-OH, Phenolic compounds are soluble in water, which sometimes combined with sugar molecule as glycosides and they are usually located in cell vacuoles (Harborne 1984). The tree of *Melia azedarach*

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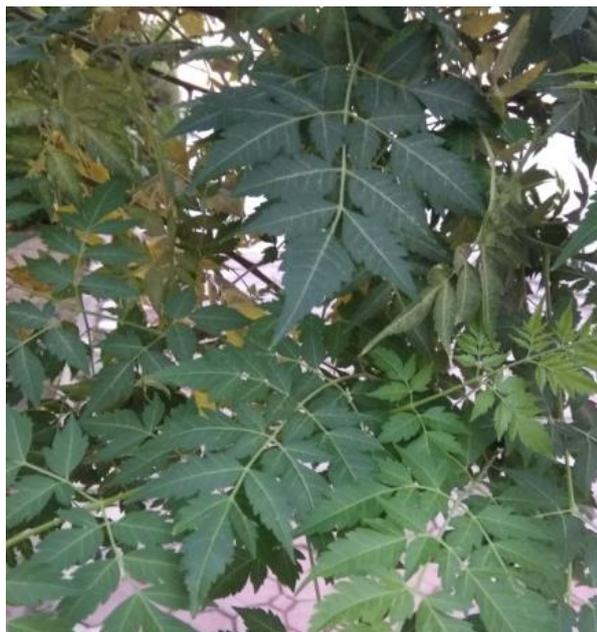


Fig. 1. *Melia azedarach* L.

is classified in Meliaceae (Mohangy); family (Khan *et al.* 2008). There are (2-15) species in the tropical and subtropical regions of the old world, one species widely grown in Iraq as a shade and ornamental tree. *Melia azedarach* is deciduous trees or shrubs, reach to (12 m) in length, with a thick trunk and spreading branches. Leaves (2-3) pinnate and (30-100 cm) long; leaflets (3-13) opposite, sub opposite or alternate, elliptic-ovate to lanceolate (2-5 cm) long, serrate or more rarely shallowly lobed or subentire, the base regular or sometimes weakly asymmetrical, sparingly pubescent along the margins and midrib or glabrous with evanescent stellate hairs when young. *Melia azedarach* has different common name in different countries, in India called Pride of India, in China known as Chinaberry, in Australia known as Bastard cedar or Cape lilac, in Iran known as Zanzalakht, and in Iraq known as Sibahbah or Rosary tree or Hawthorn or Lilac (Townsend and Guest 1974). However, the aimed of this investigation to investigate the biological activity of secondary metabolites like Phenolic, Terpenoid, and Alkaloid extracted from leaves of (*Melia azedarach* L.) against to pathogenic microorganisms isolated from burns and gingivitis infections.(Bagheri *et al* 2018, PONS11 2017).

MATERIALS AND METHODS

Collection and Identification of Plant Material

Leaves of chinaberry (*M. azedarach* L.) were collected from gardens at University of Babylon, during October 2018. Plants were identified at Babylon University, College of Science for women (Fig. 1). Leaves of this plant were cleaned, dried, and kept according to (Harborne *et al.* 1975).

Table 1. The antibacterial effect of phenolic compounds extracted from (*Melia azedarach* L.) against pathogenic microorganisms

Bacteria	Phenolic compounds		
	Concentration (mg/ ml)		
	100	200	300
	Inhibition zone/mm		
<i>Escherichia coli</i>	0±0	0±0	13± 1
<i>Pseudomonas aeruginosa</i>	0±0	0±0	13± 1
<i>Staphylococcus aureus</i>	0±0	0±0	0±0
<i>Streptococcus mitis</i>	0±0	0±0	0±0
<i>Lactobacillus acidophilus</i>	0±0	12± 1	15±1

*Mean± standard deviation

Extraction of the Crude Alkaloid Compounds

Crude Alkaloid compounds were extracted according to (Harborne 1984).

Extraction of the Crude Terpenoid Compounds

Crude terpenoids compounds were extracted according to (Harborne 1984).

Extraction of the Crude Phenolic Compounds

Crude Phenolic compounds were extracted according to (Ribereau-Gayon 1972).

Antibacterial Activity

The anti-bacterial activity of the secondary metabolites extracted from the (*Melia azedarach* L.) was tested against the isolated bacteria by using agar-well diffusion method; Measurement of diameters of clear inhibition zones was recorded as antibacterial activity according to Perez *et al.* (1990).

Microorganism Strain

Escherichia coli, *Pseudomonas aeruginosa*, and *Staphylococcus aureus* are isolated from people suffering from Burns, while *Streptococcus mitis* and *Lactobacillus acidophilus* are isolated from people suffering from Gingivitis.

RESULTS

The biological activity of secondary metabolites like phenolic, terpenoids, and alkaloid compounds extracted from (*Melia azedarach* L.), leaves against human pathogenic organisms is presented in **Tables 1-3**.

The results obtained from **Table 1** revealed that, that, gram-negative bacteria such as *Escherichia coli* and *Pseudomonas aeruginosa* unaffected to the phenolic compounds at concentrations of (100 and 200 mg/ml) and showed sensitivity to the phenolic compounds at high (300mg/ml) concentrations with inhibition zone reached to 13±1 mm/diameter. On the other hand, gram-positive bacteria such as *Streptococcus mitis* and *Staphylococcus aureus* showed entirely unaffected to all concentration of phenolic secondary metabolites. While *Lactobacillus acidophilus* showed sensitivity to two concentrations (200 and 300) mg/ml with inhibition zones reached to 12± 1 and 15± 1 respectively.

The present investigation also revealed that, gram-negative pathogens like *Escherichia coli* and

Table 2. The antibacterial effect of terpenoids compounds extracted from (*Melia azedarach* L.) against pathogenic microorganisms

Bacteria	Terpenoids compounds		
	Concentration (mg/ ml)		
	100	200	300
	Inhibition zone/ mm		
<i>Escherichia coli</i>	0±0	0± 0	0±0
<i>Pseudomonas aeruginosa</i>	0±0	0± 0	0±0
<i>Staphylococcus aureus</i>	0±0	0± 0	0±0
<i>Streptococcus mitis</i>	0±0	11± 0.5	18± 0.5
<i>Lactobacillus acidophilus</i>	0± 0	0± 0	0± 0

*Mean± standard deviation

**Fig. 2.** The antibacterial effect of terpenoids compounds extracted from (*Melia azedarach* L.) against *Streptococcus mitis***Table 3.** The antibacterial effect of alkaloids compounds extracted from (*Melia azedarach* L.) against pathogenic microorganisms

Bacteria	Alkaloids compounds		
	Concentration (mg/ ml)		
	100	200	300
	Inhibition zone/mm		
<i>Escherichia coli</i>	0±0	13± 1	16± 0.5
<i>Pseudomonas aeruginosa</i>	0±0	0±0	15
<i>Staphylococcus aureus</i>	0±0	0±0	0±0
<i>Streptococcus mitis</i>	0±0	13± 0.5	15± 0.3
<i>Lactobacillus acidophilus</i>	0±0	0±0	15± 1

*Mean± standard deviation

Pseudomonas aeruginosa and gram-positive microorganisms such as *Staphylococcus aureus* and *Lactobacillus acidophilus* completely resistant to terpenoids compounds at different concentrations (100, 200, and 300) mg/ml (**Table 2**). Except, *Streptococcus mitis* was showed sensitivity to terpenoids compounds at two concentrations (200 and 300) mg/ml with inhibition zone reached to (11± 0.5 and 18± 0.5) respectively (**Fig. 2**).

In context, results of antibacterial activity of alkaloids compounds extracted from (*Melia azedarach* L.) against pathogenic microorganisms were described in **Table 3**.

**Fig. 3.** The antibacterial effect of terpenoids compounds extracted from (*Melia azedarach* L.) against *Pseudomonas aeruginosa*

In the present study, alkaloids compounds unaffected in gram-negative and gram-positive microorganisms in concentration (100) mg/ml. But, was showed activity at high concentration (300) mg/ml in all bacteria in this study except *Staphylococcus aureus* was showed totally resistant to all concentrations of alkaloids compounds (**Fig. 3**).

DISCUSSION

Past and present medicinal plants play an important role in human life because of they possess effective compounds with a high ability to treat diseases. In recent times, many antibiotic-resistant microorganisms have emerged. Therefore, new sources must be required to overcome the resistance of these dangerous microorganisms. Many scientists believe that the solution exists in the plant kingdom. Khan *et al.* (2011) reported that, seeds extract with different solvents demonstrated antibacterial activity against microorganisms isolated from hospitals. Ramya *et al.* (2009) and Neycee *et al.* (2012) were used leaves extract of (*Melia azedarach* L.) as antibacterial by using disk diffusion method and showed significant results counter to gram-negative and gram positive bacteria. Meziane and Goumri (2014) Were used essential oils extracted from different parts of (*Melia azedarach* L.) such as leaves, seeds, and flowers as antimicrobial against yeast and other microorganisms like *E. coli*, *S. aureus*, and *E. aerogenes*. Al-Khafaji *et al.* (2016) Were used fruit extract of (*Melia azedarach* L.) as antibacterial by using well diffusion method against pathogenic bacterial like *E. coli*, *P. aeruginosa*, *B. subtilis*, *S. aureus*, *Klinsiella*, and *Proteus* and showed significant inhibition.

Bioactive compounds such as phenolic, terpenoids, and alkaloids extracted from several medicinal plants like (*Lactuca serriola* L., *Lepidium sativum* L., *Myrtus Communis* L., *Cassia senna* L., *Ricinus communis* L. and *Cassia didymobotrya* (Fresenius) Irwin & Barneby) have antibacterial efficacy against different pathogenic microorganisms (Al-Marzoqi *et al.* 2015, 2016, Hussein *et al.* 2017, 2018a, 2018b, 2019). Hussein *et al.*, (2018c) Used primitive plant like *Chlorella vulgaris* as antibacterial against pathogenic microorganisms. Kamal *et al.* (2019) was used *Hibiscus sabdarifa* extracts against members of Enterobacteriaceae microorganisms. In contrast, natural bioactive compounds extracted from medicinal plants make their effects by many mechanisms, for example polyphenols binding with polysaccharides and proteins (Macromolecules), thus inhibiting their roles in biochemical metabolites. Terpenoids and flavonoids make their effects by disruption of microbial membranes

and Polypeptides embarrassment of linkage of bacterial proteins to host polysaccharide receptors and alkaloids complexes make their effect by inhibiting of efflux pump (Okusa *et al.* 2009). Sultana *et al.* (2013) were screened many secondary metabolites compounds like phenols, alkaloids, flavonoids, saponins, tannins, and cardiac glycosides in leaves extract of (*Melia azedarach* L.). Finally, antibacterial efficacy of (*Melia azedarach* L.) might be belonging to phytochemical compounds and their effect in proteins and polysaccharides and disruption in membranes permeability or inhibiting of efflux pump.

CONCLUSION

Secondary metabolites compounds extracted from leaves of (*Melia azedarach* L.) considered a good source for making drugs against microorganisms isolated from burns and gingivitis.

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