

Against Antibacterial and Antifungal Activity of Jojoba Wax Liquid (*Simmondsia chinensis*)

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ABSTRACT

Introduction: Plants are a rich source of bioactive compounds. *Simmondsia chinensis*, also known as Jojoba, is the sole member the Simmondsiaceae's family and has been known traditionally for many medical uses. Objectives: Herein we evaluate the value of crude jojoba oil (J.O) as an antimicrobial agent *in vitro*. **Methods:** J.O was tested for potential antimicrobial activity against *Bacillus subtilis*, *Staphylococcus aureus*, *Proteus vulgaris*, *P. mirabilis*, *Salmonella typhimurium*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Candida albicans* and *Aspergillus flavus*. **Results:** Our results did not show any effect on fungi or yeast. However, a significant antibacterial activity was observed against *B. subtilis*, *S. aureus*, *P. vulgaris*, *P. mirabilis*. A high activity was observed for J.O at Minimum inhibitory concentration (MIC) level of 12.5 mg/ml. Interestingly, *S. typhimurium*, *E. coli* and *Ps. aeruginosa* were found to be highly resistant. **Conclusion:** Our findings suggest that J.O may have a medicinal potential as natural antibacterial agent.

Key words: Jojoba oil, Antibacterial, Antimicrobial activity, *Simmondsia chinensis*, Minimum inhibitory concentration (MIC).

INTRODUCTION

There are thousands of species of medicinal plants used globally for the cure of different infections.^{1,2} For example previous studies proved that the extracts of *Casuarina equisetifolia* Forest., *Euphorbia hirta* L. and *Euphorbia tirucalli* L. have antibacterial activities.³ These plants, and others, are used as antimicrobial agents and, extensive work has been carried out to determine their scientific basis.^{4,5,6,7,8} Other plants including *Zingiber officinale*, have been used as herbal drugs to treat inflammation by local inhabitants from ancient times until today.⁹ The medicinal value of the plants lies in some chemical substances that can either produce a definite physiological action on the human body or even act as antibiotics by attacking bacterial cells.^{10,11,12}

Simmondsia chinensis is a plant from the family (*Simmondsiaceae*) known as Jojoba. It is native of southern Arizona, USA. The seeds of Jojoba plant produce more than 45% w/w of colourless, odourless oily material which was discovered by the native Americans who recognised its important medicinal values.^{13,14} Due to its high economic value, Jojoba is being cultivated in different parts of the world including the Egyptian dessert and Saudi Arabia.^{15,16} Many studies have focused on understanding the antibacterial features of Jojoba.^{17,18,19}

Jojoba oil has a unique chemical structure; it is composed of oil sterols, and different tocopherols.^{13,20,21}

Jojoba seeds also contain a considerable amount of tannins.^{22,23,24} It has straight chains of C-20 and C-22 acids and alcohol monoesters, in addition to some triglycerides and stanols.^{13,25} Flavonoids are believed to be responsible for the antibacterial activity of Jojoba oil.^{26,27} Since it also works as a carrier substance for oxidation sensitive materials such as Vitamin A; the crude J.O. was used as a cosmetic and skin care material.^{26,27,28,29} Moreover, Jojoba wax has been shown to be the best liquid wax to stabilize penicillin products.²⁸

The aim of the present work is to study the antimicrobial activity of J.O at different concentrations against different microorganisms.

Experimental MATERIALS

Tested oil Crude J.O. was obtained from Egyptian Natural Oil Company. It was prepared from Jojoba nuts (*Simmondsia Chinensis*).

Microorganisms The microorganisms used in this study were isolated locally and consisted of bacterial and fungal strains. These strains (*Bacillus subtilis*, *Staphylococcus aureus*, *Proteus vulgaris*, *P. mirabilis*, *Salmonella typhimurium*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Candida albicans* and *Aspergillus flavus*) were obtained from the Microbiological Department of Animal Health Research Institute

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Table 1: Antimicrobial activity of jojoba oil on different tested microorganisms tested using agar gel growth inhibition test.

Micro-organism	conc mg/ml	Standard strain +J. O				Tested strain +J. O			
		12.5	25.5	50	100	12.5	25.5	50	100
<i>Bacillus subtilis</i>	Inhibition zone diameter mean value (mm)	10.07	15.13	22	27.1	10	15	22	27
	Standard deviation	0.351	0.306	0.2	0.346	0.3	0.231	0.2	0.3
<i>Staphylococcus aureus</i>	Inhibition zone diameter mean value (mm)	10.1	13.07	20.3	24.1	10	13	20	24
	Standard deviation	0.404	0.416	0.889	0.436	0	0.3	0.3	0.3
<i>Salmonellatyphimurium</i>	Inhibition zone diameter mean value (mm)	-----*	-----	-----	-----	-----	-----	-----	-----
	Standard deviation	-----	-----	-----	-----	-----	-----	-----	-----
<i>Escherichia coli</i>	Inhibition zone diameter mean value (mm)	-----	-----	-----	-----	-----	-----	-----	-----
	Standard deviation	-----	-----	-----	-----	-----	-----	-----	-----
<i>Pseudomonasaeruginosa</i>	Inhibition zone diameter mean value (mm)	-----	-----	-----	-----	-----	-----	-----	-----
	Standard deviation	-----	-----	-----	-----	-----	-----	-----	-----
<i>Proteus vulgaris</i>	Inhibition zone diameter mean value (mm)	8.1	10.1	14.17	20				
	Standard deviation	0.361	0.265	0.451	0.1				
<i>P.mirabilis</i>	Inhibition zone diameter mean value (mm)		8.1	10.27	15		8	10	15
	Standard deviation		0.458	0.493	0		0.3	0.1	0.3

*No inhibition zone

Table 2: antimicrobial activity of jojoba oil on different tested fungi.

Strain of Fungi	control	Jojoba concentration			
	0.0	12.5	25	50	100
<i>Candida albicans</i>	-----*	---	---	--	--
<i>Aspergillus flavus</i>	-----	---	---	--	--

* No inhibition zones.

(AHRI), Cairo, Egypt. All the used microorganisms were prepared and tested according to Kone man and Cruickshank.^{30,31}

METHODS

Microorganisms maintenance Bacterial strains were grown and maintained on Nutrient Agar slants and on Sabouraud Glucose Agar slants, and then stored at 4°C. Both bacteria and *Candida albicans* were sub-cultured in fresh media at regular intervals while, *Aspergillus flavus* was cultured on Potato Dextrose Agar (PDA) and sub-cultured at regular intervals until used for the antimicrobial tests. All bacterial strains were compared with a reference (standard strains) which were obtained from bacterial strain bank.

All tested strains were prepared and tested against J.O. for estimating the Minimum inhibitory concentration (MIC). Each isolate was tested three times to determine the mean reading. At the same time, the reference isolates, were tested against the same extract with the same concentrations and the same environmental conditions to determine the MIC mean reading (each isolate was tested 3 times)

Antibacterial Activity

The antibacterial activity of J.O was determined using the Agar Diffusion Method with 1 ml of inoculum, containing 10⁵ bacterial cells (Bookye -Yiadam, 1979). Fresh broth cultured of test organisms (standardized

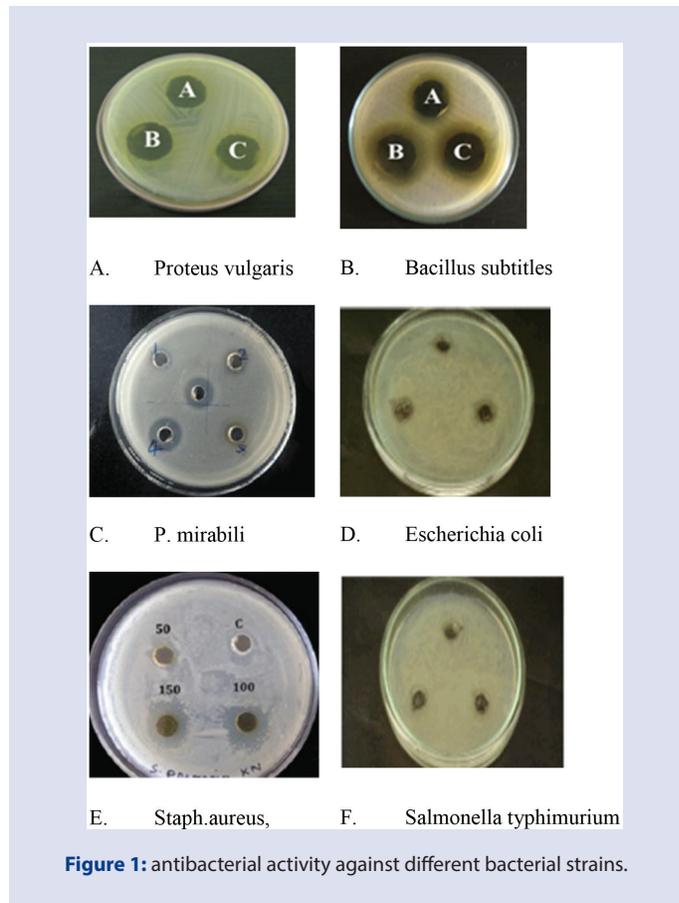


Figure 1: antibacterial activity against different bacterial strains.

inoculate) was swabbed onto sterile Mueller Hinton Agar in petri dishes. A sterile stainless-steel corn borer (12mm) was used to make the wells on the plates. The holes were filled with crude J.O. in different concentrations in water (12.5, 25, 50 and 100 mg/ml). For control experiments, holes were filled with sterile distilled water. Incubated petri dishes were left for an h at room temperature for the J.O. to diffuse before the growth of organisms commenced and then incubated at 37°C for 24h. The microbial growth was determined by measuring the diameter of the zone of inhibition (mm). The experiments were done three times and mean values have been presented in our results.

Antifungal Activity

Pour Plate Method was used for the assay of J.O effect against *Aspergillus flavus* (4x10⁵ fungal spores / plates). J.O was introduced into the test tubes containing sterile Potato Dextrose Agar (PDA). Different J.O Concentrations (12.5, 25, 50 and 100 mg /ml) were used. These were dispensed on petri dishes and could set. Each plate was bored with sterile corn borer of 12 mm in diameter. Control experiments were also set up performed without the presence of J.O. Plates were incubated at 30 °C for 3 days.

Determination of Minimum Inhibitory Concentration (MIC)

After determining the inhibition, the MIC of tested samples at different concentrations was measured against the tested organisms. Agar Diffusion Method described for antibacterial test was also used in determining antifungal action of J.O against *Candida albicans*.

RESULT

Antibacterial Activity

The antibacterial activity tests of J.O was tested against different bacterial strains. Standard bacterial strains obtained from bacterial strain bank were used as a reference. All results are shown below Table 1. Growth inhibition is indicated by clear zones. As shown, the J.O was effective against some of the common bacteria (*B. subtilis*, *S. aureus*, *P. vulgaris*, *P. mirabilis*).

Antifungal Activity

The antifungal activity tests of J.O results are shown below Table 2.

Determination of Minimum Inhibitory Concentration (MIC)

MIC results are shown in both Table 1 and Table 2. Results for Agar Diffusion Method are shown in Figure 1(A-F).

DISCUSSION

The results of testing the antibacterial activity of J.O on nine different microorganisms demonstrated the presence of antimicrobial activity in J.O. These results are in line with previous reports.^{32,33,34} However, the results disagree with *Hani et al* who reported that Jordanian J.O. did not exhibit any antimicrobial activity although it exhibited strong anti-oxidant activity. This can be related to the lower doses used in their experiments.³⁵ In addition, the Jojoba used in our study and that used by *Hani et al* come from two different sources; whether the content and the efficacy of the plant differs when grown in different countries and under different climates is an interesting matter of discussion. In fact, *El-Mallah et al* have reported on the presence of unique properties and differences in the oil components of the Jojoba seeds cultivated in Egypt in comparison with Jojoba seeds cultivated in Arizona,²¹ however further assessment and investigations on this can be the subject of future studies. There was no significant difference in the results observed for our teste bacterial strains and the reference strains, this indicates that the bacteria which was used for the tests had the same expected sensitivity of the standard bacterial strains. Our data showed that the control samples

were not sensitive towards any of the microbial species used. Moreover, *S. typhimurium*, *E. coli*, *Ps. aeruginosa* were not sensitive (with no zone of inhibition) to all concentrations of J.O. Similarly, *C. albicans* and *A. flavus* did not show any sensitivity to J.O.

P. mirabilis was the least sensitive bacterium with 15mm and 10mm zones of inhibition at concentrations of 100 and 50 mg/ml respectively. On the other hand, *B. subtilis* and *S. aureus* were the most sensitive with 10 mm at concentration of 12.5 mg/ml and reached to 27mm at concentration of 100 mg/ml.

Those results reinforce the previous findings on the presence of antimicrobial activities in Jojoba.^{2,36} We do agree that the antibacterial constituents in some plants may not be well effective if the concentrations are inadequate.^{2,35}

CONCLUSION

Our findings suggest that J.O exhibits potent antimicrobial properties. Antimicrobial tests showed that J.O exhibited a broad spectrum of activity by inhibiting the growth of some of the investigated bacteria. J.O appears to be a promising source of bioactive compounds with antimicrobial properties.

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CONFLICT OF INTEREST

The authors certify that they have NO affiliations with or involvement in any organization or entity with any financial or non-financial interest in the subject matter or materials discussed in this manuscript.

ABBREVIATIONS

JO: jojoba oil

MIC: Minimum inhibitory concentration

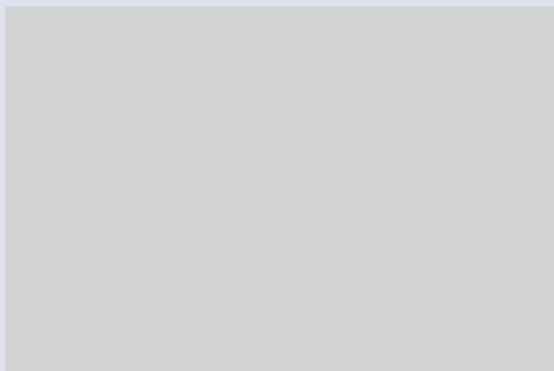
PDA: Potato Dextrose Agar

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GRAPHICAL ABSTRACT



ABOUT AUTHORS



SUMMARY

- Jojoba oil has been attracting researcher's attention in the medical and pharmaceutical field for a while. Many studies about the use of Jojoba oil antibacterial and antifungal activities has been performed with different findings. In this study, we evaluate the value of crude jojoba oil (J.O) as an antimicrobial agent *in vitro*. J.O was tested for potential antimicrobial activity against *Bacillus subtilis*, *Staphylococcus aureus*, *Proteus vulgaris*, *P. mirabilis*, *Salmonella typhimurium*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Candida albicans* and *Aspergillus flavus*. Our results did not show any effect on fungi or yeast. However, a significant antibacterial activity was observed against *B. subtilis*, *S. aureus*, *P. vulgaris*, *P. mirabilis*. A high activity was observed for J.O at Minimum inhibitory concentration (MIC) level of 12.5 mg/ml. Interestingly, *S. typhimurium*, *E. coli* and *Ps. aeruginosa* were found to be highly resistant. These results, suggest that J.O may have a medicinal potential as natural antibacterial agent.

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