

Comparison of the inhibitory activity of *Moringa oleifera* and *Moringa peregrina* extracts against two types of human pathogenic bacteria

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Abstract

The study aimed to investigate the effect of the alcoholic extract of the leaves of two *Moringa* species on two types of bacteria, Gram-positive *Staphylococcus aureus* and Gram-negative *Klebsiella pneumoniae*, which are pathogenic to humans. The results of the analysis of the alcoholic extract of the leaves of two *Moringa* species indicated that the *Moringa oleifera* leaf extract had an inhibitory effect on the growth of *Staph. aureus* bacteria. All concentrations of the ethanolic extract of *Moringa oleifera* leaves had a significant inhibitory effect on the growth of *Staph. aureus* and the effect increases with the increase of the used concentration, as the diameter reached a maximum of 18.17 mm at a concentration of 100 mg ml⁻¹. *Moringa oleifera* leaf extract did not have an inhibitory effect on the growth of *Klebsiella pneumoniae* bacteria at all extract concentrations. The results of the analysis of the alcoholic extract also showed that the *Moringa peregrina* leaf extract had an inhibitory effect on the growth of *Staph. aureus* bacteria. All concentrations of the ethanolic extract of *Moringa oleifera* leaves had a significant inhibitory effect on the growth of *Staph. aureus* bacteria. The effect increases with the increase of the used concentration, as the diameter reached a maximum of 16.17 mm at a concentration of 100 mg ml⁻¹. *Moringa oleifera* leaf extract did not have an inhibitory effect on the growth of *Klebsiella pneumoniae* bacteria at all extract concentrations.

Keywords. *inhibitory activity, Moringa oleifera, Moringa peregrina.*

1.Introduction

Plants contain large and diverse amounts of chemicals that have the ability to prevent and cure many diseases (Jamil et al. 2007). *Moringa* is considered a complete food and has many medicinal uses in addition to its high nutritional value (Sultan et al. 2008). *Moringa* has many pharmaceutical effects that are used in the treatment of many diseases in the traditional medical system (Abalaka et al. 2012). *Moringa* leaf extracts have biological properties, and these properties vary depending on the type of solvent used to extract the active ingredients contained in these leaves (Doughari et al. 2007). *Moringa* has antimicrobial properties, which explains why it is widely used in the treatment of respiratory diseases in humans (Anwar et al. 2007 ; Lockett et al. 2000).). showed that Doughari et al. 2008 the antibacterial activity of the aqueous and ethanolic acetate extracts of *Moringa* leaves was highest in the ethanolic extract, and lowest in the aqueous extract at a concentration of 1 liter. (Renitta et al. 2009 ; Mahewish et al. 2021). found that the ethanolic extract of *Moringa* leaves, flowers and seeds had antibacterial activity against *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Escherichia coli*. (Bukar et al. 2010). found that the ethanolic extract of *Moringa* leaves had a broad spectrum of activity against the tested bacteria. (Devendra et al. 2011; Muhe et al. 2024). noted that the chloroform extract of *Moringa* leaves had an antibacterial activity against *Singh* bacteria. They found that the aqueous, ethanolic, methanolic and hexane extracts of *Moringa* leaves had an inhibitory effect against several strains of bacteria, namely *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Escherichia coli* (Singh , 2015 ; Muhe et al. 2023). The aqueous, ethanolic, methanolic and hexane extracts of *Moringa* leaves had an inhibitory effect against several strains of bacteria, namely *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and the hexane extract produced

the least inhibition of all tested bacteria, while the methanolic extract produced the highest inhibition for all types. This experiment aimed to know the antagonistic activity of the aqueous and alcoholic extracts of Moringa leaves in inhibiting the growth of some types of Gram-positive and Gram-negative bacteria.

II. Materials and Methods

Plant Sample Collection and Preparation

Leaves were collected from the field, cleaned, washed with distilled water, and dried. 20 g of each were air-dried and placed in a Panasonic electric mixer. 200 ml of 70% ethanol was added to the mixture. The mixer was operated for 15 minutes, then the mixture was stirred intermittently in a hotplate magnetic stirrer for 48 hours at 45-50°C. The solution was then centrifuged at 3000 rpm for 30 minutes. The sediment was discarded, and the filtrate was taken. The process was repeated three times to ensure the removal of the sediment. The filtrate was filtered using Whatman No. 1 filter paper. The filtrate was dried in a water bath at 60°C. The extract was placed in sterile bottles and stored in a refrigerator at 5°C until use (Balajee et al. 2004).

Medicinal Efficacy of Plant Extracts

The inhibitory activity of two Moringa species' leaf extracts was tested as a simple cross-linked (CRD) experiment with a single factor comprising four treatments and three replicates. Four concentrations of Moringa species' leaf extracts (0, 25, 50, and 100 mg/ml) were applied to two separate bacterial species.

Bacterial Isolates

Two bacterial isolates were selected: Gram-positive *Staphylococcus aureus* and Gram-negative *Klebsiella pneumoniae*, a human pathogen.

Testing the effectiveness of two Moringa species' leaf extracts against two types of bacteria

The effectiveness of four concentrations of Moringa leaf extract (0, 25, 50, and 100 mg/ml) was tested against the two selected bacteria, as reported in (1999). Smânia et al. and Vanden and Vlietinck (1991) used Mueller Hinton Agar culture medium for bacterial growth, according to the manufacturer's instructions. 15-20 ml of the culture medium was placed in each Petri dish and left to solidify. The plates were then seeded using the spreading method, placing 50 microliters of bacterial suspension prepared from the bacterial isolates. The culture was spread using sterile cotton swabs. 6 mm diameter holes were then made in the agar, into which 0.1 ml of the sample to be tested for the inhibitory effectiveness of the leaf extract was placed, according to the concentrations used. The plates were then incubated for 24-48 hours at 37°C under aerobic conditions. The result was recorded by measuring the diameter of the inhibition zones in millimeters using a ruler.

III. Results and Discussion

It is clear from the results of Table (1) that the *Morinca oleifera* leaf extract has an inhibitory effect on the growth of *Staph. aureus* bacteria. All concentrations of the ethanolic extract of *Morinca oleifera* leaves had a significant inhibitory effect on the growth of *Staph. aureus* bacteria, and the effect increased with the increase in the concentration used, as the diameter reached a maximum of 18.17 mm at a concentration of 100 mg ml⁻¹. The *Morinca oleifera* leaf extract did not have an inhibitory effect on the growth of *Klebsiella pneumoniae* bacteria at all extract concentrations.

Table 1. The effect of *Moringa oleifera* leaf extract on inhibiting the growth of two types of bacteria pathogenic to humans

Inhibition zone diameter (mm)		Leaf extract concentration (mg/ml)
<i>kleb.pneumoniae</i>	<i>Staph. aureus</i>	
0	0	control
0	15.17	25
0	17.00	50
0	18.17	100
0	1.675	LSD 0.05

Table (2) also shows that the *Moringa peregrina* leaf extract has an inhibitory effect on the growth of *Staph. aureus* bacteria. All concentrations of the ethanolic extract of *Moringa oleifera* leaves had a significant inhibitory effect on the growth of *Staph. aureus* bacteria, and the effect increased with the increase in the concentration used, as the diameter reached a maximum of 16.17 mm at a concentration of 100 mg ml⁻¹. The *Moringa oleifera* leaf extract did not have an inhibitory effect on the growth of *Klebsiella pneumoniae* bacteria at all extract concentrations.

Table 2. The effect of *Moringa peregrina* leaf extract on inhibiting the growth of two types of bacteria pathogenic to humans

Inhibition zone diameter (mm)		Leaf extract concentration (mg/ml)
<i>kleb.pneumoniae</i>	<i>Staph. aureus</i>	
0	0	control
0	14.00	25
0	15.17	50
0	16.17	100
0	1.803	LSD 0.05

The antibacterial effect of *Moringa* is due to the fact that its various parts contain numerous biologically active compounds that have beneficial effects. *Moringa* leaves contain many active compounds, which can be attributed to the inhibitory effect (Tables 1, 2). The reason for the difference in the inhibitory effects of the leaf extracts of the two *Moringa* species on bacterial growth is due to the difference in the type and quantity of active substances present in these extracts of the two species, as shown in the gas chromatography results mentioned above. The inhibitory differences resulting from the different concentrations of the extracts used are due to the structural nature of these microorganisms and the concentration of active

substances in these extracts (Awadh et al., 2001). The optimal effects of medicinal plants may not be due to a single active substance, but rather to the interaction of several compounds (Kianbakht and Jahaniani, 2003). Several studies have indicated that plant extracts and oils containing 9,12-octadecadienoic acid (Z,Z) (linoleic acid) and ((Z,Z,Z)-9,12,15-octadecadienoic acid (linolenic acid) exhibit antioxidant and antimicrobial activity (Krishna et al., 2012; Tian et al., 2018). The high content of n-hexadecanoic acid (palmitic acid) in Moringa leaf extract has anti-inflammatory and anticancer effects (Aparna et al., 2012; Korbecki et al., 2019; Kim et al., 2020).

IV. References

- Abalaka, M. E.; S. Y. Daniyan1; S. B. Oyeleke and S. O. Adeyemo .2012.** The antibacterial evaluation of Moringa oleifera leaf extracts on selected bacterial pathogens. Journal of Microbiology Research, 2(2): 1-4. 3
- Anwar, F.; S. Latif; M. Ashraf and A. H. Gilani . 2007.** Moringa oleifera: a food Plant with multiple medicinal uses. Phytother. Res., 21(1):17-25.
- Balajee et. al., 2004.** The ethanolic extracts of leaves of o.sanctum against Cryptococcus neoforms. (1):096-8157.
- Bukar, A.; A. Uba and T. I. Oyeyi . 2010.** Antimicrobial profile of Moringa oleifera Lam. extracts against Mal. J. Microbiol., 8(2): 59-67.
- Cruickshank, R.; G. P. Duguide; B. P. Marmion and R. H. A. Swain .1975.** Medical microbiology, 2nd ed. Churchill Livingstone, Edinburgh, London.
- Devendra, B.N.; N.Srinivas; V.S.S.L. Prasad.Talluri and P. Swarna Latha . 2011.** Antimicrobial activity of Moringa oleifera Lam. Leaf extract against selected bacterial and fungal strains. International Journal of Pharma and Bio Sciences, 2(3): 13-18.
- Doughari, J. H.; M.S. Pukuma and N. De (2007).** Antibacterial effects of Balanites aegyptiaca L. Drel. and Moringa oleifera Lam. on Salmonella typhi. African. J. Biotechnol., 6(19): 2212- 2215.
- Fahey, J. W. 2005 .** Moringa oleifera: A review of the medical evidence for its nutritional, therapeutic and prophylactic properties. Trees of life Journal 2005, 1,5 [Electronic copy: <http://www.tfljournal.org/articla.php/20051201124931586>].
- Farooq, A., Sajid, L., Muhammad, A. and Anwarul-Hassan, G. 2007.** Moringa oleifera: a food plant with multiple medicinal uses. Phytotherapy Research. 21:17 – 25.
- Harbone, J. B. 1982 .** Introduction to ecological biodiversity. London Academic Press: pp. 227-259.
- Jamil, A.; M. Shahid; M. M. H. Khan and M. Ashraf . 2007.** Screening of some medicinal plants for isolation of antifungal proteins and peptides. Pakistan Journal of Botany, 39(1): 211-221.
- Lockett, C.T.; C. C. Calvet and L. E. Grivetti . 2000.** Energy and micronutrient composition of dietary and medicinal wild plants consumed during drought. Int. J. Food Sci. Nutr., 51(3):195-208.
- Renitta, R. E.; J. Anitha and P. Napolean .2009.** Isolation, analysis and identification of phytochemicals of antimicrobial activity of Moringa oleifera Lam. Current Biotica, 3(1):33- 37.
- Singh, S. K. 2013.** Antibacterial activity of different extract of Moringa oleifera leaf against some pathogenic bacteria. Journal of Pharmaceutical and Scientific Innovation, 2(2): 13-15.
- Srinivasan, D.; L. P. Perumalsamy; S. Nathan and T. Sures .2001.** Antimicrobial activity of certain Indian medicinal plants used in folkloric medicine. Journal of Ethnopharmacology, 94: 217-222.
- Stahl, R. 1969.** Thin layer chromatography, A laboratory Handbook, 2nd. Translated by Ashworth M. R. Springer, Verlag, Berlin.
- Suarez, M.; J. M. Entenza and C. Dorries .2003.** Expression of a plant – derived peptide harbouring water – cleaning and antimicrobial activities. Biotechnol. Bioeng., 81:13 – 20.
- Sultan, J.I., I. Rahim, H. Nawaz, M. Yaqoob and I. Javed. 2008.** Mineral composition, palatability and digestibility of free rangeland grasses of northern grasslands of Pakistan. Pak. J. Bot., 40: 2059-2070.



Awadh, A. N.; W. Jülich ; C. Kusnick U. and Lindequist (2001). Screening of Yemeni medicinal plants for antibacterial and cytotoxic activities. *Journal of Ethno pharmacology*, 74(2): 173-179.

Kianbakht, S. and F. Jahaniani (2003). Evaluation of antibacterial activity of *Tribulus terrestris* L. growing in Iran. *Journal of Iranian Pharmacol. Ther.*, 2:22–24.

Tian, Y., et al. (2018). Chemical composition and antibacterial activity of essential oils from different plants.

Krishna, M. S., et al. (2012). Phytochemical screening and antimicrobial activity of medicinal plants.

Aparna et al. (2012) Anti-Inflammatory Property of n-Hexadecanoic Acid: Structural Evidence and Kinetic Assessment

Al-Muhe, R. M., & Abdul-Wahid, M. S. (2023). Effect of Spraying with Moringa Leaf Extract and Potassium Silicate on some Chemical Properties of Banana Plant *MUSA* spp. *IOP Conference Series: Earth and Environmental Science*, 1262(4). <https://doi.org/10.1088/1755-1315/1262/4/042021>

Al-Muhe, R. M., & Abdul-Wahid, M. S. (2024). Study of the Protein Patterns under the Effect of Spraying with Moringa Plant Extract and Potassium Silicate for Banana Obtained from Tissue Culture *Musa* spp. *Journal of Global Innovations in Agricultural Sciences*, 12(1), 155–160. <https://doi.org/10.22194/JGIAS/12.1126>

Mahewish, Hamid H., Falah H. Radi, and Mohammed N. Radhi. "Response of rosemary plant to the effect of Nano-NPK fertilizer and biological factors and their effect on the active substances." *University of Thi-Qar Journal of agricultural research* 10.1 (2021): 39-48.