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Mini Review on Antimicrobial Activity and Bioactive Compounds of *Moringa oleifera*

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Abstract

Moringa oleifera is a kind of woody tree traditionally used as a nutritional source and as a medicinal plant. It grows wild in the tropical and subtropical areas of Asia, Africa and the Middle East. In China, *Moringa oleifera* trees are planted at a large scale in Yunnan, Guangdong and Guangxi Provinces. As a nutritional and medicinal plant, *Moringa oleifera* is a rich source of bioactive compounds with diverse pharmacological activities. It has been widely used in the treatment of certain diseases as a traditional medicinal herb. Antimicrobial activity is the most studied property of *Moringa oleifera*. Many studies have shown that the leaf, flower, bark, root, seed, and nearly all types of *Moringa oleifera* tissues exhibit antimicrobial activity including antibacterial, antifungal, antiviral and antiparasitic activity. This review describes progress on research conducted to understand the antimicrobial activity and related bioactive properties of *Moringa oleifera* compounds, and discusses the potential use of *Moringa oleifera* in the control of pathogenic microbes.

Keywords: *Moringa oleifera*; Antimicrobial activity; Chemical component; Antibacterial activity; Antifungal activity; Antiviral activity; Antiparasitic activity

Introduction

Discovery of novel antimicrobial agents is very important for the control of pathogenic microbes, especially for the treatment of infections caused by resistant microbes. Medicinal herbs with antimicrobial activities are considered a potent source of novel antimicrobial function. *Moringa oleifera* is a woody tree mainly distributed in the tropical and subtropical regionsof Asia, Africa and the Middle East [1]. In China, *Moringa oleifera* was planted at a large scale in Yunnan, Guangxi and Guangzhou provinces about twenty years ago. It is widely used as a vegetable, functional food and medicinal plant that has rich nutritional composition with diverse pharmacological activities [2-4].

Previous studies have shown the chemical composition of *Moringa oleifera* and its suite of bioactive compounds to include vitamins, carotenoids, polyphenols, flavonoids, essential amino acids, and phenolic acids [5-7]. *Moringa oleifera* has traditionally been used in the treatment of malaria, parasitic diseases, skin diseases, hypertension and diabetes. It has been demonstrated that *Moringa oleifera* exhibits pharmacological properties such as antioxidant [8], anti-inflammatory, anti-cancer, anti-hyperglycemic and anti-hyperlipidemic properties [2]. Leone et al. have reviewed the cultivation, genetics, ethnopharmacology, phytochemistry and pharmacology of *Moringa oleifera* [1], whereas the health benefits and medicinal use of *Moringa oleifera* has been reviewed by Abdull Razis et al. [2] and Anwar et al. [3]. Stohs et al. found that aqueous, hydroalcohol or alcohol extracts of *Moringa oleifera* exhibited various biological activities including antioxidant, tissue protective, and analgesic properties, whereas the leaf extracts were shown to be safe to use based on safety studies in animals [9].

Antimicrobial activityhas been extensively studied in *Moringa oleifera*. Crude extracts of different tissues of *Moringa oleifera* display different antimicrobial activities. Moreover, several molecules with antimicrobial activities have been identified from different tissues of *Moringa oleifera*. In this review, we summarize the research progress in understanding and characterizing the antimicrobial activity of *Moringa oleifera* tissues including antibacterial, antifungal, antiviral, and antiparasitic activity, and discuss the potential use of *Moringa oleifera* in the control of pathogenic microbes.

Antibacterial Activities of *Moringa oleifera*

Many studies have suggested that different crude extracts from different tissues of *Moringa oleifera* show antibacterial activities against both Gram-negative and Gram-positive bacteria.

Using the zone of inhibition test, Pal et al. [10] evaluated the antimicrobial activity of *Moringa oleifera* leaf extracts. Four Gram-positive bacteria (*Bacillus cereus, Bacillus subtilis, Staphylococcus aureus and Sarcinalutea*) and two Gram-negative bacteria (*Escherichia coli and Acid fast Mycobacterium phlei*) were used to test the antibacterial activities of *Moringa oleifera*. The results revealed that the average zone of four Gram-positive bacteria and two Gram-negative bacteria was inhibited by the ethanol extract of *Moringa oleifera* leaves. Using modified disk diffusion to screen antibacterial activity, Peixoto et al. [11] found that aqueous ethanol extracts of *Moringa oleifera* leaves showed effective antibacterial activity against *Staphylococusaureus*, *Vibrio parahaemolyticus*, *Enterococcus faecalis* and *Aeromonas caviae,* whereas no effects were seen against *Escherichia coli*, *Pseudomonas aeruginosa* and *Salmonella enteritidis*. The experiments carried out by Daotam et al. [12] confirmed that the methanol extract of *Moringa oleifera* leaves showed different inhibition patterns against 13 different bacteria strains out of the 19 Gram-negative bacteria strains tested, including *Escherichia coli, Enterobacter aerogenes, Klebsiella pneumonia, Pseudomonas aeruginosa* and *Providencia stuartii*. Further

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chemical composition analysis revealed that the *Moringa oleifera* leaf extract with antibacterial activities contains alkaloids, polyphenols, flavonoids, anthraquinones, coumarins, tannins, triterpenes, sterols saponins and some secondary metabolites. It was suggested that *Moringa oleifera* leaf extracts might be used in the control of many infectious diseases alone or together with other antibiotics [12].

In addition to the leaves, crude extracts of the bark of *Moringa oleifera* also possess antibacterial activities. Zaffer et al. reported that methanol, chloroform, ethyl acetate and aqueous bark extracts show different degrees of antibacterial activity against four bacterial species, *Staphylococcus aureus, Citrobacter freundii, Bacillus megaterium and Pseudomonas fluoescens* [13]. Of these, the ethyl acetate extract exhibited the maximum antibacterial activity against the four tested bacteria strains and the most sensitive speciesto all four different extracts was *Staphylococcus aureus*.

The methanol crude extract of *Moringa oleifera* seeds exhibited clear antibacterial activity using thin layer chromatography bioassay against *Escherichia coli, Pseudomonas aeruginosa, Staphylococcus aureus, Cladosporium cladosporioides and Penicillium sclerotigenum* [14]. Three compounds, 4-(α-L-rhamnopyranosyloxy)benzyl isothiocyanate, methyl *N*-4-(α-L-rhamnopyranosyloxy)benzyl carbamate, and 4-(β-D-glucopyranosyl-1→4-α-L-rhamnopyranosyloxy)-benzyl thiocarboxamide were identified from the extract of *Moringa oleifera* seed. These compounds show potent antibacterial activity against some of the tested pathogens at 5 mg/L. Among them, 4-(β-D-glucopyranosyl-1→4-α-L-rhamnopyranosyloxy)-benzyl thiocarboxamide showed the most potent antibacterial activity again *Shigella dysenteriae*, *Bacillus cereus*, *Escherichia coli* and *Salmonella typhi* [14]. The synergistic 4-(α-L-rhamnopyranosyloxy) benzyl isothiocyanate, which was originally isolated from the methanol extract of *Moringa oleifera* seed, showed more effective antimicrobial activity against 3 bacterial species (*Staphylococcus aureus, Escherichia coli* and *Pseudomonas aeruginosa*) and 2 fungal species (*Candida albicans* and *Aspergillus niger*) than niazimicin, which was also isolated from the seed of *Moringa oleifera* [15]. *Staphylococcus aureus* is a major human pathogen that is wide spread in hospitals [16]. The compound 4-(α-L-rhamnopyranosyloxy) benzyl isothiocyanate was found to inhibit the growth of BAA-977, a *Staphylococcus aureus* strain [17]. It appears that *Moringa oleifera* has a promising future in the control of infectious pathogenic bacteria and can be used in the discovery of novel antibacterial agents.

In addition to the various chemical compounds that exhibit antibacterial activity, several antimicrobial peptides have also been identified and characterized from *Moringa oleifera*. Lectin from *Moringa oleifera* was originally isolated from the seed aqueous extract by chitin column chromatography as a larvicidal agent again *Aedes ageypti* [18]. The water-soluble *Moringa oleifera* lectin shows effective antibacterial activity by reducing the growth of *Staphylococcus aureus* in polluted waters [19]. Moura et al. [20] revealed that the water-soluble *Moringa oleifera* lectin show bactericidal effects against *Bacillus cereus, Bacillus pumillus, Bacillus megaterium, Micrococcus sp., Pseudomonas sp., Pseudomonas fluorescens, Pseudomonas stutzeriand Serratia marcescens* by affecting the growth, survival and cell permeability of these bacteria. Furthermore, some of the bacteria tended to undergo biocorrosion following lectin treatment.

The flocculating cationic polypeptide is another antimicrobial peptide isolated from the seed of *Moringa oleifera* and also referred as Flo or MOCP [21-23]. The flocculation cationic polypeptide showed antibacterial activity by damaging bacterial cell membranes and causing fusion between membranes. Also, it has been used widely in water purification processes [24]. Structure-function studies of Flo peptides revealed that a sequence prone to form a helix-loop-helix structural motif is important to the bactericidal activity and the hydrophobic proline residues within the protruding loop are essential for maintaining bactericidal activity [25].

Besides the bioactive compounds and antimicrobial peptides, silver nanoparticles were also shown to be involved in antibacterial activity. Prasad and Elumalai [26] showed that the extract of *Moringa oleifera* leaves exhibits considerable antimicrobial activity by producing silver nanoparticles extracellularly via the rapid reduction of silver ions.

Moringa oleifera was also shown to inhibit pathogenic bacteriathatcan affect marine aquaculture products [27]. Brihanteet al. [28] showed that the ethanol extract of *Moringa oleifera* pods and chloroform extract of flowers could effectively inhibit three strains of *Vibrio cholera*, *Vibrio vulnificus* and *Vibrio mimicus* isolated from *Macrobrchium amazonicum* prawns. However, the ethanol extract of stem and seeds showed low efficiency in inhibiting the growth of these pathogenic bacteria.

Taken together, results from the various studies described in this section suggest that the identification of several bioactive molecules might contribute to the discovery of novel antibacterial agents from the medicinal plant *Moringa oleifera*.

Antifungal Activity of *Moringa oleifera*

Antifungal activity of *Moringa oleifera* has been described in multiple studies. Chuang showed the antifungal effect of *Moringa oleifera* against the human pathogenic fungi *Trichophyton rubrum*, *Trichophyton mentagrophytes*, *Epidermophyton floccosum* and *Microsporum canis*. The results showed that both the seed extracts and essential oils exhibited antifungal activity on the four different fungi with different MICs while the leaf extract and the sub-fractions had little effect on these dermatophytes [29]. This study suggests that *Moringa oleifera* can be used to treat fungal infections of skin.

Saprophytic fungi were also found to be inhibited by the extract of *Moringa oleifera* leaves [30]. This antifungal property of *Moringa oleifera* can prevent culture media contaminations by some saprophytic fungi.

Phytopathogenic fungi cause disease and threaten the production of economic crops. Several reports suggest that *Moringa oleifera* show the ability to control fungal disease in plants. A thermo-stable protein named chitin-binding protein (Mo-CBP3) was isolated by affinity chromatography on chitin together with ion exchange chromatography from the seeds of *Moringa oleifera*. Mo-CBP3 shows *in vitro* antifungal activity on the phytopathogenic fungi *Fusarium solani, Fusarium oxysporum, Colletotrichum musae and Colletotrichum gloesporioides* at a concentration of 0.05 mg/ml with 62% inhibition within 49 hour [31], but has no effect on *Pythium oligandrum* [32]. Further studies revealed that Mo-CBP3 exhibits antifungal activity by inhibiting mycelia growth and conidial viability of *Fusarium solani*. It is suggested that, through interactions with the cell membrane, Mo-CBP3 induced the production of ROS and caused cell death in the fungi [31]. Till now, 4 different Mo-CBP3 isoforms (Mo-CBP3-1, Mo-CBP3-2, Mo-CBP3-3, Mo-CBP3-4) have been identified from *Moringa oleifera* seed. By analyzing the full-length cDNAs encoding Mo-CBP3, it is predicted that the precursor of different Mo-CBP3 isoforms consist of 160 and 163 amino acids with a 20 residue signal peptide. Mo-CBP3 was previously regarded as a member of 2S alumbin family [33]. Now, Mo-CBP3 is considered as a new antifungal drug in the development of transgenic crops for traits

such as thermo-stability, broad antifungal spectrum and low toxicity [33,34].

Four secondary metabolites were isolated from the endophytic fungus of *Moringa oleifera* by Zhao et al. [35] and the chemical structures were determined as griseofulvin, dechlorogriseofulvin, 8-dihydroramulosin and mellein. Among them, griseofulvin, which was indirectly isolated from *Moringa oleifera,* showed clear inhibition on the growth of 8 plant pathogenic fungi in an *in vitro* antifungal assay [35]. This study provides a new strategy for the discovery of novel antimicrobial agents from *Moringa oleifera*.

Antiviral Activity of *Moringa oleifera*

As a traditional medicinal plant, *Moringa oleifera* show antiviral activities in several different studies. Murakami et al. [36] reported that Epstein-Barr virus (EBV) activation in Raji cells could be inhibited by niaziminin, a thiocarbamate identified from the leaves of *Moringa oleifera*. The acetoxy group at the 4'-positon of niaziminin plays an important role in maintaining antiviral activity. Lipilun et al. [37] showed that the ethanol extract of *Moringa oleifera* exhibits anti herpes simplex virus (HSV) type activity at a dose of 750 mg/kg per day. Treatment with *Moringa oleifera* extract could delay skin lesion development, prolong the mean survival times, and reduce the mortality of HSV-1 infected mice [37,38]. Waiaput [39] suggested that 80% ethanol crude extracts of *Moringa oleifera* fruit showed anti-HBV activity by inhibiting HBV replication with mild cytotoxicity on HepG2 cells [39]. *Moringa oleifera* has also traditionally been used in the treatment of HIV/AIDS related symptoms, possibly by improving the immune system. However, very few reports reveal the anti-HIV activity mechanism of *Moringa oleifera,* and there is no evidence to confirm the effect of *Moringa oleifera* on HIV.

By reviewing the studies discussed in this section, we can conclude that *Moringa oleifera* can be used in the development of promising antiviral drugs.

Antiparasitic Activity of *Moringa oleifera*

Leishmania is a genus of trypanosomes that are responsible for Leishmaniasis disease [40]. Kaur et al. [41] showed that 70% ethanolic extract of *Moringa oleifera* roots exhibit antileishmanial activity with IC_{50} values of 83 µg/ml and the methanolic extract of leaves showed antileishmanial activity with IC_{50} values of 47.5 µg/ml. Niazinin, which was isolated from the methanolic extract of leaves, showed the most antileishmanial activity with IC_{50} values of 5.25 µM. Singh et al. [42] showed that the extract of *Moringa oleifera* flower (MoF) showed the most antileishmanial activity compared to extracts from other tissues. The ethyl acetate fraction of MoF, named MoE showed the most antileishmanial activity both *in vivo* and *in vitro*.

Schistosomiasis is a chronic tropical disease caused by parasite *Schistosoma mansoni* and about 200,000 people are killed each year by this disease [43]. *Biomphalaria glabrata* is the intermediate host of *Schistosoma mansoni* [44]. The control of *Schistosoma mansoni* and *Biomphalari aglabrata* is critical in preventing the spread and infection of schistosomiasis. The aqueous extract of *Moringa oleifera* flowers, which contain tannins, saponins, flavones, flavonols, xanthones and trypsin, were found todelay the development of *Biomphalari aglabrata* embryos and promoted the mortality of adult snails significantly [45].

Aedes aegypti females transmit the virus that causes dengue and arboviral diseases commonly occurring in tropical and subtropical regions. Several studies have reported that the extract of *Moringa oleifera* flowers with trypsin inhibitor activity show larvicidal activity on *Aedes ageypti* [46,47]. Agra-Neto et al. confirmed that lectins WSMoL and cMoL from *Moringa oleifera* seeds show larvicidal activity on fourth-stage *Aedes ageypti* larvae by affecting the digestive and detoxifying related enzymes of larvae [18,48]. These studies suggest that *Moringa oleifera* could be used in mosquito control to disrupt the spread of related diseases.

In many developing countries including Ghana, irrigation water is widely used in urban farming, which can get polluted by the helminth parasite and their eggs. Senguptaet et al. showed that *Moringa oleifera* seed extracts could reduce helminth eggs and turbidity in irrigation water [49]. It is well known that *Moringa oleifera* seeds have been widely used in water purification for a long time in underdeveloped countries. The mechanism of *Moringa oleifera* seeds as water purifying agents was shown by the flocculent protein from *Moringa oleifera* seed forming flocculate particles in suspension in water [22,50]. The ability to reduce helminth eggs is one of the functions of *Moringa oleifera* seed extracts in the water purifying process.

Haemonchus contortus is the main nematode species in the gastrointestinal tract of small ruminants [51]. A previous study reported that the crude water extract of *Moringa oleifera* seed showed high egg hatch inhibition on *Haemonchus contortus* [52]. The results of this work describe a potential use of *Moringa oleifera* in controlling nematode parasites [53]. On the whole, *Moringa oleifera* shows high potential in the control of parasites, which could inhibit the spread of related diseases.

Future Applications

Moringa oleifera can produce a large variety of novel bioactive compounds and has been widely used in food and treatment to certain diseases through traditional medicine. Several studies have shown that extracts and some identified compounds from *Moringa oleifera* exhibit antimicrobial activity against various pathogenic bacteria, fungi, viruses and parasites that affect the health of humans, animals and crops. The antimicrobial activities of different tissues with different bioactive compounds are summarized in Table 1. Nearly all the tissues of Moringaoleifera show different antimicrobial activities. Considering their broad antimicrobial spectrum, low toxicity as a traditional food and high production for growing fast, *Moringa oleifera* can be effectively used in the discovery of novel antimicrobial agents. It can be used in medicinal treatments to control the infection of pathogenic microbes. In agriculture, *Moringa oleifera* can improve the control of phytopathogenic fungi that cause disease and affect the production of economic crops. In marine aquaculture, *Moringa oleifera* also shows a promising future in the control of infectious diseases.

For developing novel agents, it is important to promote the isolation and identification of certain molecules with antimicrobial activity from *Moringa oleifera*. Evaluation of the safety and toxicity of antimicrobial agents form *Moringa oleifera* is necessary before implementing the use of these compounds. The detailed antimicrobial molecular mechanism of compounds form *Moringa oleifera* is also important to determine. In brief, this review provides valuable information on *Moringa oleifera* antimicrobial activity and its potential applications.

Conclusion

In conclusion, the leaves, bark, root, flower, fruit and seed of Moringaoleifera are all reported with antimicrobial activities against different microorganisms. The leaves show antibacterial, anti- saprophytic fungi, anti-HSV, anti-EBV and antileishmanial

Tissues	Compounds	Activities	References
Leaves	Ethanol extract	Antibacterial activity	[10-12]
		Antisaprophytic fungi activity	[30]
		Anti-HSV activity	[37]
	Methanol extract	Antileishmanial activity (IC50=47.5 ug/ml)	[41]
	Niaziminin	Inhibit the activation of EBV in Raji cells	[36]
	Niazinin	Antileishmanial activity (IC50=1.8ug/ml)	[41]
	Silver nanoparticles	Antibacterial activity	[26]
Bark	Methanol, chloroform, ethyl acetate and aqueous bark extracts	Antibacterial activity against four bacterial species	[13]
Root	70% ethanol extract	Antileishmanial activity (IC50=83 ug/ml)	[41]
Flower	Aqueous extract (MoF) and ethyl acetate fraction (MoE)	Antileishmanial activity	[42][45]
	Trypsin inhibitor	Larvicidal activity on *Aedesageypti*	[46-47]
Fruit	80% ethanol crude extracts	Anti-HBV activity	[39]
Seed	Methanol crude	Antibacterial activity	[14]
	Extract and essential oil	Against four human pathogenic fungi	[29]
	Seed extract	Reduce helminth eggs in irrigation water	[49]
	N-4-(α-L-rhamnopyranosyloxy)benzyl carbamate	Antibacterial activity	[14]
	4-(β-D-glucopyranosyl-1→4-α-L-rhamnopyranosyloxy)-benzyl thiocarboxamide	Antibacterial activity	[14] [15]
	4-(α-L-rhamnopyranosyloxy)benzyl isothiocyanate,	Antibacterial activity	[17]
	Lectin	Antibacterial activity	[18-20]
	Lectins WSMoL and cMoL	Larvicidal activity on fourth-stage *Aedesageypti* larva	[18][48]
	Flo	Antibacterial activity	[21-25]
	Mo-CBP3	Antifungal activity on 4 phytopathogenic fungi	[31-34]
Endophytic fungus	Secondary metabolite, griseofulvin	Antifungal activity on 8 plant pathogenic fungi	[35]

Table 1: The antimicrobial activities of different tissues with different bioactive compounds from *Moringaoleifera*.

activities. The bark show antibacterial activity against four bacterial species and the root show antileishmanial activity. The flower exhibit antileishmanial activity and larvicidal activity on Aedesageypti. The fruit own anti-HBV activity. The seed display antimicrobial activities against some bacteria, human pathogenic fungi, phytopathogenic fungi, and parasits. Niaziminin and Niazinin are two compounds with antimicrobial activity identified from the leaves. N-4-(α-L-rhamnopyranosyloxy)benzyl carbamate, 4-(β-D-glucopyranosyl-1→4-α-L- rhamnopyranosyloxy)-benzyl thiocarboxamid, 4-(α-L-rhamnopyranosyloxy)benzyl isothiocyanate, Lectin, Flo and Mo-CBP3 are identified from the seed, and show different antimicrobial activities. It was suggested that the leaves and the seeds display broader antimicrobial spectrum than other tissues of Moringaoleifera.

Conflict of Interest Statement

The authors declare that they have no conflicts of interest to this work.

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