

Antimicrobial efficacy of moringa seed extracts against some postharvest disease pathogens of tomato

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ABSTRACT

Moringa tree has an impressive range of medicinal uses and high nutritional value. The present study deals with the antimicrobial activities of the ethanolic, petroleum ether and aqueous extracts of moringa seeds in controlling some bacteria and fungi and improving postharvest handling of tomato fruits. A sample of 100g of moringa seed powder was extracted successively with petroleum ether and also with ethanol using a Soxhlet apparatus for about four hours with petroleum ether and eight hours with ethanol. Solvents were then evaporated; extracts were left till complete drying. The petroleum ether extract (oil) produced 34.9 ml and the ethanol extract 14.5 g. Various concentrations of seed extracts of moringa (10%, 20%, 30% and 50%) were prepared and evaluated, using a randomized complete block design, for their antimicrobial efficacy against five species of fungi, namely (*Natrassia mangifera*, *Fusarium oxysporum*, *Aspergillus niger*, *Alternaria alternata* and *Metarhizium anisoplae*) and three species of bacteria (*Bacillus subtilis*, *Actinomycetes* sp. and *Erwinia caratovora*). The ethanolic extract showed a strong antibacterial activity against *Actinomycetes* sp. and fungicidal activity against *F. oxysporum*. The aqueous extract presented strong antibacterial activity against *B. subtilis* and strong antifungal activity against *A. niger*, but the ethanol extract did not show any activity against *A. niger* and *M. anisoplae*. All seed extracts, irrespective of their types, in different concentrations, inhibited the growth of all microbes to varying degrees except petroleum ether extract. The inhibition zone increased gradually with the increase of extract concentration. Tomato fruits were treated with moringa aqueous extract coatings with wax in order to preserve quality and enhance shelf life during storage. Weight loss of uncoated and coated samples was determined throughout storage at 10°C and 37°C for 10 days. The four experimental coatings were wax, wax mixed with 10 ml of crude aqueous extract of moringa, crude extract only and untreated tomatoes. *Moringa* extract was effective in extending the shelf-life of treated tomato fruits when compared to the untreated, in the following order: Wax, moringa extract and untreated control.

INTRODUCTION

The Moringaceae is a mono- generic family with 13 known species; the most popular is *Moringa oleifera* Lam, which is grown throughout tropical and sub-tropical region. For centuries, moringa has been advocated for traditional medicinal and industrial uses (Nepolean, *et al.*, 2009). All parts of moringa tree are edible and have long been consumed by humans. Various parts of the moringa tree such as root, bark, leaves, flowers, pods and seeds are used in nutrition, water purification, cattle fodder and as a fertilizer (Palada and Chang, 2003).

The plant has many antimicrobial properties as secondary metabolites such as alkaloids, and phenolic compounds. moringa seed oil yield is 30-40 % by weight. It is a sweet, non sticking, non drying oil that resists rancidity (Nepolean, *et al.*, 2009). This versatile property of moringa has recently attracted the attention of many scientists and encouraged them to screen its biological activities (Kekuda *et al.*, 2010).

Several low molecular weight proteins or peptides with antibacterial or antifungal activity have been isolated in recent years from various plants and are believed to be involved in defense mechanism against phytopathogenic microorganisms by inhibiting their growth through diverse molecular modes. The plants can also produce localized production of antimicrobial low molecular weight secondary metabolites known as phytoalexins (Dahot 1998). Edible coatings may provide additional protection against contamination of microorganisms.

The aim of the present work was to provide information about the antimicrobial activity of moringa seed extracts against selected microorganisms and the efficacy of the above named extracts in controlling selected postharvest diseases and extending the shelf-life of tomato fruits.

MATERIALS AND METHODS

Preparation of extracts

Moringa seeds were collected from a single tree in Wad Madani city in 2013 and used for crude extraction. The organic solvent extract was prepared according to the method adopted by Saadabi and Abu zaid, (2011). An amount of 186 g of moringa seeds were sun dried, the testa and wings were manually removed and the white kernels were ground to fine powder using a coffee mill, to produce about 136 g kernel weight. A sample of 100 g of the powder was extracted successively with petroleum ether and ethanol using a Soxhlet apparatus for about four hours with petroleum ether and eight hours with ethanol. Solvents were then evaporated and extracts were air dried. The petroleum ether extract produced 34.9 ml of oil and the ethanolic extract 14.5 g. To make the organic solvent extracts, 10g, 20g, 30g and 50g of the ethanol extract were mixed with 100 ml distilled water to make a 10%, 20%, 30% and 50% concentrations. The petroleum ether extract was dissolved in 10 ml hexane and similar concentrations to the ethanolic extract were prepared. The aqueous

extract from moringa seeds was prepared for the bioassay. An amount of 100g of crushed seeds was added to 500 ml distilled water in a water bath for 15 minutes and filtered. The extract was dried and then kept in a freezer. Concentrations of 10%, 20%, 30% and 50% were prepared, then 100 μ L of each was added to the plate with the microbial strain.

The aqueous extract for postharvest disease control was prepared according to the method of Adetunji *et al.*, (2012). Moringa seeds were dried in the laboratory and ground to a very fine powder and sieved using muslin cloth. Fifty grams of the dried moringa seed powder was weighed and introduced into a conical flask containing 250 ml of distilled water, covered with aluminum foil and placed on a water bath and then filtered with Whatman filter paper. The filtrate obtained was evaporated to dryness.

Microbial cultures and growth conditions

Moringa seed ethanolic extracts were assayed for antifungal activity against the fungi *Natrassia mangifera*, *Fusarium oxysporum*, *Aspergillus niger*, *Alternaria alternata* and *Metarhizium anisoplae*. The antibacterial activity was assayed against *Bacillus subtilis*, *Actinomyces* sp. and *Erwinia amylovora*. All the microbes were collected from diseased plant tissue, isolated and identified at the Department of Plant Pathology, Faculty of Agriculture and Natural Resources, Gezira University in 2013. The fungi were grown on Potato Dextrose Agar (PDA) plates while the bacteria were cultured on Nutrient Agar (NA) at 32°C and maintained with periodic sub-culturing in a refrigerator at 4°C.

Antifungal activity

The extracts of moringa seeds were screened for antifungal activity by the agar well diffusion method (Perez *et al.*, 1990) with a sterile cork borer of size 6.0 mm. The 48 hours old cultures grown in PDA plates were prepared. After solidification, the appropriate wells were made on the agar plates using a cork borer. In the agar well diffusion method, 100 μ l of ethanolic and petroleum ether extracts (concentrations: 10%, 20%, 30% and 50%) were introduced into the wells. An incubation period of 24-48 hours at about 37 °C was allowed. The antifungal activity was evaluated by measuring the zones of inhibition of fungal growth which was put in the well in the side of the plate and the seed extract in the other side. For each concentration of the extracts, three replicate trials were conducted against the tested organisms. Filter paper discs dipped into sterile distilled water and hexane were used as control.

Antibacterial activity

The antibacterial activity of moringa seed extracts at 10%, 20%, 30% and 50% concentrations was conducted on three bacterial species which normally cause plant diseases according to the method of Saadabai and Abu zaid, (2011). Impregnated seed extract filter paper discs by each concentration were put in the center after the bacteria was streaked in the nutrient agar plate. Antibacterial activity was determined by measuring the

zone of inhibition around each paper disc. For each concentration of the extracts, three replicate trials were conducted against the tested organisms. Filter paper discs dipped into sterile distilled water and hexane were used as control. The plates were then incubated at 32°C for 48 hours.

Antimicrobial activity of aqueous extract

The aqueous extracts used in the different experiments were used to test their activity on three fungi (*Natrassia mangifera*, *Fusarium oxysporum* and *Aspergillus niger*) and one bacterium, *Bacillus subtilis*.

Standard antibiotic and antifungal assay

A sample of 100 µl concentration of 10% aqueous extract and 10% ethanolic extract were tested against *Bacillus subtilis* compared with the reference standard antibiotic Chloramphenicol (25µl/ 3g/100 ml concentration) as an antibiotic. The effect against *Fusarium oxysporum* was evaluated with Thiram fungicide (25 µl/3g/100 ml concentration).

Tomato postharvest disease control

Tomato fruits were brought from Wad Medani market. Bee wax was obtained from Gezira University honey bees project. The fruits were sorted out to remove diseased and bruised ones, washed by tap water and dried. The clean dried fruits were then divided into 3 lots, each containing 15 fruits and then subjected to the different coating treatments. Each treatment was replicated three times.

An amount of 5.5g of bee wax was taken in 250 ml conical flask and melted at 70°C and heated continuously to 80-90°C. Then 2.5g oleic acid was added to the melted wax followed by the addition of 1.5 g triethanolamine with constant stirring. Finally 90.5 ml of distilled water (which was pre-heated at the same temperature (80-90°C) was added slowly with continued stirring for 5-7 minutes. The emulsion was cooled down and stored at room temperature in a stoppered vessel (Salman *et al.*, 2008).

Four treatments were applied to the tomato fruits: Coated with bee wax +10 ml moringa aqueous seed extract, coated with 10 ml moringa aqueous seed extract, coated with 10 ml Bee wax only, uncoated fruits (control). Half the lots of fruits were coated with the above concentrations and stored at room temperature (about 37°C) and the other half with the same coating treatments were stored at 10°C. Data were recorded from randomly selected fruits at 4 and 10 days intervals during the experimental period. For the determination of weight loss during storage, five fruits were marked at the start of the experiment from each treatment and kept separate for periodic weighing by an electric digital balance (model SF-400). The number of fruits with symptoms of rot, due to the fungi *Aspregillus niger* and *Fusarium oxysporum*, were recorded and the results were expressed as disease severity and percentage.

Experimental design and statistical analysis

The experiments were arranged in a completely randomized design. The data were subjected to analysis of variance. Treatment means were separated using Duncan's Multiple Range Test at 5% level of significance.

RESULTS AND DISCUSSION

Antifungal activity of ethanol extract

As a general rule, moringa seed extracts were considered active against both fungi and bacteria when the zone of inhibition in the culture medium is greater than 6 mm (Saadabi and Abu zaid 2011; Eilert *et al.*, 1981). Accordingly, in this study, moringa ethanol extract showed an antifungal activity against *N. mangifera*, *F. oxysporum* and *A. alternata* and has antibacterial activity against *B. subtilis*, *E. caratovora* and *Actinomycetes* sp. (Tables 1 and 2). Results obtained from *in vitro* antimicrobial activity showed no inhibition against the fungal species *A. niger* and *M. anisoplae*. (Table 1). The ethanolic extract was superior to petroleum ether extract, which had no activity against all tested fungi in all concentrations (data not shown). Similarly, Bobbaral *et al.* (2009) found that moringa ethanolic extract at 0.05 ml concentration did not exhibit antifungal activity against *A. niger*. Working with the methanolic seed extracts of moringa, Saadabi and Abu zaid (2011), observed little or no activity against *A. niger*. According to Chiejina and Onaebi (2013), the ethanolic leaf extract of moringa showed fungicidal activities against *A. niger*, especially at high concentrations. In this study, only seed extracts were used. Looking closely at these results, it is clear that all the concentrations of the ethanolic extract significantly suppressed the growth of *N. mangifera* (Table 1). The inhibition zone increased significantly with increasing concentrations, reaching 13.00 mm when 50% concentration was used.

Also, all the ethanolic extract concentrations inhibited the growth of *F. oxysprum*. The addition of either 20% or 30% of the extract did not differ from each other in reducing *F. oxysporum* growth, however, they were significantly different from both 10% and 50% concentrations (Table1). *A. alternata* showed increased inhibition zone diameters as the ethanol extract concentration was increased from 10% to 50%. A concentration of 50%, however, significantly controlled the fungus compared to all other concentrations. The results are also in line with the findings of Jabeen, *et al.* (2008), who found that crude extracts of moringa seed showed very strong activity against *Fusarium solani*, and little activity against *A. niger* and *M. anisoplae*.

The sensitivity of *N. mangifera*, *F. oxysporum* and *A. alternata* to a concentration of 10% of the ethanolic extract was similar (Table 1). As the concentration was increased to 20%, *F. oxysporum* and *A. alternata* became more affected than *N. mangifera* by the application

of the extract. At the higher concentrations of 30% and 50%, *F. oxysporum* was the most sensitive of all the tested fungi which was exhibited by the diameter of the inhibition zone.

Table 1. Antifungal activity of ethanol extract of moringa seeds against five fungal species expressed as inhibition zone diameter (mm).

Fungi	Extract concentrations (%)			
	1	2	30	5
	0	0		0
<i>Natrassia mangifera</i>	7	9	12.	1
	.	.	7	3
	0	3	^b _y	.
	d	c		0
	x	y		a
	y			y
<i>Aspergillus niger</i>	0	0	0.0	0
	.	.	0.0	.
	0	0	^a _z	0
	^a _z	a		a
		z		z
<i>Fusarium oxysporum</i>	7		14.	2
	.	1	0	2
	3	1	^b _x	.
	c	.		0
	x	7		a
		b		x
		x		
<i>Alternaria alternata</i>	8		13.	1
	.	1	0	8
	7	1	^b _y	.
	c	.		7
	x	7		a
		b		y
		c		
		x		

<i>Metarhizium</i>	0		0.0	
<i>anisoplae</i>	.	0	^a _z	0
	0	.		.
	^a _z	0		0
		a		a
		z		z

Means having the same letters in the rows (a-d) or columns (x-z) are not significantly different according to Duncan's Multiple Range Test at 5% level of significance.

Antibacterial activity of ethanol extract

In this experiment, the ethanol extract of moringa was found to be inhibitory to all bacterial species studied except *B. subtilis* in low concentrations of 10% and 20% (Table 2). The inhibition zones increased significantly with the increased concentration of the applied extract. The growth of *Actenomyces* sp. was the most retarded by the ethanolic extract of moringa. This was followed by *E. caratovora*, which is a very important causal organism of postharvest diseases. It came second in sensitivity to the extract. This justifies the trials in this study of moringa extract as a postharvest disease control. In case of *B. subtilis*, no inhibition was evident at low concentrations. Nevertheless, when using higher concentrations, the inhibition zone diameters significantly increased (Table 2).

Results obtained from *in vitro* antibacterial activity showed that the ethanol extract was superior to petroleum ether extract. The latter had no activity against all bacterial species and in all tested concentrations in suppressing the bacterial growth (data on petroleum ether is not shown). The antimicrobial activity of the extracts may be due to the presence of lipophilic compounds (Saadabi and Abu zaid, 2011). The inactivity of petroleum ether seed extract may be due to the fact that the active compounds which possess the antimicrobial properties are polar in nature and not possibly extracted by petroleum ether (Saadabi and Abu zaid, 2011). The results are in contrast with the findings of Jabeen *et al.* (2008) who found that the crude ethanol extracts of moringa seed showed very strong activity against *Bacillus subtilis*. Here, *Actenomyces* sp. was the most sensitive to most concentrations.

Saadabi and Abu zaid (2011) evaluated moringa seed extracts against four bacteria, including *B. subtilis*. All of the seed extracts, irrespective of their types, in different concentrations inhibited the growth of all microbes to varying degrees. The maximum inhibition zone was observed against *Staphylococcus aureus* followed by *B. subtilis*. This confirms the results of the present study where 30% and 50% concentration inhibited the growth of *B. subtilis*. Similarly, Uma and Sasikumar (2005) found that different organic and alcoholic extracts of moringa had antimicrobial activity against certain bacterial pathogens including *B. subtilis*. Walter *et al.* (2011) showed that an antibacterial activity of

methanol and n-hexane extracts of moringa seeds were used on three bacterial species, which normally cause water borne diseases. The highest inhibitions were observed at lower dilutions of moringa methanol extracts. This disagrees with the present finding where the effect increases with increasing concentration. Nepolean *et al.* (2009) found that the ethanolic extracts of moringa demonstrated the highest activity, while the aqueous extracts showed the least activity against *Salmonella typhi*.

Table 2. Antibacterial activity of ethanol extract from moringa seeds against different species of bacteria expressed as inhibition zone diameter (mm).

Bacteria	Extract concentrations (%)			
	10	2	3	5
		0	0	0
				%
<i>Bacillus subtilis</i>	0.0 ^{c_z}	0 .	6 .	9 .
		0 ^{c_z}	7 ^{b_y}	7 a
				z
<i>Actenomyces</i> sp.	6.0 ^{c_x}	1 7	2 2	3 0
		.	.	a
		7 ^{c_x}	3 ^{b_x}	x
<i>Erwinia caratovora</i>	6.7 ^{d_y}		1	1
		1	.	.
		.	0	3
		0 ^{c_y}	^{b_x}	a
				y

Means having the same letters in the rows (a-d) or columns (x-z) are not significantly different according to Duncan's Multiple Range Test at 5% level of significance.

Antifungal activity of aqueous extract

In this experiment, the aqueous extract showed significant antifungal and antibacterial activity on the tested species of fungi and bacteria (Table 3). In case of the fungus *F. oxysporum*, there was a positive response to the application of the extract at the lower concentration. This was more apparent as the concentration was increased up to 50%. The inhibition zone increased to become the largest compared to the other tested fungi. Saadabi and Abu zaid (2011) found that the aqueous extract was very active against *A. niger*. Tijjani

et al. (2014) showed that the aqueous moringa seed extract exhibited more control of *A. flavus* isolated from rotted tomato fruits than the aqueous neem extract. The present study showed that the aqueous extract was effective in inhibiting the growth of *A. niger* contrary to the ethanolic extract. The inhibition zone diameter increased progressively as the concentration increased. Considering *N. mangifera*, there was no significant difference between 10% and 20% concentration but the difference was significantly high when adding 30% and 50% concentrations.

Results obtained from *in vitro* antimicrobial activity showed that the aqueous extract has considerable inhibitory effects against the tested bacteria and fungi at all tested concentrations. *F. oxysporum* was the most sensitive to the aqueous extract at the lower concentrations. As the concentration was increased to 30% and 50%, *B. subtilis* became as sensitive to the extract as *F. oxysporum*.

Antibacterial activity of aqueous extract

The aqueous extract of all concentrations was found to be inhibitory to the bacterium *B. subtilis* (Table 3). The inhibition zone increased significantly with the increase of extract concentration. As shown before (Table 2), 10% and 20% concentrations of the ethanol extract did not exhibit any effect on this bacterium. This clearly manifests the efficacy of the aqueous extract in controlling the bacterium even at the lower concentrations. Saadabi and Abu zaid (2011) reported that aqueous extract showed strong and superior antibacterial activity against all tested bacterial strains in 5%, 10% and 20% concentrations especially with regard to gram positive bacteria (*Staphylococcus aureus* and *B. subtilis*) as compared to methanol or petroleum ether.

Table 3. Antimicrobial activity of aqueous extract of moringa seeds against three species of fungi and one strain of bacteria

	Concentrations (%)			
	10	20	30	50
<i>Bacillus subtilis</i>	6.3 ^d _y	10.7 ^c _y	15.0 ^b _x	21.3 ^a _x
<i>Fusarium oxysporum</i>	9.0 ^c _x	13.7 ^b _x	15.3 ^b _x	23.3 ^a _x
<i>Aspergillus niger</i>	6.7 ^d _y	10.0 ^c _y	12.0 ^b _y	14.7 ^a _y
<i>Nattrassia mangifera</i>	8.0 ^c _{xy}	9.3 ^c _y	12.0 ^b _y	15.7 ^a _y

Means having the same letters in the rows (a-d) or columns (x-z) are not significantly different according to Duncan's Multiple Range Test at 5% level of significance.

Standard antibiotic and fungicide

The activity of moringa aqueous extract and ethanolic extract at 10% concentration against *F. oxysporum* were compared with the commercial fungicide (Thiram) at 25 μ l/ 3 g/ 100 ml concentration used as a reference. The efficacy of the aqueous extract was similar to Thiram, and both of them were better than the ethanolic extract (Table 4). Saadabi and Abu zaid (2011) also concluded that the methanol extract of the seeds obtained from moringa was less effective than the standard antibiotics used, but the aqueous extract of moringa seeds, on the other hand, was more effective.

Table 4. Antifungal and antibacterial effects of two moringa seed extracts compared to standard antifungal compound (Thiram) against *Fusarium oxysporum* and standard antibiotic (chloramphenicol) against *Bacillus subtilis*.

Antifungal compound	Inhibition zone diameter (mm)	Antibacterial compound	Inhibition zone diameter (mm)
Ethanolic extract	7.3 \pm 1.2 ^b	Ethanolic extract	0.7 \pm 0.0 b
Aqueous extract	9.0 \pm 1.0 ^{ab}	Aqueous extract	6.3 \pm 0.6 a
Thiram	10.3 \pm 1.5 ^a	Chloramphenicol	7.7 \pm 1.6 a

Means having the same letters in the rows are not significantly different according to Duncan's Multiple Range Test at 5% level of significance.

Similarly, the activity of moringa aqueous extract and ethanolic extract (10%) against *B. subtilis* was tested, using the antibiotic chloramphenicol (25 μ l/ 3g/100 ml) compound as a reference. The inhibition zone from the aqueous extract and chloramphenicol were similar and both of them showed a better control against the bacterium than the ethanolic extract (Table 4).

Postharvest disease control by coating and waxing

Edible surface coatings, such as waxes, are often applied to improve the cosmetic features of fruit and vegetables. They also contain ingredients which reduce water vapour loss, and also provide a medium for fungicides application (Hagenmaier and Baker, 1993; Debeaufort *et al.*, 1998; Alleyne and Hagenmaier, 2000). The mechanism by which fruit coatings delay fruit senescence is explained primarily as a response to the modification of internal atmosphere, including CO₂, O₂, and ethylene (Banks *et al.*, 1993; Saftner, 1999).

In this study, moringa aqueous extract alone or in combination with bee wax was used to improve the appearance and shelf life of tomato fruits. The results in Table 5 show that coating tomato fruits with the aqueous extract of moringa seed significantly reduced weight

loss in tomato fruits by about 50% in 4 days at 10 °C and up to 68% in 4 days at 37 °C compared to untreated control. It also reduced rotting of fruits resulting from fungal/bacterial infection by 33.3% and 56.6 % at 10 °C and 37 °C, respectively, compared to untreated control. Moreover, a synergistic effect was observed in improving the shelf life of tomatoes when combining moringa aqueous extract with bee wax emulsion in coating tomatoes. This combination significantly reduced the weight loss as high as 74% in 4 days. It also reduced microorganism infection by 90% at 10 °C and 80% at 37 °C (Tables 5 and 6). In case of wax emulsion alone, it reduced weight loss by about 76% at 37°C. Rot infection was also reduced by 43% and 40% at 10 °C and 37 °C, respectively (Tables 6 and 7). Irokanulo *et al.* (2015) demonstrated the effectiveness of coating with moringa seed powder in preserving tomato fruits inoculated with two bacteria, *Pseudomonas aeruginosa* and *Bacillus* sp.

Postharvest weight loss in tomato fruit is due to transpiration and respiration. Bee wax and moringa emulsion acted as barrier between the inner and outer environments of the fruit, and therefore reduced weight loss in tomato fruits beside the preservative nature of the moringa extract throughout the storage period.

Table 5. Weight loss (%) of tomato fruits coated with various emulsions and uncoated tomato after 4 days (4 D) or 10 days (10 D) at 10°C or 37°C.

Treatment	4 D at	10 D	4 D at	10 D
	10°C	at 10°C	37°C	at 37°C
Control	10.3 ^{c_x}	23.5 ^{b_x}	16.7 ^{bc_x}	69.2 ^{a_x}
Moringa	5.1 ^{c_y}	18.1 ^{b_y}	5.3 ^{c_{yz}}	47.1 ^{a_z}
Wax	2.4 ^{c_z}	16.4 ^{b_y}	6.1 ^{bc_z}	56.4 ^{a_y}
Moringa+Wax	2.6 ^{b_z}	11.9 ^{b_z}	6.6 ^{b_z}	53.2 ^{a_y}

Means having the same letters in the rows (a-d) or columns (x-z) are not significantly different according to Duncan's Multiple Range Test at 5% level of significance.

Table 6. Tomato fruit rotting (%) observed on tomatoes treated by moringa extract and wax emulsions stored at 10°C and 37°C for 10 days.

Treatment	10°C		37°C	
	Severity	%	Severity	%
Control	7.5a	50.0	12.5a	83.3
Moringa	2.5a	16.7	4.0b	26.7
Wax	6.5a	43.3	6.0ab	40.0
Moringa+Wax	1.5a	10.0	3.0b	20.0

Means having the same letters in the columns are not significantly different according to Duncan's Multiple Range Test at 5% level of significance.

CONCLUSION

This study has successfully shown that the ethanolic seed extracts of moringa seeds, with concentrations as low as 10% bear antifungal properties against the fungi *N. mangifera*, *F. oxysporum* and *A. alternata* and the bacteria *Actinomyces* sp. and *E. caratovora*. The bacterium *Bacillus subtilis* needs a minimum concentration of 30% to be controlled. The experiments showed that the aqueous seed extract had even more antimicrobial properties against the bacterium *B. subtilis*, and the fungi *F. oxysporum*, *N. mangifera* and *A. niger*. The latter was not inhibited by any of the ethanol concentrations. These extracts could be promising natural antimicrobial agents with potential applications in controlling bacteria and fungi that cause plant diseases. The extracts can provide a cheap (especially aqueous extract) and sustainable method toward disease reduction when issues of safety and toxicity are evaluated. More work is needed to purify and identify the exact phytochemicals responsible for this outstanding activity to be exploited commercially.

REFERENCES

- Adetunji, C. O., O.B Fawole, K.A. Arowora, S.I. Nwaubani, E.S. Ajayi, J.K. Oloke, O.N. Majolagbe, B.A. Ogundele, J.A. Aina, and J.B. Adetunji. 2012. Quality and safety of *Citrus sinensis* coated with hydroxypropyl methyl cellulose edible coatings containing *Moringa oleifera* extract stored at ambient temperature. *Global Journal of Waxing and Lining Materials on Storage Life of some Citrus Fruits Process* 92: 237-240.
- Alleyne, V. and R. Hagenmaier. 2000. Candelilla-shellac. An alternative formulation for coating apples. *Horticueral Science Journal* 35: 691–693.
- Banks, N.H., B.K. Dadzie, and D.J. Cleland. 1993. Reducing gas exchange of fruits with surface coating. *Postharvest Biololgy and Technology* 3: 269–284.
- Bobbaral V., K.P. Kumar, S.N.K. Chandra, and S. Penumajji. 2009. Antifungal activity of selected plant extracts against phytopathogenic fungi *Aspergillus niger*. *Indian Journal of Science and Technology* 4: 87-90.
- Chiejina, N.V. and C.N. Onaebi. 2013. *In vitro* fungicidal activity of two plant extracts against five phytopathogenic fungi of cucumber (*Cucumis sativus* L.) fruit. *International Journal of Applied and Natural Sciences* 2: 359-401.
- Dahot, U. M. 1998. Antimicrobial activity of small protein of *Moringa oleifera* leaves. *Journal of Islamic Academy of Science* 1: 27-32.
- Debeaufort, F., J.A. Quezada-Gallo, and A. Voilley. 1998. Edible films and coatings: Tomorrow's packaging: *Critical Review in Food Science* 38: 299–313.
- Eilert, U., B. Wolters, and A. Nahrstedt. 1981. The antibiotic principles of seeds of *Moringa oleifera*. *Planta Medica* 1: 55-61.
- Hagenmaier, R.D. and R.A. Baker. 1993. Reduction in gas exchange of citrus fruit by wax coatings. *Journal of Agriculture and Food Chemistry* 41: 283–287.
- Irokanulo, E. O., I.L. Egbezien and S. O. Owa. 2015. Use of *Moringa oleifera* in the preservation of fresh tomatoes. *Journal of Agriculture and Veterinary Science* 8: 127-132.
- Jabeen, R., M. Shahid, A. Jamil and M. Ashraf. 2008. Microscopic evaluation of the antimicrobial activity of seed extracts of *Moringa oleifera*. *Pakistan Journal of Biological Science* 40:349 - 358.
- Kekuda, T.R., N. Mallikarjun, D. Swathi, K.V. Nayana, B.M. Aiyar and T.R. Rohini. 2010. Antibacterial and antifungal efficacy of steam distillate of *Moringa oleifera* Lam. *Journal of Pharmaceutical and Research* 1: 34 -37.
- Nepolean, P., J. Anitha, and R. Emilin. 2009. Isolation, analysis and identification of phytochemicals of antimicrobial activity of *Moringa oleifera* (Lam.) essential oils for

the preservation of whole beef muscle. *Journal of Agricultural and Food Chemistry* 52: 68 – 95.

Palada, M. C. and L. C. Chang. 2003. Suggested cultural practices for *Moringa oleifera*. *The World Vegetable Center* 23: 1- 4.

Perez, C., M. Paul, and P. Bezique. 1990. An antibiotic assay by the agar well diffusion method. *Alta Biomed Group Experiences* 15: 1-13.

Saadabi, A. M. and E.I. Abu Zaid. 2011. An *in vitro* antimicrobial activity of *Moringa oleifera* L. seed extracts against different groups of microorganisms. *Australian Journal of Basic and Applied Sciences* 5: 129-134.

Saftner, R.A. 1999. The potential of fruit coating and film treatments for improving the storage and shelf-life qualities of ‘Gala’ and ‘Golden Delicious’ apples. *Journal of American Society for Horticultural Science* 124: 682–689.

Salman M., J. Anwar, W. Zaman, U.M. Shafique and A. Irafan. 2008. Preparation of oil, water emulsions of paraffin and bees waxes with water. *Journal of Scientific Research* 2: 555- 674.

Tijjani1, A., S.A. Adebitan, A.U. Gurama, S.G. Haruna and T. Safiya. 2014. Effect of some selected plant extracts on *Aspergillus flavus*, causal agent of fruit rot disease of tomato (*Solanum lycopersicum*) in Bauchi State. *International Journal of Biosciences* 4: 244-252.

Uma, C. and J.M. Sasikumar. 2005. Antimicrobial activity of traditional medicinal plants from Southern, Western Ghats. *Asian Journal of Microbiology, Biotechnology and Environmental Science* 7: 665-670.

Walter A., W. Samuel, A. Peter and O. Joseph. 2011. Antibacterial activity of *Moringa oleifera* and *Moringa stenopetala* methanol and n- hexane seed extracts on bacteria implicated in water borne diseases. *African Journal of Microbiology Research* 2: 153-157.

على بعض الميكروبات التي تصيب الطماطم كفاءة مستخلصات بذور المورينقا

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الخلاصة

شجرة المورينقا ذات قيمة غذائية وطبية عالية. هدفت هذه الدراسة لمعرفة مدى كفاءة مستخلصات بذور المورينقا باستخدام الكحول الإيثيلي، والأثير البترولي والماء في مكافحة البكتيريا والفطريات وتحسين معاملات ما بعد الحصاد لثمار الطماطم. أخذت 100 جرام من مسحوق بذور المورينقا، وتم الاستخلاص بجهاز سوكسلت أولاً بالبتروليم إيثر لمدة أربعة ساعات ثم الإيثانول لمدة ثمانية ساعات وتم تبخير المذيبات بتعريضها للهواء. وكان الناتج من المستخلص 34.94 مل للأثير البترولي و 14.47 جرام للإيثانول. أعدت تركيبات مختلفة من المستخلصات وكانت كالاتى (10%، 20%، 30%، 50%) لمعرفة كفاءتها لوقف نشاط خمسة انواع من الفطريات وهي : *Natrassia mangiefra* و *Aspergillus niger* و *Fusarium oxysporum* و *Alternaria alternata* و *Metarhizium anisoplae* وثلاثة انواع من البكتيريا هي *Bacillus subtilis*، *Actinomyces sp.* و *Erwinia caratovora*. مستخلص الكحول الإيثيلي اظهر فعالية كبيرة كمضاد بكتيرى ضد *Actinomyces sp.* وتأثيراً كبيراً ضد فطر *F. oxysporum* بينما كان للمستخلص المائي تأثير كبير في وقف نشاط *B. subtilis* وأكثر كفاءة ضد *A. niger* مقارنة بالكحول الإيثيلي والأثير البترولي التي لا يوجد لها تأثير *M. anisoplae* و *A. niger*. ازدادت مسافة التثبيط بزيادة نسبة التركيز للمستخلص تدريجياً. وفي تجربة اخرى تم استخدام المستخلص المائي للمورينقا مع شمع النحل لتغليف ثمار الطماطم وتخزينها في درجة حرارة 10 و 37 درجة مئوية لمدة 10 أيام وذلك لمعرفة تأثير مستخلص بذور المورينقا لزيادة فترة صلاحية ثمار الطماطم وإطالة فترة التخزين مع المحافظة على الجودة. وفي الأربع معاملات (شمع النحل و مستخلص المورينقا و إضافة الشمع مع 10 مل من المستخلص والثمار غير المعاملة) كان مستخلص المورينقا مع شمع النحل أكثر كفاءة في المحافظة على وزن ثمار الطماطم وتقليل تعرضها للعفن وتلها المعاملة بالشمع ثم مستخلص المورينقا مقارنة بالثمار غير المعاملة.