Antibacterial Activity of the Capsules of *Moringa oleifera* Lamk. (Moringaceae)

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ABSTRACT

This study determined the antibacterial activity of methanolic crude extract, purified dichloromethane extract and isolated parts from column chromatography of Moringa oleifera Lamk. capsules by agar-well diffusion. The methanolic crude extract showed no activity against Staphylococcus aureus ATCC 25923, Escherichia coli ATCC 25922, Pseudomonas aeruginosa ATCC 27853 and Klebsiella pneumoniae ATCC 67120. The purified dichloromethane extract and isolated parts from column chromatography showed antibacterial activity against these bacteria. Antibacterial activity began at 5-10% W/V concentration. It was proved that purified samples consisted of more active components, so they showed antibacterial activity against gram-positive and gram-negative bacteria.

Key words: *Moringa oleifera* Lamk., Antibacterial activity

INTRODUCTION

Moringa oleifera Lamk. is a member of the *Moringaceae* family. It originated in India, Sri Lanka and can be grown-up in Asia Minor and Africa as well. However, it is commonly found in Thailand. Several evidences revealed that *M. oleifera* had various pharmaceutical activities such as antibacterial (Eilert et al., 1981; Dayrit et al., 1990), antifungal, antispasmodic, anti-inflammatory, and diuretic activities (Carceres et al., 1992). Mutagenic activity of *M. oleifera* was also proposed by Villasena et al., 1989. The nutritional values of this plant were demonstrated by Verma et al., (1976) and Chakraborti et al., (1988).

As mentioned above, there is a good positive direction if the *M. oleifera* cultures from the northern part of Thailand have the similar valuable activity. In this present study, the technique for purification of crude extract of *M. oleifera* to screen the antibacterial activity was established. About 5 to 10% W/V of chromatographic fractions and purified dichloromethane extract showed the potential broad spectrum antibacterial activity against gram positive bacteria (*Staphylococcus aureus*) and gram negative bacteria (*Escherichia coli, |Pseudomonas aeruginosa,* and *Klebsiella pneumoniae*). Further studies are required to confirm and clarify the mechanism of action of *M. oleifera.*

MATERIALS AND METHODS

Plant material and preparation of extracts

Moringa oleifera Lamk. specimens were collected from Chang Puak market, Chiang Mai in 1996. A Voucher specimen (Nantachit 4401) is kept in the Chiang Mai University Pharmacy Herbarium. The capsules were dried at 40°C and powdered. Three kilograms of powder were macerated with 1 litre of methanol. Each replicate was macerated for 1 day, filtered and repeated 2 times. The filtrate was evaporated in a vacuum. The residue (crude extract) was dark green and the percent yield was 20.0.

Purification of crude methanolic extract

Crude methanclic extract 22.10 gm was dissolved in 100 ml of dichloromethane and was extracted with 100 ml of water. The dichloromethane layer was washed with 150 ml of water and emulsion occurred. Anhydrous calcium chloride was added in order to break the emulsion and dichloromethane was evaporated to dryness by a vacuum pump. The dichloremethane extract was further purified by column chromatography. Silica gel 60 (35-70 mesh) was used as an adsorbent and the column was eluted with 3% methanol in dichloromethane. A total 24 fractions, 20.0 ml each, were collected. The total fractions were separated into 4 parts by a thin layer chromatogram.

Part 1, fraction 4 to fraction 7 was 0.34 gm
Part 2, fraction 8 to fraction 12 was 0.83 gm
Part 3, fraction 13 to fraction 17 was 0.92 gm
Part 4, fraction 18 to fraction 24 was 0.08 gm

Determination of antibacterial activity of crude methanolic extract, purified dichloromethane extract and column chromatographic fractions by agar-well diffusion

Crude methanolic extract, purified dichloromethane extract and column chromatographic fractions were dissolved in methanol and screened for antibacterial activity against *Staphylococcus aureus* ATCC 25923, *Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853 and *Klebiella pneumoniae* ATCC 67120 by using 5-10% W/V concentrations of the extracts and fractions (Lenette, 1980; Washington, 1981).

RESULTS

Crude methanolic extract at 10% W/V concentration showed no activity against the bacteria tested.

Purified dichloromethane extract and four parts of column chromatographic fractions showed antibacterial activity against all four pathogenic bacteria tested at 10% W/V, except fractions 18 to 24 which showed antibacterial activity at 5% W/V concentration because there was not enough of the last chromatographic fractions, so it was tested at 5% W/V concentration (Table 1).

Table 1. Antibacterial activity of crude methanolic extract, purified dichloromethane extract and four parts of column chromatographic fractions.

Fraction Tested	Inhibition zone (mm)			
	***S. aureus* ATCC 25923**	***E. coli* ATCC 25922**	***P. aeruginosa* ATCC 27853**	***K. pneumoniae* ATCC 67120**
Control (MeOH)	0	0	0	0
Crude methanolic extract (10% W/V)	0	0	0	0
Purified dichloromethane extract (10% W/V)	11, 11	11, 11	11, 11	10, 11
	11, 11	10, 10	11, 11	9, 10
average	11	10.5	11	9.8
F_4 - F_7 (10% W/V)	10, 10	11, 10	11, 11	9, 10
	10, 10	10, 10	10, 10	9, 9
average	10	10.2	10.5	9.2
F_8 - F_{12} (10% W/V)	11, 10	11, 10	9, 10	10, 10
	11, 10	10, 10	9, 10	10, 10
average	10.5	10.2	9.5	10
F_{13} - F_{17} (10% W/V)	10, 10	10, 10	11, 10	12, 12
	10, 10	10, 10	11, 11	11, 11
average	10	10	10.8	11.5
F_{18} - F_{24} (5% W/V)	11, 11	9, 10	10, 10	10, 11
	11, 11	9, 10	10, 10	10, 11
average	11	9.5	10	10.5

F = column chromatographic fraction

DISCUSSION AND CONCLUSION

Purified dichloromethane extract and four parts from column chromatographic fractions showed antibacterial activity against gram-positive and gram-negative bacteria at 5-10% W/V concentration. Crude methanolic extracts at the same concentrations showed no activity because there was not enough active constituent in the crude methanolic extract and the extracts must be freshly prepared.

From the literature reviewed, it was found that capsules, leaves and seeds of *M. oleifera* Lamk. have high nutritional value, vitamins and many minerals. Roasted seeds showed mutagenic activity and water extracts from flowers, leaves, roots, seeds and bark of *M. oleifera* Lamk. showed anti-spasmodic, anti-inflammatory and diruetic activities. The seeds of this plant also showed anti-microbial activity. From my preliminary investigation, I found that *M. oleifera* Lamk. capsules from northern part of Thailand showed antibacterial activity against gram-positive and gram-negative bacteria. From this information, it is reasonable to promote *M. oleifera* Lamk. as an economic plant.

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