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Antimicrobial activities of extracts for some of medicinal plants

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ABSTRACT

Comparative antimicrobial effects of extracts from seeds of *Linum usitatissimum* and *Lepidium sativum*, leaves and stems of *Monelchma ciliatum* against standard organisims (*Bacillus subtilis, Staphylococcus aureus, Escherichia coli, Proteus vulgaris, Pseudomonas aeruginosa, Candida albicans* and *Aspergillus nige*r were investigated. Methanolic extract of *Lepidium sativum* caused significant antimicrobial activity while chloroformic and water extracts have low activity. *Linum ustatissimum* had very low activity. *Monechma ciliatum* stems methanolic extract was active more than chloroform extract. Water extract caused inhibition zone against *Psedomonas vulgaris* only. Methanolic extract of *Monelchma ciliatum* leaves was the most active extract against standard organisms.

1. Introduction

The use of plants for treatment of various diseases is universal, and has been practiced for many years even up to now where many people are treated by modern drugs. Both literate and illiterate people still use local plants as drugs in many conditions (Kokwaro, 1976).

Although plants may represent a potential source of antibiotic none has been reported of higher plant origin (Aizenman, 1978).

Monechma ciliatum belongs to the family Acanthaceae, locally known as "Black Mahlab.

Black Mahlab is a famous medicinal plant in western Sudan; its seeds are used as an effective laxative and contain an essential oil which emits a sweet and pleasant odor. It also played an important role as a medicine. It is used for general body pain, bowel trouble and sterility in women (Hedberg and Stangard, 1989).

Phytochemical and pharmacological characters of *Monechma ciliatum* was studied by Uguru and Evans (2000). The leaves of *M. ciliatum* were found to contain alkaloids, glycosides, proteins, tannins and saponins. The hot methanol extract of the leaves was found to have potent oxytocic effect, which may constitute an amino acidic derivative.

Lepedium sativum **t**he plant belongs belongs to the family Brassicaceae.The native names are cress, pepper ress and pepper wort; the Arabic name is ELRashad.The heial onstituents of the plant include phenolic compounds (Ozeker and Esiyok, 1999), imidazole and alkaloids (Maier, *et al*., 1998), hydroxylated glutamic acids (Bell *et al*., 1981) sinigrin, K-salt (Thies, 1988); flavonoid compounds (Paszkowski and Kremer, 1988), sterols and Benzyl glucosinolate degradation protect (Gil and Macleod, 1980). *Lepidium sativum* contained glucotropaeolin as reported by Songsak and Lockwood (2002).

Phytochemical screening of *Lepidium sativum* studied by Nuha (2006) showed that the plant was positive for Triterpenses, Alkaloids, Flavonoids, Tannins and Cumarins but negative for Saponins, Cynogenicglycoside and Anthraquinoneglycoside

The seeds of *Lepidium sativum* are used for treatment of fracture healing in Saudi traditional medicine (Ahsan *et al*., 1989). The mucilage in the outer seed is used as substitute for tragacanth and Gum Arabic (Mathews *et al*., 1993).

Linum usitatissimum **t**he plant belongs to the family *Linaceae*ca. The Arabic name of the plant is Kettan, Berber, Tifert and Delkmouch and English name is Commonfax (Kokwaro, 1976).

Traditionally the plant was used to treat fever.Seeds used as a laxative, soothing and pain – relieving .It is used for inflammation of digestive and urinary tracts, antidiarrhoeic, often mixed with Althaea flowers. Seeds used in preparation of cataplasms for their emollient prosperities against boils and inflammations (Kokwaro, 1976).*Linum usitatissimum* contains linamarin which contains a peptide, linatine and has an antipyridoxine action in chickens (Humphreys, 1988).

2. Materials and methods

2.1. Plants extract

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Three plants were used; *Monechma ciliatum* (leaves and stems) and *Lepidium sativum* were collected from Medicinal and Aromatic Plants Research Institute (MAPRI) Farm while *Linum usitatissimum* seed was supplied by herbalist.

The plants were authenticated by the scientist MAPRI. The plants were coarsely powdered soaked in chloroform and extracted according to Harborne, (1988).

2.2. Microbes

Gram positive and gram negative organisms were used. The gram positive organisms were Bacillus subtitis (NCT 8236) and Staphylococcus aureus (ATCC25923) while the gram negative organisms were Escherichia coli (NCTC8196), Proteus vulgaris (ATCC6380) and Pseudomonas areuginosa (ATCC27853)).Two fungi, Aspergillus niger (ATCC9763) and Candida albicans (7596) were used All organisms are obtained from National collection of culture Type Colindale (NCTC) England and American Type culture collection Rockville Maryland, USA.

2.3. Antimicrobial assay

The cup –plate –agar diffusion method described by Kavanagh (1972) was adopted with some minor modifications, to assess the antibacterial activity of the extracts.

Three ml of each of the five standardized bacterial stock suspensions (10^8-10^9 C.F.U. /ml) were thoroughly mixed with 300ml of sterile melted nutrient agar maintained at 45ºC.

20 ml aliquots of the inoculated nutrient agar were distributed into sterile Petri-dishes. The plates were divided into two halves, two cups in each half (10mm in diameter). Alternate cups were filled with 0.1ml samples of each of the extracts and allowed to diffuse at room temperature for 2 hour.

The plates were then incubated in the upright position at 37ºC for 18 hours.

Two replicates were carried out for each extract against each of the tested organisms.

Simultaneously positive controls involving the addition of the respective solvents instead of the extracts were carried out separately. The diameters of the growth inhibition zones were measured and the mean values were tabulated.

The antifungal activity was measured by similar method for testing antibacterial activity using sabouraud dextrose agar. The inoculated media was incubated at 25ºC for two days for the *Candida albicans* and three days for *Aspergillus Niger*.

3. Results

The methanolic extract of *Monechma ciliatum* stems showed high antimicrobial activity against *Proteus vulgaris* (24) and *Staphylococcus aureus* (20), moderate activity against *Escherchia coli* (18) and low inhibition zone against *Bacillus subtilis* (12), *Pseudomonas aeruginosa* (12) and *Candida albicans* (12). On the *other hand* the Chloroform extract showed moderate activity against *Staphylococcus aureus* (17), *Proteus vulgaris*, and *Bacillus subtilis* (15). It had low activity against *Escherchia coli* (13), *Pseudomonas aeruginosa* (13) and *Candida albicans* (13).

The water extract of *Monechma ciliatum* stems, had high antimicrobial activity against *Pseudomonas aeruginosa* (23), while the activity was absent against all other standard organisms tested.

Aspergillus niger was resistant to the all plant stem extracts (Table1) The antimicrobial activity of methanolic extract of *Monechma ciliatum* leaves was moderate against *Proteus vulgaris* (19) and *Escherchia coli* (18), low against *Bacillus subtilis* (17), *Pseudomonas aeruginosa* (17) and *Staphylococcus aureus* (17). The activity was absent aganst *Candida albicans* and *Aspergillus niger*.

The chloroform extract of *Monechma ciliatum* leaves caused high inhibition zone against *Staphylococcus aureus* (23) and low against *Bacillus subtilis* (15) and *Candida albicans* (14). However, the extract showed low activity against *Escherchia coli* (13), *Proteus vulgaris* (13) and *Pseudomonas aeruginosa* (13).

Water extract of *Monechma ciliatum* leaves caused moderate activity against *Pseudomonas aeruginosa* (17) and low inhibition zone activity against *Bacillus subtilis* (13) and *Staphylococcus aureus* (13). The activity was absent against *Escherchia coli*, *Proteus Vulgaris, Candida albicans* and *Aspergillus Niger* (Table 2).

The methanolic extract of *Linum usitatissimum* caused an inhibition growth zone diameter. It was moderate in *Pseudomonas aeruginosa* (18) and *Bacillus subtilis* (16), low against *Proteus vulgaris* (15), *Staphylococcus aureus* (14) and *Escherchia coli* (13). The activity was absent against *Candida albicans* and *Aspergillus niger*.

The antimicrobial activity of Chloroform extract of *Linum usitatissimum* was low against *Pseudomonas vulgaris* (13) and inactive against *Bacillus subtilis, Staphylococcus aureus, Escherchia coli, Proteus vulgaris, Aspergillus niger* and *Candida albicans*.

Water extracts showed low activity against *Candida albicans* (14) while there was no activity against other organisms. (Table 3)

The methanolic extract of *Lepidium sativum* had high inhibition against *Candida albicans* (21) and *Proteus vulgaris* (20). The antimicrobial activity of the extract was low against *Bacillus subtilis* (13), *Staphylococcus aureus* (13), *Escherchia coli* (13) and *Pseudomonas aeruginosa* (13). The extract was inactive against *Aspergillus Niger*.

The chloroform extract of *Lepidium sativum* had very low activity against *Staphylococcus aureus* (12) and *Pseudomonas aeruginosa* (12) and the activity was absent against *Bacillus subtilis, Escherchia Coli, Proteus Vulgaris, Candida albicans* and *Aspergillus niger*.

The antimicrobial activity of water extract of *Lepidium sativum* was low against *Bacillus subtilis* (15) and *Escherchia coli* (15). The extract had no effect against *Staphylococcus aureus, Proteus Vulgaris, Candida albicans* and *Aspergillus Niger* (Table 4).

Table 1: The antimicrobial activity of Monechma ciliatum stems against standard organisms

Solvent System	Yield	Conc**. ** (mg/ml)	MIZD*(Mm) Bacteria					Fungi	
			B. s	S. a	E. coli	Pro. v	Pseudo. a	Asp. n	C.a
Methanol	4.88	100	12	20	18	24	11	-	12
Chloroform	6.05	100	15	16	13	17	12	-	13
Water	0.04	-	-	-	-	-	23	-	-

* MIZD: Mean of inhibition growth zone diameter. (Mm): Millimeter
** Conc: Concentration of extractin solvent.
Bacillus subtilis (B. s); Staphylococcus aureus (S .a) ; Escherichia coli (E .coli) ; Proteus vulgaris (pro. v) ; Pseudomonas aeruginosa (Pseudo. a) ; Aspergillus niger (Asp. n) ; Candida albicans (C.a.).
Low=< 15mm, Moderate = 19-20 mm, High=20-25mm, V. High=>25mm.

Table 2: The antimicrobial activity of Monechma ciliatum Leaves against the standard organisms

Solvent System	٪Yield	Conc**. ** (mg/ml)	MIZD*(Mm) Bacteria					Fungi	
			B. s	S. a	E. coli	Pro. v	Pseudo. a	Asp. n	C.a
Methanol	12.43	100	17	16	18	19	17	-	-
Chloroform	11.13	100	15	23	13	13	13	-	14
Water	0.09	-	13	12	-	-	17	-	-

* MIZD: Mean of inhibition growth zone diameter. (Mm): Millimeter
** Conc: Concentration of extractin solvent.
Bacillus subtilis (B. s); Staphylococcus aureus (S .a) ; Escherichia coli (E .coli) ; Proteus vulgaris (pro. v) ; Pseudomonas aeruginosa (Pseudo. a) ; Aspergillus niger (Asp. n) ; Candida albicans (C.a.).
Low=< 15mm, Moderate = 19-20 mm, High=20-25mm, V. High=>25m

Table 3: The antimicrobial activity of Linum usitatissmum against the standard organisms

Solvent System	٪Yield	Conc.** (mg/ml)	MIZD (Mm) * Bacteria					Fungi	
			B. s	S. a	E. coli	Pro. v	Pseudo. a	Asp. n	C.a
Methanol	4.6	100	16	14	13	15	18	-	-
Chloroform	28.5	100	-	-	-	-	13	-	-
Water	0.041	-	-	-	-	-	-	-	14

* MIZD: Mean of inhibition growth zone diameter. (Mm): Millimeter
** Conc: Concentration of extractin solvent.
Bacillus subtilis (B. s); Staphylococcus aureus (S .a) ; Escherichia coli (E .coli) ; Proteus vulgaris (pro. v) ; Pseudomonas aeruginosa (Pseudo. a) ; Aspergillus niger (Asp. n) ; Candida albicans (C.a.).
Low=< 15mm, Moderate = 19-20 mm, High=20-25mm, V. High=>25mm.

Table 4: The antimicrobial activity of Lepidium sativum against the standard organisms

Solvent System	Yield%	Conc. * * (mg/ml)	MIZD*(Mm) Bacteria					Fungi	
			B. s	S. a	E. coli	Pro. v	Pseudo. a	Asp. n	C.a
Methanol	98.2	100	13	13	13	20	13	-	21
Chloroform	9.0	100	-	11	-	-	12	-	-
Water	0.03	-	15	-	15	-	-	-	-

* MIZD: Mean of inhibition growth zone diameter. (Mm): Millimeter
** Conc: Concentration of extractin solvent.
Bacillus subtilis (B. s); Staphylococcus aureus (S .a) ; Escherichia coli (E .coli) ; Proteus vulgaris (pro. v) ; Pseudomonas aeruginosa (Pseudo. a) ; Aspergillus niger (Asp. n) ; Candida albicans (C.a.).
Low=< 15mm, Moderate = 19-20 mm, High=20-25mm, V. High=>25mm.

4. Discussion

Plants are known to produce certain bioactive molecules which react with other organisms to the environment inhibiting bacteria or fungal growth (Harbone, 1988). This finding is interesting because antibiotic used are often more active against Gram positive rather than Gram negative bacteria. Lee *et al*. (2007) reported that plant extracts demonstrated strong antibacterial activity against Gram positive and Gram negative bacterial strains.

In the present study the aqueous extract of the plant shoed no antimicrobial activity against the tested organisms, whereas the organic extracts presented antimicrobial activity of least for most of the microorganism tested. The methanolic extract was found to be more effective than chloroform extract. This could be due to the fact that the active constituents which had antimicrobial effect dissolved readily in methanol rather than chloroform. Similarly Mohammed (1979) reported wide range of antimicrobial activity of *Hibiscus*

sabdariffa (karkadeh) extracts using different solvents.

From the present study we find that *Monechma ciliatum* was the most effective antimicrobial agent compared to *Lepidium sativum* and *Linum usitatissimum*. This is mainly due to the variation chemical constituents of the plants. Moreover the leaves extract of *Monechma cilatum* was more potent than the stem extracts. This may be due to the different concentration of the active antimicrobial component in different parts of the plant. All standard bacteria, Gram positive and Gram negative, were sensitive to the methanolic extract of *Monechma cilatum* but fungi responded less which indicated the narrower spectrum of the plant.

The present investigation has shown that methanolic extract of *Lepidium sativum* had high activity against Gram-negative *Proteus vulgaris* and the yeast *Candida albicans*, but low against Gram-positive *Bacillus subtilis, Staphylococcus aureus*, while the chloroform extract was inactive against all standard organisms tested. Asma (2003) reported similar effects in her study of the antimicrobial activities of *Lepidium sativum* seeds. She found that the methanol extract of *Lepidium sativum* was the most active extract. She also pointed out that the clinical isolates exhibited low susceptibility to the plant extract compared with the standard organisms.

In the present investigation the *Linum usitatissimum* seeds extracts showed very low antimicrobial activity against standard organisms compared with the other plants extracts tested in this study. Methanol extract of *Linum usitatissimum* caused moderate to low inhibitory zone in Gram-positive and Gram-negative bacteria. However, the topical wound healing properties of Canisep cream containing oils from *Linum usitatissimum* had an antimicrobial property (Amresh *et al*., 2005).This may indicate that *Linum usitatissimum* has a synergistic effect as antimicrobial properties.

The response of the three plants was very week or negative for *Candida albicans*. None of the plant extract was positive for Asperigellus Niger. This indicated that the plants had very little antifungal components. In conclusion these results suggested that *Monechma ciliatum* and *Lepidium sativum* are a source of bioactive substances endowed with interesting antibacterial activites. Further investigations are required to confirm its activities, to find active components of the extract and to confirm the mechanism of action.

References

Ahsan SK, Tarig M, Ageel M, Alyanya MA and Shah AH (1989). Studies on some herbaldrugs used in fracture healing. International J. of Crude-Drug Research, 27(4), 235-239.

Aizenman BE (1978) .Higher Plants as a source For Preparation of new antibiotics Microbial Zh. (Kiev) 40:233.

Amresh K, Dass LL, Samir S and Anwar T (2005). Birsa Agricultural University, Ranchi (Jharkhand), India. Indian Journal of veterinary surgery Vol. (26) (No. 1) 28- 30.

Asma, A.M. (2003). Antimicrobial activity of *Lepidium sativum*. Sudan University of Science and Technology – College of Post Graduate Studies. For Higher Diploma in clinical Microbiology – Khartoum.

Bell EA, Meiei LK and Sorensen H (1981). Hydroxylated glutamic acids in phlox, Lepidium and Rheum species.Phytochemestry, 20(9), 2213-2216.

Gil V and Macleod AL (1980). The benzyl glucosinolate degradation in Lepidium sativum: effect of plants age and time of autolysis.Phytochemistry, 19(7), 1365- 1368.

Harborne JB (1988). Phytochemical method. 2nd Edition.

Hedberg I and Stangard F (1989). Traditional Medicine in Botswana. Traditional Medicinal Plants. I pelegeny Publisher Sweden.

Kavanagh F (1972). Analytical Microbiology. Vol. 11. Academic Press (Pub.). Newyork and London.

Kokwaro JO (1976). Medicinal Plants of east Africa. Nairobi University.

Lee SB, Cha KH, Kim SN, Altantsetseg S, Shatar S, sarangerel O and Nho CN (2007). The antimicrobial activity of essential oil from Dracocephalm foelidum against pathogenic organisms. J. Microbiol. 45(1), 53-57.

Maier UH, Gundlash H and Zenk MH, (1998). Seven imidazole alkaloids from Lepidium sativum. Phytochemistry, 49(6), 1791- 1795.

Mathews S, Singhal RS and Kulkarni PR (1993). Some Phsiochemical characteristics of Lepidium sativum seeds. Nahrung, 37(1), 69- 71.

Mohamed WK (1979). A phytochemical investigation of certain pigment bearing plants. A thesis presented for degree of Master of Pharmaceutical Science in Pharmacognosy. Pharmacognosy Department, Faculty of Pharmacy, Cairo University, Cairo, Egypt.

Nuha H (2006). Hepatoprotective activities of some Sudanese medicinal plants. M.V. Sc. Thesis, University of Khartoum. Sudan.

Ozeker E and Esiyok D (1999). Identification of phenolic compounds in seeds of different rocket species (Eruca saliva and Diplotaxin tenuifolia) and Land cress (Lepidium sativum). Crui ferae-News letter, No. 21, 21- 22.

Paszkowski WI and Kremer RJ (1988). The Biological activity and tentative identification of flavonoid components (Abatilon Thephrasti Medik) seed coat. J. of Chemical Ecology, 14 (7), 1573- 1582.

Songsak T and Lockwood GB (2002). Glucosinolates of seven medicinal plants from Thailand. J. Fitoterapia 73, 209-216.

Thies W (1988). Isolation of sinigrin and glucotropaeolin from cruci ferous seeds –fat-science and technology, 71(8), 311- 314.

Uguru MO and Evans F (2000). Phytochemical and pharmacological studies on Monechma ciliatum. J. of Ethnopharmacol, 73(1-2): 289- 92.