Journal of Medicinal Plants Research Vol. 5(27), pp. 6299-6308, 23 November, 2011
Available online at http://www.academicjournals.org/JMPR
ISSN 1996-0875 ©2011 Academic Journals
DOI: 10.5897/JMPR11.533

Full Length Research Paper

Anti-microbial efficacy and biochemical analysis from different parts of *Acacia nilotica L.* and *Ricinus communis* L. extracts

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Accepted 2 August, 2011

The present study was under taken to explore the potential of *Acacia nilotica* L and *Ricinus communis* L as antimicrobial agents in relation with various important bio-molecules and to check their correlation as antimicrobial agent. In present study water and methanol extract (20%) of different parts of *A. nilotica* and *R. communis* showed good inhibition against Gram positive (*Staphylococcus aureus* and *Streptococcus pneumoniae*), and Gram negative bacteria (*Pseudomonas aeruginosa*, *Escherichia coli, Klebsiella pneumoniae)* and some fungi (*Aspergillus niger*, *Penicillium expansum* and *Aspergillus fumigatus*). The presence of significant concentration of total protein, total sugar, reducing sugar, some identified free amino acids and sugars provided good correlation among antimicrobial compounds which are present in both plants extracts and have capability to kill the pathogens or inhibit the growth of microbes.

Key words: *Acacia nilotica* L., *Ricinus communis* L., antibacterial, antifungal, biomolecules.

INTRODUCTION

Pathogenic microorganisms have capability to develop resistance against commercially available antibiotic or antimicrobial drugs, which are commonly used for the treatment of infectious diseases. To avoid undesirable side effect of certain antibiotics and emergence of previously unknown infections, researchers are focusing to explore new antimicrobial substance from various sources, such as medicinal plants (Marchese and Shito, 2001).

Furthermore, medicinal plant extract are rich source and contain mixture of compounds or secondary metabolite like tannis, terpeniod, alkaloids, flavonoids, etc that exhibits a new potential source of remedy against anti-infectious pathogens and is cheaper than modern drugs (Amani et al., 1998; Cowan, 1999; Dahanukar et al., 2000; Mahesh and Satish, 2008). Medicinal plants are used by 80% of the world population as the only available medicines especially in developing countries (Hashim et bark, roots, fruits, twigs exudates and modified plant organs have various medicinal properties and are al., 2010). Different parts of medicinal plant such as stem, collected as raw drugs by local communities and traded in the market as a raw material for herbal industries (Uniyal et al., 2006). Accordingly, plants and its metabolites are safe and its continuous use as a drug is observed to be an alternative source to cure the human pathogenic diseases and this approach is in practice from the ancient time (Archana et al., 2011). The *A. nilotica* (Babool) and *R. communis* (Castor bean plant) are available in large quantities in Pakistan and used as folk medicine against various human pathogens. These medicinal plant Castor bean (*R. communis*) family Euphorbiaceae and Babool (*A. nilotica*) family *Fabaceae* are native to Eastern Africa and South Asia. Indo-Pakistan region.

The annual crop grows on marginal lands and coastal sandy belts under warm climates (Maslin et al., 2003; Conceicao et al., 2007a, b). These genus contain variety

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of secondary metabolites such as flavonoids, phenolic acid, saponins, saponin glycosides, volatile oil, triterpenoid, tannins, alkaloids which have numerous biological and pharmacological properties and are very valuable phytochemicals of these plants (Singh et al., 2009a; Solomon and Shittu, 2010).In the medical point of view, castor bean has been used with confidence for the treatment of several diseases, asthma, boils burns cold, colic, catarrh, chancre, cholera, cancer, carbuncle convulsions, and 'craw-craw' (Lakshminarayana and Sujatha, 2005; Boeck-Neto et al., 2005; Korwar et al., 2006). The seeds are the most toxic part of the plant although the leaves are also poisonous. The seeds contain 0.2 to 3% ricin and one mg of pure ricin can be isolated from its grain. Methanolic extracts of the leaves of *R. communis* were used antimicrobial testing against eight pathogenic bacteria in rats and showed antimicrobial properties. However, extract did not show any toxicity (Oyewole, 2010). Although, lethal dose considered to be 4 to 8 seeds, reports of actual poisoning are relatively rare (Wedin et al., 1986). Babool (*A. nilotica*) is primarily grown for the purpose of fuel and timber.

However, the pods of *A. nilotica* are fed to cattle to increase the milk yield and also used ethno-medicinally for the treatment of skin, sexual, stomach, malaria, sore, throat as well as for tooth problems. The fresh parts of this plant have been reported to be most active against Hepatitis C virus (Hussein et al., 2000). The herbal product derived from *Acacia* species are sold in the market either pure or mixed form like babool tooth past, Ayur shampoo and Nyle Shampoo, etc. Few reports are available on different parts of *A. nilotica* and *R. communis* as antimicrobial agent and their correlation with bio-molecules of both plant extracts against bacterial and fungal species.

MATERIALS AND METHODS

Plant material and growth of micro-organism

The different parts of *A. nilotica* (leaves, stem and bark) and *R. communis* (leaves and stem) were collected from adjunct area of district Jamshoro, Sindh, Pakistan. The voucher specimens of the plants were deposited in the Institute of Plant Sciences, University of Sindh, Pakistan.

Bacterial culture of *E. coli, P. aeruginosa, K. pneumoniae, S. aureus* and *S. pneumoniae* were maintained on L.B. solid medium (Luria and Burrous, 1955; Miller, 1972) and fungal species *A. niger*, *P. expansum* and *A. fumigatus* were grown and maintained on nutrient agar medium at 37°C.

Gram staining method and preparation of inoculum

A thin smear of species on the glass slide was made by Gram staining method (Gram, 1884). The Gram positive (*S. aureus* and *S. pneumoniae*) and Gram negative bacteria (*P. aeruginosa*, *E. coli* and *K. pneumoniae)* were pre-cultured in L.B broth for overnight at 37°C. The fungal inoculums (*A. niger*, *P. expansum* and *A. fumigatus*) was prepared from 5 to 7 day old culture grown on glucose medium.

Preparation of aqueous and methanol extracts of plant

Collected plant material was dried at room temperature in the shade and was ground to fine powder in a homogenizer. 20 g of powder sample was crushed with glass powder with distilled water and 90% methanol and centrifuged at 7000 rpm for 10 min; filtered the supernatant through a Whatt-man No. 1 filter paper. The supernatant was transferred into 100 ml volumetric flask and this procedure was repeated twice. The final volume was made up to 100 ml with distilled water or 90% methanol (Dahot, 1999).

Antibacterial and antifungal activity

The cultures of bacteria grown overnight at 37°C were used for testing the antibacterial activity from *A. nilotica* and *R. communis* extracts (not cultures were used for testing antibacterial activity but extracts). The antibacterial activity was checked by agar diffusion method (Mothana and Lindequist, 2005). While Antifungal activity was tested against *A. niger*, *P. expensum* and *A. fumigatus* through diffusion plate method with slightly modification as reported by Terras et al. (1995). L.B. nutrient agar was inoculated with a microbial cell suspension (300 µl in 20 ml of medium) and poured into sterile petri dishes. Sterile filter paper discs 6 mm in diameter were impregnated with 5 µl of each plant extract sample and placed on the inoculated agar surface. Standard 6 mm discs containing Ampiclox 30 µg/disc; Enxocibid 30 µg/disc; Nystain 100 units/disc; Erythromycin 5 µg/disc; Fucidin 100 µg/disc; Romicef 30 µg/disc; Cefspan 5 µg/disc and Cefizox 30 µg/disc were used as positive controls. Plates were incubated overnight at 37°C for 24 h. In contrast, fungus was incubated at 37°C for 120 h. After incubation antimicrobial activity was evaluated by measuring the inhibition zones (mm) ±SD of three replicates.

Determinations of total protein

Total protein content from 20% water and methanol extract of *A. nilotica* (leaves stem and bark) and *R. communis* (leaves and stem) were determined by Lowry et al. (1951). The extract of both plants (0.5 ml) was mixed with 2.5 ml alkaline copper solution and after 10 min 0.25 ml of diluted Folin Ciocalteu (1:1 v/v with water) was added. The absorbance of colour produced was measured after 30 min at 750 nm against blank.

Determinations of total sugar and reducing sugar

Total sugar contents from 20% water and methanol extract of *A. nilotica* (leaves stem and bark) and *R. communis* (leaves and stem) were determined by phenol sulfuric acid method reported by Montgomery (1960). The extract sample of both plants (0.5 ml) was mixed with 2.5 ml sulphuric acid and 0.05 ml 80% phenol. The colour intensity of reaction mixture was read at 485 nm after 15 min against blank. In case of reducing sugar contents from extracts 1.0 ml of sample was mixed with 1.0 ml DNS and the reagent was heated in boiling water bath for 5 min. The absorbance was measured at 540 nm against blank (Miller, 1959).

Table 1. Antibacterial activity of different parts of *A. nilotica* and *R. communis* from 20% methanol extract.

***Acacia nilotica* methanol extract**				
Microorganisms	**Inhibition zones (mm)**			
Gram-positive	**Leaves**	**Stem**	**Bark**	**Methanol (control)**
Staphylococcus aureus	14	7	7	Nil
Streptococcus pneumoniae	12.5	10	7.5	Nil
Gram-negative				
Escherichia coli	8.5	10.5	7	Nil
K. pneumoniae	7	8	7	Nil
P. aeruginosa	7	7	6	Nil
***Ricinus communis* methanol extract**				
Gram-positive				
Staphylococcus aureus	7.5	7	N.T	Nil
S.spneumoniae	11	8.5	N.T	Nil
Gram-negative				
Escherichia coli	8	9	N.T	Nil
K. pneumoniae	9	7	N.T	Nil
P. aeruginosa	6	7	N.T	Nil

N.T: not tested.

Identification of amino acids and sugars by thin layer chromatography (TLC)

The identification of amino acid and sugar from the water and methanol extract of *A. nilotica* and *R. communis* were checked by TLC. Free amino acids were identified by following solvent composition as butanol, acetic acid and water (4:1:5 v/v/v) while for sugar identification, butanol, acetic acid and water (5:1:4 v/v/v) were used as solvent for thin layer chromatography. The spots were visualized and marked each one and assigned with Rf value.

RESULTS AND DISCUSSION

Medicinal plants contain different antimicrobial compounds, which are responsible to kill the pathogen or inhibit their growth. Several scientists revealed similar results from both plants against Gram positive (*S. aureus* and *S. pneumoniae*), Gram negative bacteria (*P. aeruginosa*, *E. coli* and *K. pneumoniae*) (Solomon and Shittu, 2010; Mwine and Damme, 2011) 20% water and methanol extract of *A. nilotica* and *R. communis* leaves and stems were tested against both Gram negative *E. coli, K. pneumoniae, P. aeruginosa* and Gram positive, *S. aureus* and *S. pneumoniae* pathogens and three species of fungus that is, *A. niger, A. fumigatus* and *P. expansum.* 20% methanol extracts of *A. nilotica* and *R. communis* exhibited significant antimicrobial activity and maximum inhibition zones against both Gram positive and Gram negative pathogens were noted as 14 mm, 12.5 mm *A. nilotica* leaves, 7.5 and 11 mm *R. communis* leaves against *S. aureus, S. pneumoniae,* and 10.5 mm, 10 mm *A. nilotica* stem against *E. coli* and *S. pneumonia,* respectively. However, minimum inhibition zones also observed around 6 to 9 mm against tested pathogens by both different parts of methanol plant extract as reported in Table 1. Mahesh and Satish (2008) reported that leaf extracts of *A. nilotica* showed significant antimicrobial activity against *E. coli*, *S. aureus* and *Xanthomonas axonopodis pv. Malvacearum* against *Bacillus subtilus.* While the bark extract of the same plant also showed highest antimicrobial activity against *B. subtilus* and *S. aureus.* The influence of 20% water extracts of different parts of *A. nilotica* and *R. communis* were tested as antibacterial agent. The data depicted in Table 2, high inhibition zones were noted 13 and 10 mm against *E. coli* when water extract of both plants stems were applied, while water extract of different parts of both plants also showed good inhibition zones against both Gram positive and negative bacterial pathogens around 6 to 9 mm zones. Dabur et al. (2007) revealed that the water extract of *A. nilotica,* was found to be the most active against bacteria *E. coli*; *Salmonela typhi*, *P. aeruginosa, S. aureus*; *B. cereus*, *K. aerogenes*, *Proteus vulgaris* and *Shigella Boydii*.

However, methanol extract of *A. nilotica* showed significant activity against *E. coli* and *S. typhi*. Whilst in another report, *A. nilotica* exhibited highest activity against three bacterial strain *E. coli, S. aureus* and *S. typhi* (Siani et al., 2008). An antibacterial activity of *R. communis* seed against bacterial species by using disc diffusion method was reported by Jombo et al. (2008) .In

Table 2. Antibacterial activity of different parts of *Acacia nilotica* and *Ricinus communis from* 20% water extract.

***Acacia nilotica* water extract**			
Microorganisms	**Inhibition zones (mm)**		
Gram-positive	**Leaves**	**Stem**	**Bark**
Staphylococcus aureus	6.5	7	6
Streptococcus pneumoniae	8	9	9
Gram-negative			
Escherichia coli	8	10	6
K. pneumoniae	8	7	9
P. aeruginosa	6	7	6
***Ricinus communis* water extract**			
Gram-positive			
Staphylococcus aureus	7	8	N.T
S.Spneumoniae	9	8	N.T
Gram-negative			
Escherichia coli	9	13	N.T
K. pneumoniae	8	8	N.T
P. aeruginosa	6	7	N.T

N.T: not tested.

Table 3. Antifungal activity of different parts of *A. nilotica* and *R. communis* 20% methanol and water extract against some pathogenic fungus.

***A. nilotica* and *R. communis* methanol extract**			
Name of plant	***Aspergillus niger* (mm)**	***Penicillium expansum* (mm)**	***Aspergillus fumigates* (mm)**
A. nilotica leaves	Nil	Nil	Nil
A. nilotica stem	Nil	Nil	Nil
A. nilotica bark	Nil	Nil	Nil
R. communis leaves	Nil	Nil	Nil
R. communis stem	Nil	11	Nil
A. nilotica and *R. communis* water extract			
A. nilotica leaves	Nil	10	11
A. nilotica stem	Nil	Nil	Nil
A. nilotica bark	Nil	Nil	Nil
R. communis leaves	Nil	Nil	10
R. communis stem	6	11	Nil

this study, methanol and water extract of *R. communis* seed was checked against *K. pneumoniae, E. coli, P. vulgaris* and *S. aureus* which were highly susceptible to both methanol and water extract of seed. However, *P. aeruginosa* reduced susceptibility at some stage in this study. Banso (2009) investigated the phytochemical and antimicrobial activity of different parts of *A. nilotica* against bacterial species that is, *Streptococcus viridans*, *S. aureus*, *E. coli*, *B. subtilis* and *Shigella sonnei* by using agar diffusion method. In this report, *A. nilotica* extract showed significant activity against all tested microorganisms. However, *B. subtilis* was the most susceptible against plant extract while most resistance specie was *Candida albicans.* Therefore this study represented that *A. nilotica* could be a good source of antimicrobial agents. Furthermore, it is reported that the inhibition of tested microorganisms caused by the active contents, such as terpenoids, tannis, alkaloids, saponins and glycosides which are possessed in stem bark extract of plant. Different parts of both plants methanol extract did not show any antifungal activity against tested fungal species *A. niger*, *A. fumigatus* and *P. expansum*, only *R. communis* stem was found active against *P. expansum* 11 mm zone as shown in Table 3. Beyond this 20% water

Table 4. Antibacterial activity of standard antibiotic.

Name of antibiotic	*Escherichia coli* (mm)	*Pseudomonas aeruginosa* (mm)	*Klebsiella pneumoniae* (mm)	*Staphylococcus aureus* (mm)	*Streptococcus pneumoniae* (mm)
Ampiclox (30 µg/disc)	N.T	18	8	Nil	8
Exohabid (30 µg/disc)	24	42	18	32	18
Nystain (100 units/disc)	N.T	Nil	8	N.T	8
Erythromycin (5 µg/disc),	14	N.T	10	20	Nil
Fucidin (100 µg/disc	30	34	18	Nil	12
Romicef (30 µg/disc)	Nil	Nil	6	N.T	6
Cefspan (5 µg/disc)	Nil	Nil	8	Nil	8
Cefizox (30 µg/disc)	Nil	10	8	Nil	8

NT= not tested.

Table 5. Antifungal activity of standard antibiotic.

Name of antibiotic	*Penicillium expansum* (mm)	*Aspergillus niger* (mm)	*Aspergillus fumigates* (mm)
Ampiclox (30 µg/disc)	Nil	Nil	Nil
Exohabid (30 µg/disc)	N.T	Nil	Nil
Nystain (100 units/disc)	10	Nil	6
Efizox	Nil	Nil	Nil
Fucidin (100 µg/disc)	Nil	Nil	Nil
Romicef (30 µg/disc)	8	Nil	Nil
Cefspan (5 µg/disc)	Nil	Nil	6
Cefizox (30 µg/disc)	Nil	Nil	Nil

NT= not tested.

extract of different parts of *A. nilotica* leaves showed good inhibition zones against *A. fumigatus* 11 mm and *P. expansum* 10 mm whilst *R. communis* leaves and stems inhibited individually as *A. fumigatus* 10 mm and *P. expansum* 11 mm, respectively.

Any part of both plants water extract did not show effectiveness against *A. niger* except *R. communis* stem which exhibited minimum potency 6 mm as summarized in Table 3. Dabur et al., (2007) reported beyond than our results that *A. nilotica* was inactive against fungal pathogen *A. fumigatus*; *A. flavus*; *A. niger* and *C. albicans* but in our report it was observed that *A. nilotica* inhibited and active against *A. fumigatus* and *A. niger.* Siani et al. (2008) found similar results as our findings with two fungal strains *C. albicans* and *A. niger.* It was also observed that commercially available antibiotics used in this study as positive control as summarized in Tables 4 and 5, inhibited strongly against both Gram positive and negative bacteria and fungi in comparison to both plants extracts in some conditions. It is suggested that may be less concentration of plants extracts used in this study than standard antibiotics (conc. of each standard are given in Tables 4 and 5). Some bio-molecules were analyzed, identified and compared their correlation with antimicrobial activity of 20% methanol and water extract of different parts of *A. nilotica* and *R. communis* plants. The pH of all samples was also checked from each part of both plant extract. 20% water and methanol extract of different parts of *A. nilotica* and *R. communis* leaves, stem and bark exhibited slightly acidic in nature (around or below pH 6.1) as represented in Figure 1. The protein concentration from 20% water and methanol extract of different parts of *A. nilotica* and *R. communis* was recorded not more than 8.493 mg/ml as depicted in Figure 2. The maximum concentration of total sugar 18.34 mg/ml, and 12.294 mg/ml from water extracts of *A. nilotica* and *R. communis leaves* than other parts of same plant water extract, However, higher total sugar contents 12.214 mg/ml and 10.057 mg/ml from methanol extract of *R. communis* leaves and *A. nilotica* stem, respectively in comparison to other parts of both plants extracts Figure 3. The maximum reducing sugar contents from water and methanol extracts of *A. nilotica* leaves 4.486 mg/ml and 5.88 mg/ml were isolated, respectively in comparison to other parts of both plant extracts water and methanol as depicted in Figure 4. Some free amino acid and sugars from methanol and water extract of *A. nilotica* and *R. communis* leaves stem

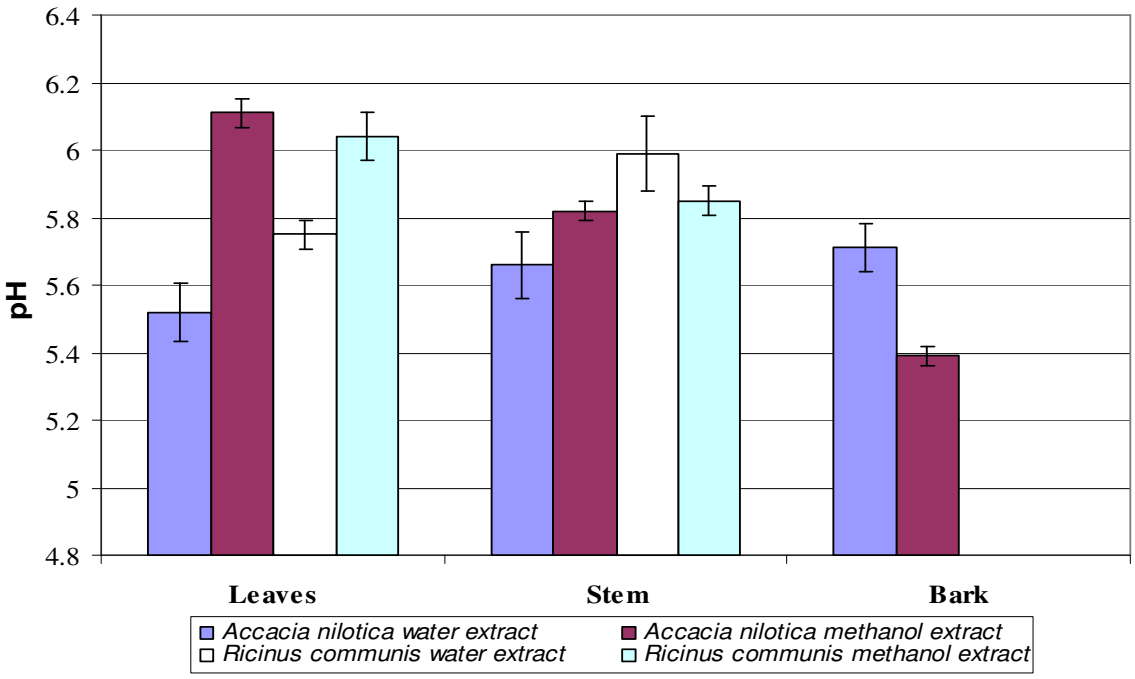

Figure 1. pH of different parts of *Acacia nilotica* and *Ricinus communis* 20% water and methanol extracts.

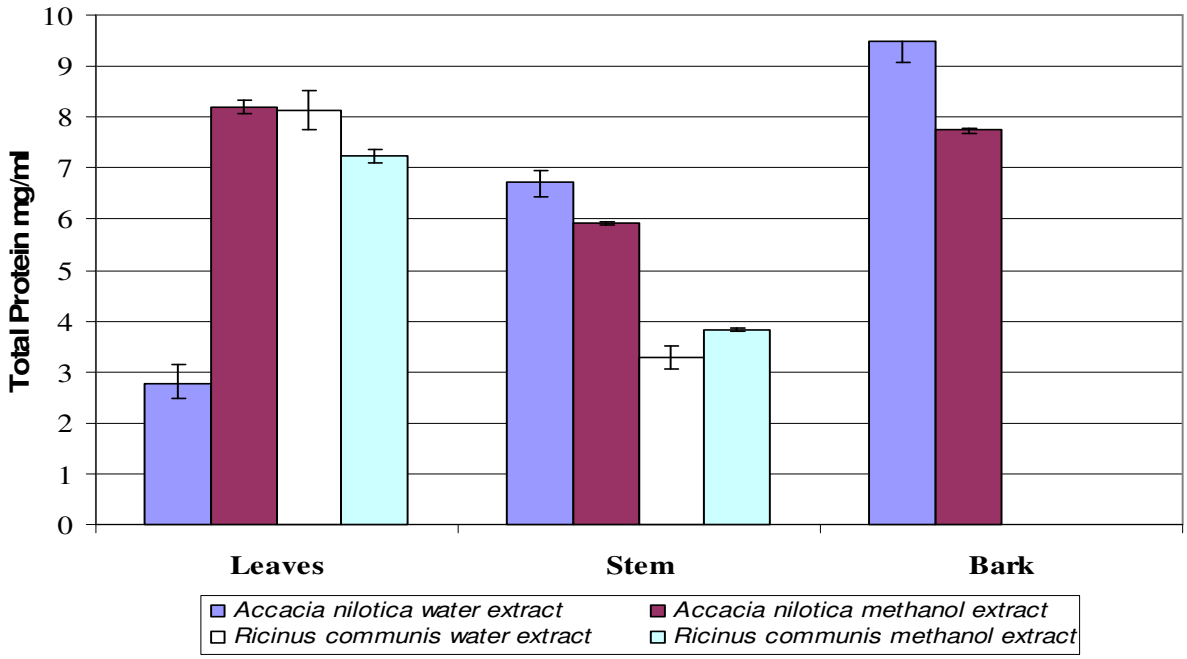

Figure 2. Total protein from different parts of *Acacia nilotica* and *Ricinus communis* 20%water and methanol extracts

and bark were identified through TLC.

The Rf values of different sample were matched with Rf value of standard amino acid and sugars. On the basis of identified free amino acids Table 6 and sugars Table 7, strongly confirms that *A. nilotica* and *R. communis* leaves, stem and bark extract consist of peptide / protein or sugar containing antimicrobial compound in both extracts. Barman and Rai (2006) have reported that these plants contain all the essential amino acids in good proportions comparable to egg protein. Furthermore,

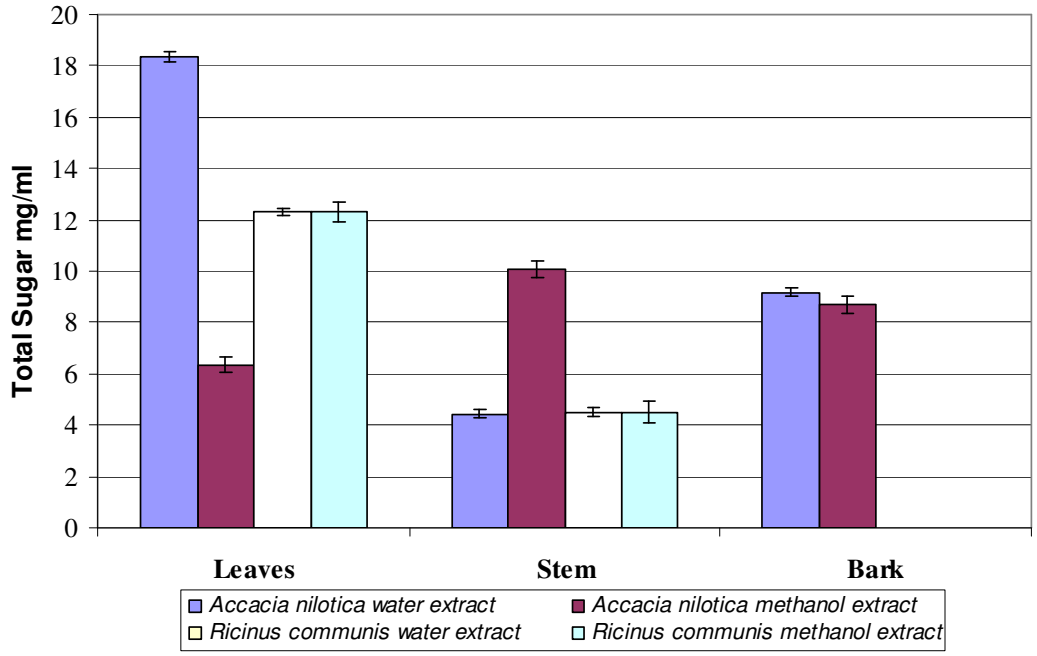

Figure 3. Total sugar from different parts of *Acacia nilotica* and *Ricinus communis* 20% water and methanol extracts.

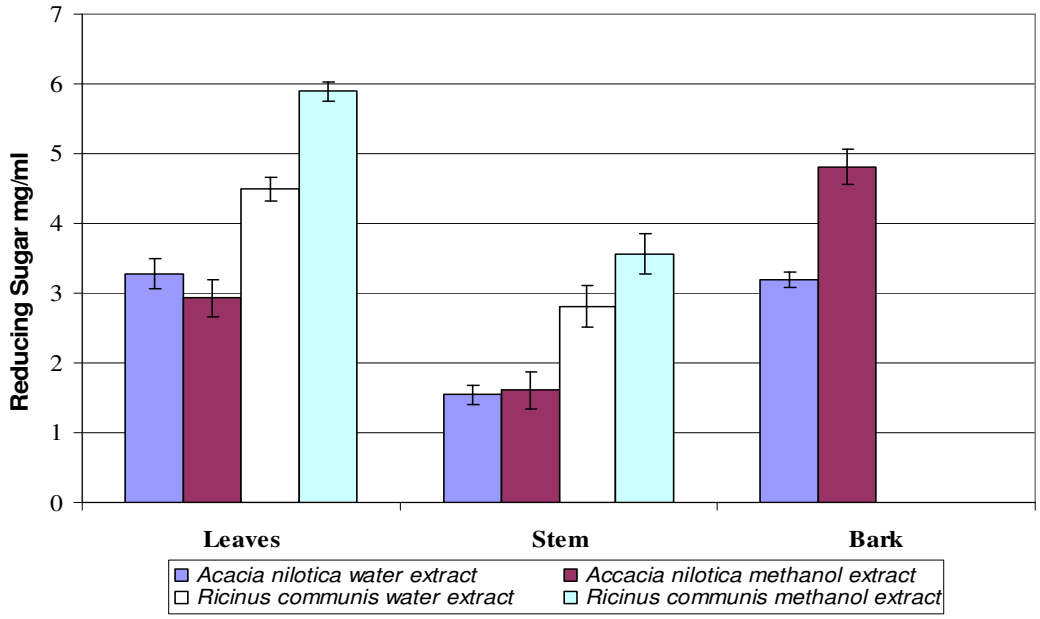

Figure 4. Reducing sugar from different parts of *Acacia nilotica* and *Ricinus communis* 20%water and methanol extracts.

antibiotic substances origin from plants exhibits to be more resistance against Gram positive than to Gram negative bacterial species. It is reported that some prominent antibiotic agents of fungal origin like penicillin are known to best in their inhibitory activity and most of them being inhibitory to Gram positive bacterial species. In comparison to Gram positive bacteria, Gram negative organisms have lipopolysaccharide layer with the combination of proteins and phospholipids, which are the major components of the outer surface of Gram negative

Table 6. Identification of free amino acid from different parts of *A. nilotica and R.communis* of 20%water and methanol extracts by TLC.

Name of plants	Standard of amino acid	Rf value of sample	Identification of amino acid
***A. nilotica* and *R. communis* water extract**			
Acacia nilotica leaves	0.628	0.628	Phenlyalalnine
	0.100	0.95	Aspartic acid
Acacia nilotica stem	0.252	0..257	Serine
	0.363	0.364	Asparagine
	0.439	0.442	Cystine
Acacia nilotica bark	0.100	0.138	Aspartic acid
	0.312	0.314	Hydroxyproline
	0.521	0.521	Tryptophane
	0..628	0.907	Unknown
Ricinus communis leaves	0.100	0.5	Un known
Ricinus communis stem	0.628	0.657	Un known
***A. nilotica* and *R. communis* methanol extract**			
Acacia nilotica leaves	0.496	0.485	Leucine
Acacia nilotica stem	0.363	0.357	Asparagine
Acacia nilotica bark	0.312	0.312	Hydroxyproline
	0.628	0.907	Unknown
Ricinus communis leaves	0.496	0.492	Leucine
	0.628	0.707	Unknown
	0.628	0.592	Unknown
Ricinus communis stem	----------	-------	--------

Table 7. Identification of carbohydrates from different parts of *A. nilotica and R. communis* of 20%water and methanol extracts by TLC.

Name of plants	Standard of carbohydrate		Rf value of sample	Identification of carbohydrate
***A. nilotica* and *R. communis* water extract**				
Acacia nilotica leaves	Lactose	0.849	0.850	Lactose
	Glucose	0.938	0.903	Unknown
	Fructose	0.938	1.026	Unknown
Acacia nilotica stem	Ribose	0.319	0.526	Unknown
Acacia nilotica bark	Glucose	0.938	0.957	Unknown
	Fructose	0.938		
Ricinus communis leaves	Ribose	0.319	0.518	Unknown
	Maltose	0.885	0.885	Maltose
Ricinus communis stem	Lactose	0.849	0.684	Unknown

Table 7. Contd.

***A. nilotica* and *R. communis* methanol extract**				
Acacia nilotica leaves	Glucose	0.938	----------	----------
	Fructose	0.938		
Acacia nilotica stem	Ribose	0.319	0.342	Unknown
Acacia nilotica bark	Fructose	0.938	0.938	Fructose
	Glucose	0.938	0.938	Glucose
Ricinus communis leaves	Ribose	0.319	0.482	Unknown
Ricinus communis stem	Lactose	0.849	0.849	Lactose

bacteria (Burn, 1988). Access of most compounds to the peptidoglycan layer of the cell wall is hindered by the outer lipopolysaccharide layer. This explains the resistance of Gram negative bacterial strains to the lytic action of most plant extracts exhibiting activity. Phytochemical analysis provides strong circumstantial evidence that small proteins/peptides play a key role as an antimicrobial defense system in plants (Terras et al., 1995). This defense system of plants provide protection them against potential microbial invaders. Among these other plants secreted compounds several proteins or peptides and carbohydrates have a specific defense mechanism against phytopathogenic microbes by inhibiting their growth.

Hence, microbial growths cease through diverse molecular modes and bind to chitin or increasing the permeability of the fungal membrane or cell wall (Van-Etten et al., 1989; Maher et al., 1994). Their active mechanism may be the formation of ion channels in the microbial membrane (Terras et al., 1993; Zhang and Lewis, 1997) or competitive inhibition of adhesion of microbial proteins to host polysaccharide receptors (Sharon and Ofek, 1986) while another strategy of plants defense mechanism to thwart microbial invaders on the basis of secretion of antimicrobial low molecular weight secondary metabolites known as phytoalexins (Van-Etten et al., 1989; Maher et al., 1994). Furthermore, the synthesis of many presumed defense related proteins are induced when plants are confronted with pathogens (Linthorst, 1991). Many other workers also revealed that the monosaccharide fructose present in cranberry and blueberry juices competitively inhibited the adsorption of pathogenic *E. coli* to urinary tract epithelial cells, acting as an analogue for mannose (Avorn, 1996). Many fruits also contain fructose; however, researchers are now seeking a second active compound from cranberry juice which contributes to the antimicrobial properties of this juice (Zafriri et al., 1987). The exceptional water-soluble compounds, such as polysaccharides for example, starch and polypeptides, including fabatin and various lectins, are commonly more effective as inhibitors of pathogen (Zhang and Lewis, 1997).

Conclusions

This study was revealed the preliminary investigation on antimicrobial efficacy of different parts of *A. nilotica* and *R. communis* against certain human pathogenic microorganisms and their correlation with bio-molecules as microbial agent. Antimicrobial Activity from methanol and water extract of different part of *A. nilotica and R. communis* (leaves stem and bark) showed significant inhibition against both Gram-positive and negative bacteria. 20% methanol extract of leaves of both plants *A. nilotica* and *R. communis* exhibited highest zone of inhibition 13 and 11 mm against *E. coli* and *S. penumoniae,* respectively. Whilst water extract showed strong zone of inhibition in the range of 14 and 12 mm against *S. aureus* and *S. penumoniae,* respectively. Methanol and water extract also showed antifungal activity against selected fungus species. Some biomolecules were also analyzed identified from 20% methanol and water extract of different part of *A. nilotica* (leaves, stem and bark) and *R. communis* (leaves and steam). The highest total protein 8.493 mg/ml from methanol extract of *A. nilotica* leaves, maximum total sugar 18.340 mg/ml from methanol extract of *A. nilotica* leaves, while highest reducing sugar 5.88 mg/ml from methanol extract of *R. communis* were obtained. Free Amino acid and sugar were also identified through TLC, which is strongly indicates that both plants extract consist of peptide/ protein or sugar containing antimicrobial compound in both extracts.

AKNOWLEDGEMENT

The authors would like to thank the University of Sindh, Jamshoro, Pakistan for their support in this research work.

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