RESEARCH ARTICLE PHARMACOLOGIA

DOI: 10.5567/pharmacologia.2014.199.204

Antibacterial Activity of Methanol Extracts and Fractions from *Kalanchoe crenata, Terminalia avicennioides* and *Sarcocephalus latifolius*

[1]Foutse Yimta, [2]Mouokeu Raymond Simplice, [1]Nguimatsia François, [3]Njateng Guy Sedar Singor, [3]Tamokou Jean de Dieu and [3]Kuiate Jules Roger

[1]Laboratory of Pharmacognosy, University of Montains, P.O.Box 208, Bangangte, Cameroon

[2]Laboratory of Microbiology and Food Quality Control, Institute of Fisheries and Aquatic Sciences, University of Douala, P.O.Box 7236, Douala, Cameroon

[3]Laboratory of Microbiology and Antimicrobial Substances, Faculty of Sciences, P.O.Box 67, Dschang, Cameroon

ABSTRACT

Background: Alternatives to available antibiotics for disease management are increasingly felt due to the increase in the resistance of bacterial strain. Plants are known to be a rich source of medicines because they produce wide array of bioactive molecules. The present study was undertaken to investigate the antibacterial properties of the methanol extract bark and leaves of *Kalanchoe crenata* (Crassulaceae), *Terminalia avicennioides* (Combretaceae) and *Sarcocephalus latifolius* (Rubiaceae). **Materials and Methods:** The crude extracts were prepared by maceration of plant powder in methanol. *K. crenata* extract was further partitioned into hexane, ethyl acetate and residue fractions. *T. avicennioides* extract was also fractionated by flash chromatography into eight fractions named F_c to F_j. Phytochemical tests were carried out on the extracts and fractions using standard methods. The antibacterial activity of the crude extracts and fractions were evaluated by broth microdilution method. **Results:** The phytochemical tests indicate that all tested extracts contained phenols, tannins, flavonoids and other classes of chemicals were selectively present. The antibacterial susceptibility test showed the best spectra of activity with *T. avicennioides* extract (MIC = 0.1-0.4 mg mL^{-1}), followed by *S. latifolius* (MIC= 0.2-0.8 mg mL^{-1}). *K. crenata* extract was found to be less active (MIC= 0.8-1.6 mg mL^{-1}). F_I and F_J fractions were found to have similar antibacterial activity relatively greater than F_G fraction. The highest activity of those fractions was achieved on *P. mirabilis* (MIC =0.02 mg mL^{-1}) where conventional antibiotic ciprofloxacin failed to be active. **Conclusion:** The overall results highlighted the antibacterial activity of *T. avicennioides and S. latifolius*. This constitutes a power tool for the investigation of *T. avicennioides and S. latifolius* extract for the preparation of phytomedicine against bacterial diseases as well as the isolation of active ingredient from T. *avicennioides* methanol extract.

Key words: *Kalanchoe crenata, Terminalia avicennioides, Sarcocephalus latifolius,* antibacterial activity

Pharmacologia 5 (6): 199-204, 2014

INTRODUCTION

There is a progressive increase in antibiotic resistant strains of clinically important pathogens (Djeussi *et al*., 2013; Sen and Batra, 2012). Despite the advancement in science and technology on the discovery of many natural and synthetic drugs, infectious diseases are still the leading cause of morbidity and death, especially in developing countries (Barre-Sinoussi, 2009; Vargas *et al*., 2003). The outlook for the use of antimicrobial drugs in the future is still uncertain. Alternative actions must be taken as to reduce the incidence of conventional therapeutic failure to antimicrobial treatments. Among the potential sources of new agents, plants have long been investigated. They are known to produce a variety of compounds to protect themselves against a variety of pathogens (Satish *et al*., 2008). These compounds have been associated with the used of some plants in folk medicine in the treatments of a variety of diseases. Large evaluation of such plants for various biological activities is a prerequisite in the isolation and characterization of the active ingredients and further the development of biomedicine. *Kalanchoe crenata*, *Terminalia aviceniodes* and *Sarcocephalus latifolius* are plants belonging to Cameroon flora. They are well known for therapeutic usage by local populations. *Kalanchoe crenata* is used to treat asthma, ocular affection and otitis. *Terminalia aviceniodes* is used to treat syphillis, wounds, gastric ulcer (Arbonier, 2005). *Sarcocephalus latifolius* is antimitotique, purgative and vermifuge. Earlier studies on *K. crenata* reported the antibacterial activity of the leaves palm-wine and local gine extract (Akinsulire *et al*., 2007). Analgesic, anticonvulsion, anti-inflammatory, antiarthritic and

Corresponding Author: Mouokeu Raymond Simplice, Laboratory of Microbiology and Food Quality Control, Institute of Fisheries and Aquatic Sciences, University of Douala, P.O. box 7236, Douala, Cameroon, Mouokeu, Tel: +23777442528/+23791914849

© 2014 Science Reuters, UK

antispasmolytic properties were also reported (Dimo *et al*., 2006). Previous studies on *T. aviceniodes* revealed the safety usage of the aqueous extract (Bulus *et al*., 2011) as well as the anti-malarial activity (Omonkhua *et al*., 2013). As a contribution to search of the antibacterial activities from plants, we designed the present work to determine the activity of three selected Cameroonian plants, *Kalanchoe crenata*, *Terminalia aviceniodes* and *Sarcocephalus latifolius* against gram positive and gram negative bacteria of clinically importance.

MATERIALS AND METHODS

Plant material: *K. crenata* was collected in the arboretum of the University of Mountains (Bangangte, West region of Cameroon). *T. avicennioides* and *S. latifolius* stem bark were harvested in Noun road (Bangangte). Botanical identification was carried out at the Cameroon National Herbarium by referring to the voucher specimen 35196/HNC, 7908/SRFCAM and 4492/SRFK, respectively for *K. crenata, Terminalia avicennioides* and *Sarcocephalus latifolius*.

Microorganisms: Microorganisms used included two strains of gram positive bacteria (*Staphylococcus aureus* ATCC 25922 and *Enterococcus faecalis* ATCC 10541), four strains of gram negative bacteria (*Pseudomonas aeruginosa* ATCC 27853, *Escherichia coli* ATCC 11775, *Salmonella enterica serovar thypi* ATCC 6539, *Klebsiella pneumoniae* ATCC 13883) and clinical isolates of *Proteus mirabillis* and *Shigella flexneri.* These isolates were a gift from the Pasteur Institute (Cameroon).

Preparation of crude extracts: The stem bark of *T. avicennioides* and *S. latifolius* was dried at laboratory temperature (20±2°C) for 20 days and powdered to coarse particles. Three kilograms of each powdered and fresh crushed leaves of *K. crenata* were separately macerated in 9 L methanol for 48 h with frequent stirring. The homogenate was then filtered and concentrated under reduced pressure using rotary evaporator (Buchi R2000) at 50°C to yield crude extract of 10.80% (*K. crenata*), 11.80% (*S. latifolius*) and 21.22% (*T. avicennioides*).

Fractionation of the crude extract and phytochemical screening: *T. avicennioides* extract was fractionated using flash chromatography. In so doing, 130 g extract were fixed on 200 g silica gel grade 60 and then placed on a chromatography column (25 mm diameter and 30 cm high) containing silica gel. The elution gradient was successively mixtures of hexane-ethyl acetate (100-00; 80-20; 70-30; 60-40; 50-50; 40-60; 30-70; 20-80; 0-100) and ethyl acetate-methanol (50-50; 0-100). Eighteen fractions (volume = 250 mL each) were collected and concentrated using a rotary evaporator under reduced pressure at 50°C. These fractions were further grouped based on their thin layer chromatographic profile into eight fractions named F_c to F_j.

The *K. crenata* extract was partitioned with hexane and ethyl acetate, respectively. 100 g extract were homogenized in 500 mL of hexane. The mixture was then stirred for 5 min and allowed to stand for one hour. The upper phase (hexane fraction) was recovered. The extract residue was dried at laboratory temperature and treated in same conditions with ethyl acetate to give the ethyl acetate fraction and the residue. For each solvent, extraction was made three times and fractions were concentrated separately under vacuum at 50°C using a rotary evaporator.

The phytochemical analysis was done on the extracts and fractions following standards methods (Cowan, 1999).

Antibacterial susceptibility testing: The antibacterial activity of the crude extracts and fractions was evaluated by broth microdilution method. Stock cultures of microorganisms were maintained at 4°C on slopes of Muller Hinton agar (Conda, Madrid, Spain). Inocula suspension were prepared from an overnight culture by transferring of loopful of cells from overnight culture to distilled water and adjusted to 0.5 Mc Farland turbidity standard, corresponding to 1.5×10^8 CFU/mL. Antibacterial activity was assayed using Mueller Hinton broth culture medium (Conda, Madrid, Spain) in 96 microtitre plates. Briefly, the stock solution of crude extract and fractions was dissolved in 5% DMSO. 100 μL of broth culture medium was introduced in each well of a 96 microtitre plates. Serials two fold dilutions were made with solutions of crude extracts or fractions, prepared at 128 mg mL^{-1} (crude extract) and 3.2 mg mL^{-1} (fractions). One hundred microlitters of inoculum diluted 100 times were further introduced in each well. The plates were further incubated at 37°C for 24 h. After the incubation period, growth was monitored calorimetrically using iodotetrazolium chloride (INT). Viable bacteria change the yellow dye of P. iodonitrotetrazolium violet to pink color. For a given crude extract or fraction, the smallest concentration at which no visible color change was noticed was considered as the Minimum Inhibitory Concentration (MIC) (Djeussi *et al*., 2013).

The bactericidal concentrations were determined by subculture 10 μL of the well which did not show any

© 2014 Science Reuters, UK

visible change after the incubation during MIC assays. The plates were further incubated at 37°C for 24 h. For each crude extract or fraction, the smallest concentration where no growth was recorded was considered as MBC. All the assays were carried out in triplicate. Ciprofloxacin was used as positive control where as 5% DMSO was used as negative control (Kognou *et al*., 2011; Salie *et al*., 1996).

RESULTS

Phytochemical analysis of extracts and fractions: The results of the preliminary phytochemical studies reported in Table 1 indicate that the *S. latifolius* extract contains flavonoids, phenols, saponins and tannins. *K. crenata* and *T. avicennioides* extracts contain phenols and tannins. The hexane fraction of *K. crenata* contains only sterols. Moreover, the phytochemical composition of the ethyl acetate fraction and the residue of *K. crenata* were close to the crude extract. Fractions F_C, F_D, F_E, F_F and F_G resulting from *T. avicennioides* extract fractionation were more complex in their phytochemical composition and were closed to the crude extract.

Antibacterial activity: S. latifolius, *K. crenata* and *T. avicenioides* methanol extract revealed antibacterial activity on studied bacteria that varied with the plant extract (Table 2). *Terminalia aviceniodes* extract (MIC= 0.1-0.4 mg mL^{-1}) was found to be more active compared to *S. latifolius* (MIC= 0 .2-0.8 mg mL^{-1}). *K. crenata* extract was found to be less active (MIC= 0.8-1.6 mg mL^{-1}) with Gram + bacteria having the highest MIC values. Partition of *K. crenata* extract reduced the activity of fractions on the studied bacteria.

P. mirabilis, *E. coli* and *S. flexneri* were more sensitive to *K. crenata* extract while *S. typhi* and *E. coli were* more sensitive to *S. latifolius* extract. *S. typhi*, *P. aeruginosa* and *E. coli* were more sensitive *T. avicennioides* extract.

The activity of *T. avicennioides* extract was compared to its fractions (Table 3). It was realized that F_G, F_J and F_I are the active fractions. Compared to other fractions, fraction F_G was found to be inactive against *S. flexneri*. F_I and F_J fractions were found to have similar antibacterial activity relatively greater than F_G fraction. The highest activity was achieved on *P. mirabilis* (MIC =0.02 mg mL^{-1}) where conventional antibiotic ciprofloxacin failed to be active.

MICs values of *S. latifolius*, *K. crenata* and *T. avicennioides* extracts as well as *T. avicennioides* fractions were four fold less than the MBC values, indicating that the bactericidal effect of these extracts could be expected.

DISCUSSION

Each of the extract tested in the present study displayed antibacterial activity on the bacterial strains tested. This suggests that these extract possess broad spectrum activities. These results correlate with the observation of previous workers that plants contain substances that are antimicrobial (Kuete, 2010; Olukoya *et al*., 1986). However, differences were observed between their antibacterial activities. These differences could be due to the differences in the chemical composition of these extracts as revealed by phytochemical analysis.

The antibacterial activity of *K. crenata* extract was found to be moderate in almost all the tested bacteria including *S. aureus* (MIC= 1.6 mg mL^{-1}). These data contrast with previous results on the plant which revealed the activity of the methanol extract only on *Staphylococcus aureus* with MIC value of 11.10 mg L^{-1} (Kablan *et al*., 2008), but they are in accordance with other results where both the aqueous and methanol dry leaves extract of *K. crenata* had moderate antibacterial activity with MIC values ranging from 8 to 128 mg mL^{-1} (Akinsulire *et al*., 2007). The differences in the antibacterial activity could be explained by either or both qualitative and quantitative difference in phytochemical composition, due to the environmental conditions during plant growth. This therefore poses the problem of standardization of plant extract for therapeutic usage because such variation could

Table 1: Phytochemical screening of extracts and fractions of *S. latifolius*, *K. crenata* and *T. avicenioides*

		K. Crenata			*T. avicennioides*								
Chemical class	*S. latifolius*	Extract	Hex- F	*Residue*	Extract	F_C	F_D	F_E	F_F	F_G	F_H	F_I	F_J
Alkaloïds	-	+	-	+	+	-	+	+	-	-	-	-	-
Anthraquinones	-	-	-	-	+	-	-	-	-	-	-	+	+
Flavonoïdes	+	+	-	+	+	-	-	+	+	+	+	+	+
Phenols	+	+	-	+	+	-	-	-	+	+	+	+	+
Saponins	+	-	-	-	+	-	-	-	-	-	+	+	+
Sterols	-	+	+	-	-	-	-	-	-	-	-	-	-
Tanins	+	+	-	+	+	-	-	-	-	+	+	+	+
Triterpens	-	-	-	-	+	+	+	+	-	-	-	-	-
Coumarins	-	+	-	+	-	-	-	-	-	-	-	-	-

Hex-F: Hexane fraction

© 2014 Science Reuters, UK

Table 2: Minimum Inhibitory Concentrations (MIC), Minimum Bactericidal Concentrations (MBC) and MBC/MIC ratio of extracts of *S. latifolius*, *K. crenata* and *T. avicennioides* fractions

	K. crenata																				
	Crude extract			Hex			EtAC			Residue			*S. latifolius*			*T. avicennioides*			Ciprofloxacin		
	CMI	CMB	CMB/CMI	CMI	CMB	CMB/CMI	CMI	CMB	CMB/CMI	CMI	CMB	CMB/CMI	CMI	CMB	CMB/CMI	CMI	CMB	CMB/CMI	CMI	CMB	CMB/CMI
Gram negative																					
P. aeruginosa	3.2	3.2	1	0.8	0.8	1	3.2	3.2	1	1.6	3.2	2	0.4	0.8	2	0.2	0.4	2	1.0	1	1
P. mirabilis	0.8	3.2	4	1.6	1.6	1	1.6	3.2	2	1.6	3.2	2	0.8	0.8	1	0.4	0.4	1	16.0	16	1
K. pneumonia	3.2	3.2	1	3.2	3.2	1	3.2	3.2	1	1.6	3.2	2	0.8	0.8	1	0.4	0.8	2	1.0	1	1
S. enterica serovar thypi	1.6	3.2	2	3.2	3.2	1	1.6	3.2	2	1.6	3.2	2	0.2	0.2	1	0.1	0.2	2	4.0	4	1
E. coli	0.8	3.2	4	3.2	3.2	1	0.8	3.2	4	0.8	3.2	4	0.2	0.4	2	0.2	0.4	1	0.5	1	2
S. flexneri	0.8	3.2	2	>3.2	-	-	1.6	3.2	2	1.6	3.2	2	0.8	0.8	1	0.4	0.4	1	8.0	8	1
Gram positive																					
S. aureus	1.6	3.2	2	>3.2	-	-	1.6	3.2	2	1.6	3.2	2	0.4	0.4	1	0.4	0.4	1	8.0	8	1
E. faecalis	1.6	3.2	2	3.2	3.2	1	1.6	3.2	2	1.6	3.2	2	0.4	0.4	1	0.4	0.4	1	4.0	16	4

-: Undetermined, Hex: Hexane fraction, EtAC: Ethyl acetate fraction, concentration of extracts and fractions are expressed in mg mL^{-1}

Table 3: Minimum inhibitory concentration (CMI) and minimum bactericidal concentrations of *T. avicinnioides* extract and fractions

	Extract			F_G			F_H			F_I			F_J			Ciprofloxacin ($\mu g\ mL^{-1}$)		
	CMI	CMB	CMB/CMI	CMI	CMB	CMB/CMI	CMI	CMB	CMB/CMI	CMI	CMB	CMB/CMI	CMI	CMB	CMB/CMI	CMI	CMB	CMB/CMI
Gram negative																		
P. aeruginosa	0.2	0.4	2	0.2	0.4	2	0.05	0.4	8	0.2	0.4	2	0.2	0.4	2	1.0	1	1
P. mirabilis	0.4	0.4	1	0.1	0.4	4	0.8	>0.8	-	0.02	0.4	16	0.02	0.4	16	16.0	16	1
K. pneumonia	0.4	0.8	2	0.2	>0.8	-	>0.8	-	-	0.2	0.8	4	0.2	0.8	4	4.0	4	1
S. typhi	0.1	0.2	2	0.8	>0.8	-	>0.8	-	-	0.4	0.8	2	0.4	0.8	2	0.5	1	2
E. coli	0.2	0.4	1	0.4	0.4	1	>0.8	-	-	0.4	0.8	2	0.4	0.8	2	1.0	1	1
S. flexneri	0.4	0.4	1	>0.8	>0.8	-	>0.8	-	-	0.4	>0.8	-	0.4	>0.8	-	8.0	8.	1
Gram positive																		
S. aureus	0.4	0.4	1	0.2	0.8	4	>0.8	-	-	0.4	>0.8	-	0.4	>0.8	-	8.0	8	1
E. faecalis	0.4	0.4	1	0.1	0.8	8	>0.8	-	-	0.2	0.4	2	0.2	0.4	2	4.0	16	4

- : Undetermined, Fractions F_C, F_D, F_E and F_F was found to be inactive, concentration of extracts and fractions are expressed in mg mL^{-1}, *S. typhi*: *Salmonella enterica serovar typhi*

© 2014 Science Reuters, UK

be the cause of therapeutic failure when the whole extract is used to treat a particular disease.

The various antimicrobial activities of *S. latifolius*, *K. crenata* and *T. avicennioides* extract as shown from the result of this study, confirms their use traditionally in treating antimicrobial infections.

The antibacterial activity of the three fractions from the *K. crenata* crude extract was found to be lesser, indicating unnecessary fractionation of this extract as to improve the antibacterial activity.

The fractionation of *T. avicennioides* extract improved the antibacterial activity in the fractions F_J and F_I on *P. mirabilis* where ciprofloxacin, a well known broad spectrum antibacterial agent fail to be active. This result shows that the fractionation process concentrated the active principle in those fractions. These fractions are best candidate for the treatment of diseases associated with these microorganisms than the crude extract. Similar results were reported by Mansouri *et al*. (2001) when evaluating the antibacterial activity of the crude extracts and fractionated constituents of *Myrtus communis*. The results of this study do not only show the scientific basis for some of the therapeutic uses of *T. avicennioides* plant in traditional medicine, but also confirms the impact of ethno botanical approach when investigating plants for their antimicrobial properties (Adesanya, 2005; Iwu, 1993).

Some antimicrobial extracts from plant fail to be active upon fractionation. The result thus obtained in *T. aviceniodes* fractions highlight the fact that a particular compound can be responsible for the antibacterial activity in the plant. This is an interesting tool for the isolation and purification of the active ingredient for therapeutic purpose.

Differences in sensibility among strains were observed in the plants extracts and fractions. This could be due to their genetic content and it is an evidence for the necessity of antibiogram prior to antimicrobial prescription. It's particularly important because inappropriate antimicrobial drugs enhance microbial resistance (Escalante *et al*., 2002).

CONCLUSION

The results of the present study support the traditional use of the studied plants in the treatment of bacterial infections. They also provide an important basis for the use of methanol extract of the plants used to control infectious diseases caused by Gram-negative and positive bacteria.

ACKNOWLEDGMENT

Authors are thankful to the University of Mountains for his financial support and the Cameroon National Herbarium (Yaounde) for plants identification.

REFERENCES

Adesanya, S.A., 2005. From Nature to Drugs: Theories and Realities: An Inaugural Lecture Delivered at Oduduwa Hall, Obafemi Awolowo University, Ile-Ife, Nigeria, on Tuesday 12th July, 2005. Obafemi Awolowo University Press, Osun State, Nigeria.

Akinsulire, O.R., I.E. Aibinu, T. Adenipekun, T. Adelowotan and T. Odugbemi, 2007. *In vitro* antimicrobial activity of crude extracts from plants *Bryophyllum pinnatum* and *Kalanchoe crenata*. Afr. J. Traditional Complementary Altern. Med., 4: 338-344.

Arbonier, M., 2005. Arbres, Arbustes et Lianes Des Zones Seches d'Afrique de l'Ouest. 1st Edn., CIRAD, National Museum of Natural History, France, Pages: 541.

Barre-Sinoussi, F., 2009. Globalization and infectious diseases: A challenge announced by the emergence of HIV/AIDS. Proceedings of the Solemn Meeting of the Academy of Science, June 16, 2009, France, pp: 1-5.

Bulus, T., SE. Atawodi and M. Mamman, 2011. Acute toxicity effect of the aqueous extract of *Terminalia avicennioides* on white albino rats. Sci. World J., Vol. 6.

Cowan, M.M., 1999. Plant products as antimicrobial agents. Clin. Microbiol. Rev., 12: 564-582.

Dimo, T., A.L. Fotio, T.B. Nguelefack, E.A. Asongalem and P. Kamtchouing, 2006. Antiinflammatory activity of leaf extracts of Kalanchoe crenata Andr. Indian J. Pharmacol., 38: 115-119.

Djeussi, D.E., J.A.K. Noumedem, J.A. Seukep, A.G. Fankam and I.K. Voukeng *et al*., 2013. Antibacterial activities of selected edible plants extracts against multidrug-resistant gram-negative bacteria. BMC Complementary Alternat. Med., Vol. 13. 10.1186/1472-6882-13-164

Escalante, M.A., C.B. Santecchia, N.S. Lopez, M.A. Gattuso and G.A. Ravelo *et al*., 2002. Isolation of antifungal saponins from *Phytilacca tetramera*, an argentina species in critic risk. J. Ethnopharma., 82: 29-34.

Iwu, M.M., 1993. Handbook of African Medicinal Plants. 1st Edn., CRC Press, Boca Raton, FL., ISBN-10: 084934266X, pp: 464.

Kablan, B.J., M. Adiko and D.P. Abrogoua, 2008. Evaluation *in vitro* de l'activite antimicrobienne de *Kalanchoe crenata* et de *Manotes longiflora* utilisees dans les ophtalmies en Cote d'Ivoire. Phytotherapie, 6: 282-288.

Kognou, A.L.M., R.A.N. Ngane, J.R. Kuiate, M.K. Mogtomo and A.T. Tiabou *et al*., 2011. Antibacterial and antioxidant properties of the methanolic extract of the stem bark of *Pteleopsis hylodendron* (Combretaceae). Chemother. Res. Pract. 10.1155/2011/218750.

© 2014 Science Reuters, UK

Kuete, V., 2010. Potential of Cameroonian plants and derived products against microbial infections: A review. Planta Medica, 76: 1479-1491.

Mansouri, S., A. Foroumadi, T. Ghanei and A.G. Najar, 2001. Antibacterial activity of the crude extracts and fractionated constituents of *Myrtus communis*. Pharm. Biol., 39: 399-401.

Olukoya, D.K., T.O. Odugbemi and S.O.A. Bamgbose, 1986. Some aspects of traditional therapy of Gonorrhoea in Lagos, Nigeria. J. Res. Ethno-Medicine., 1: 26-29.

Omonkhua, A.A., M.C. Cyril-Olutayo, O.M. Akanbi and O.A. Adebayo, 2013. Antimalarial, hematological and antioxidant effects of methanolic extract of *Terminalia avicennioides* in *Plasmodium* berghei-infected mice. Parasitol. Res., 112: 3497-3503.

Salie, F., P.F.K. Eagles and H.M.J. Leng, 1996. Preliminary antimicrobial screening of four South African Asteraceae Species. J. Ethanopharmacol., 52: 27-33.

Satish, S., M.P. Raghavendra and K.A. Raveesha, 2008. Evaluation of the antibacterial potentials of some plants against human pathogenic bacteria. Adv. Biol. Res., 2: 44-48.

Sen, A. and A. Batra, 2012. Evaluation of antimicrobial activity of different solvent extracts of medicinal plant: *Melia azedarach*. Int. J. Curr. Pharma. Res., 4: 67-73.

Vargas, M.L.Y., M.V. Castelli, V.V. Kouznetsov, G.J.M. Urbina and S.N. Lopez *et al*., 2003. *In vitro* antifungal activity of new series of Homoallylamines and related compound with inhibitory properties of the synthesis of fungal cell wall polymers. Bioorganic Med. Chem., 11: 1531-1550.

© 2014 Science Reuters, UK