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Full Length Research Paper

Efficacy of crude extract and fractions of *Citrullus lanatus* against some selected micro-organisms

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Multiple resistances to antimicrobial agents in recent years compel the search for newer agents from natural sources (herbal options). To evaluate the antimicrobial activities of methanol extract and fractions of *Citrullus lanatus* leaves. The leaves of *C. lanatus* were successively extracted with methanol and fractioned into n-hexane, chloroform, ethylacetate and n-butanol fractions respectively. Antimicrobial activity of the crude extract and respective fractions of *C. lanatus* leaves was evaluated against clinical isolates including, *Streptococcus pyogenes*, *Streptococcus faecalis*, *Bacillus cereus*, *Corynebacterium ulcerans, Escherichia coli, Klebsiella pneumoniae, Pseudomonas flourescens, Salmonella typhi, Candida albicans* and *Candida krusei* using agar diffusion and broth dilution methods. Susceptibility test of all the fractions revealed significant activity against the test microbes. Ethylacetate fraction exhibited the highest activity with inhibition range of 26 – 29 mm and the methanol extract showed the least activity with inhibition range of 16 – 18 mm against all the test organisms except *S. pyogenes, C. ulcerans, K. pneumonia* and *S. typhi*. The Minimum Inhibitory Concentration (MIC) range was 1.25 – 5 mg/ml and the Minimum Bactericidal/Fungicidal Concentration (MBC/MFC) range was 2.5 – 10 mg/ml against the susceptible organisms. Antimicrobial potency of the extract and fractions was in the order; EF> CF> BF>HF>ME. The result of this research suggests that the leaves of *C. lanatus* have a remarkable antimicrobial activity which validates the ethno medicinal claim.

Key words: *Citrullus lanatus*, phytochemical screening, antimicrobial, MIC, MBC/MFC.

INTRODUCTION

Microbial infection is the most burdensome infectious disease in many countries, leading to about 14 - 17 million deaths annually (Yusuf et al., 2015). Pharmacological industries have produced a number of new antibiotics in the last three decades; however, resistance to these antibiotic drugs by microorganisms is still on the increase. The problem of microbial resistance is growing and the outlook for the use of antimicrobial drugs in the future is still uncertain (Gislene et al., 2000). Hence, the clarion calls for a renewed effort in the search for new antimicrobial agents. Nature has been a good source of many modern antimicrobial agents; most of these modern drugs have been obtained from medicinal plants. Extracts from plants are continuously being sort for the treatment of microbial infection and other diseases because they are effective, cheaper and can be easily accessed by most populations.

Indigenous to Southern Africa (Robinson and Decker-Walters, 1997), *Citrullus lanatus* commonly known as watermelon belongs to the *Curcurbitaceae* family. The plant produces a fruit that is 93% water, hence the name “watermelon”. *C. lanatus* is employed in traditional

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Table 1. Bioactivity studies conducted on *Citrullus lanatus.*

S/NO	Bioactivity	Plant part	References
1	Laxative	Fruit pulp	Swanpil et al., 2011
2	Antimicrobial	Stem, fruits and seeds	Loiy et al., 2011
3	Antigardial	Fruits	Loiy et al., 2011
4	Anti-inflammatory	Seeds and seed oil	Gill et al., 2010
			Madhavi et al., 2012
5	Antioxidant	Seeds	Gill et al., 2010
			Naresh, 2011
6	Anti-ulcer	Seeds	Alok et al., 2012
			Okunrobo et al., 2012
7	Hepatoprotective	Seed oil and fruit	Madhavi et al., 2012
8	Anti-hyperlipidemic	Sentinel	Sevcan et al., 2011
9	Antidiabetic	Fruits	Aruna et al., 2012
10	Compositional studies	Seeds	Francis et al., 2013
11	Anti-prostatic hyperplasia	Seeds	Godwin et al., 2008
12	Analgesic	Seeds	Gill et al., 2010
13	Anti-secretory	Fruits	Aruna et al., 2012
14	Histology on kidney	Fruits	Adesanya et al., 2011

medicine for the treatment of several infectious diseases such as sore eye, scabies, itches, ulcer, pain, constipation, malaria and others (Rahman et al., 2008). The fruit is diuretic and effective in treatment of fever, dropsy and renal stones (Grieve and Leyel, 1984; Chiej, 1984). Others include; antihypertensive effect (USA, 2012), alcoholic poisoning and diabetes (Duke and Ayensu, 1998). The root is purgative and at higher dose is said to be emetic. The seed is demulcent, diuretic, pectoral and tonic (Duke and Ayensu, 1998). It is also used in the treatment of urinary tract infections (Grieve and Leyel, 1984), bed wetting (Moerman, 1998) and has a good vermifuge and hypotensive effect. The leaves are a good antimalarial, analgesic, anti-inflammatory, mosquitocidal, and antimicrobial agents and can be used in the treatment of gonorrhea (Personal Communication). A number of bioactivity studies have been reported on the seeds, seed oil, fruits, stem and leaves of *C*. *lanatus* (Table 1). Thus, the aim of the study is to evaluate the antimicrobial activities of the crude methanol extract and fractions (hexane, chloroform, ethylacetate and n-butanol) of leaves of *C. lanatus* against some selected bacteria and fungi.

MATERIALS AND METHODS

Collection and identification of plant material

The plant material was collected from Illela Local Government Area, Sokoto State, Nigeria in December 2014. It was authenticated by Namadi Sanusi at the Herbarium Unit, Department of Biological Sciences, Ahmadu Bello University, Zaria by comparing with existing specimen with voucher No. (1266). The leaves were shade dried, pulverized, labeled and stored at room temperature prior to extraction.

Preparation of the extract

The powdered leaves (154 g) were extracted with methanol using maceration method for 3 days. The extract was evaporated *in-vacuo* using rotary evaporator at 40°C to yield a gummy greenish residue (25.6 g) subsequently referred to as the crude methanol extract (ME). Twenty gram (20 g) of ME was suspended in distilled water, filtered and fractioned successively into hexane (3.14 g), chloroform (0.22 g), ethylacetate (2.43 g) and n-butanol (4.40 g) fractions coded as HF, CF, EF and BF respectively. The fractions were concentrated and kept in a refrigerator prior to use.

Microbial species

Ten (10) microbial species were obtained from the Department of Medical Microbiology, Ahmadu Bello University Teaching Hospital, Zaria, Nigeria. The microbes include *Streptococcus pyogenes*, *Streptococcus faecalis*, *Bacillus cereus*, *Corynebacterium*

ulcerans, Escherichia coli, Klebsiella pneumoniae, Pseudomonas flourescens, Salmonella typhi, Candida albicans and *Candida krusei*. All bacterial cultures were checked for purity and maintained in a blood agar slant while fungi were maintained on a slant of Sabouraud Dextrose Agar (SDA).

Antimicrobial evaluation

Susceptibility test

An antimicrobial activity of HF, CF, EF, BF and ME was determined via susceptibility test using agar diffusion method. 10 mg/ml (stock concentration) was prepared by dissolving 0.1 g of the fractions each in 10 ml dimethyl sulfoxide (DMSO). Mueller Hinton agar and the growth medium, was prepared according to manufacturer's instructions and sterilized for 15 min at 121°C. Nutrient broth and sabouraud dextrose broth were used for antibacterial and antifungal evaluations respectively. The test organisms were inoculated and incubated for 24 h for bacteria and 48 h for fungi. The solidified sterile medium contained in petri dishes were seeded with 0.1ml standard inocula of the test microbes at 45°C. Wells were bored into the solidified inoculated nutrients agar plates using cock borer of 6 mm diameter. The wells were filled with 0.1 ml DMSO solution of the extract. Ciprofloxacin (5 µg/ml) and Fluconazole (5 µg/ml) discs were also placed on the agar plates and served as the standard drugs for bacteria and fungi, respectively. 1 h was allowed for the extract and the standard compounds to diffuse into the agar after which the plates were incubated overnight at 37°C and 25°C for bacteria and fungi, respectively. At the end of incubation period, diameter of inhibition zone was measured using transparent ruler and recorded. The zones of inhibition of microbial growth were tested in duplicates and the mean of the results was recorded in millimeters (mm).

Minimum inhibitory concentration (MIC)

The MIC of the extract was carried out using Broth dilution method (Volekobia et al., 2001; Abdullahi et al., 2015). Mueller Hinton broth was prepared of which 10 ml was dispensed into test tubes, sterilized at 121°C for 15 min. and allowed to cool; MC-Farlands standard turbidity scale number 0.5 was prepared. Dilution of the organism suspension was done continuously using sterile normal saline until the turbidity marched that of Mc-Farland's scale by visual comparison. At that point, the concentration of the test microbe was about 1.5×10^8 cfu/ml. Two-fold serial dilution of the extract in the sterile broth was made to obtain the concentrations of 5, 2.5, 1.25 and 0.625 mg/ml. 0.1ml of the standard inoculum of the test microbe was then inoculated into the different concentrations of the extract in the broth. The tubes were incubated at 37°C for 24 h and 25°C for 48 h for bacteria and fungi respectively after which the plates were observed for turbidity (growth). The MIC was defined as the lowest concentration of the extract inhibiting the visible growth of each micro-organism.

Minimum bactericidal/fungicidal concentration (MBC/MFC)

The MBC/MFC was carried out to determine whether there is complete death of test microbes or just growth inhibition. Mueller Hinton agar was prepared, sterilized, poured into sterile petri dishes and allowed to cool and solidify. The contents of the MIC in the serial dilution were then sub-cultured on to the prepared media. Incubation was made at 37°C for 24 h and the plates were observed for colony growth. The plates with the least concentration and no colony growth were taken as MBC/MFC (Abdullahi et al., 2015).

RESULTS

The results of susceptibility test, zone of inhibition, MIC, MBC/MBF of the fractions (ME, EF, BF, CF and HF) of *C. lanatus* are presented in Tables 2-5 respectively;

The crude extract and fractions of *C. lanatus* exhibited varying degree of antimicrobial effect against all the tested microbes except *S. pyogenes*, *C. ulcerans*, *K. pneumoniae* and *S. typhi*. The standard antibiotic agent, Ciprofloxacin showed no activity against *P. flourescens*, *C. albicans* and *C. krusei*, while fluconazole, a standard antifungal drug was active against *C. albicans* and *C. krusei* only.

The results showed inhibition ranges of 16 - 18, 26-29, 20-23, 22-25 and 20-22 mm for ME, EF, BF, CF and HF respectively against *S. faecalis*, *B. Cereus*, *E. Coli*, *P. flourescens*, *C. albicans* and *C. krusei.* EF was the most active with highest sensitivity against *B. ceureus* and *P. flourescens* (29 mm). The least sensitive organism was *S. faecalis* (16 mm) to ME. The MIC of ME was 5mg/ml, 1.25 - 2.5 mg/ml for EF and 2.5 mg/ml for BF, CF and HF. Highest activity was recorded at 1.25mg/ml in EF against *S. faecalis*, *B. cereus*, *P. flourescens* and *C. albicans.* With MIC of 5 mg/ml, ME had the least activity against all the test organisms. The MBC/MFC were recorded as 10mg/ml for ME and HF, ranges of 2.5 - 5mg/ml (EF) and 5 - 10mg/ml for BF and CF. This revealed that the ethyl acetate fraction has the highest bactericidal/fungicidal activity at 2.5 mg/ml against *S. faecalis*, *B. Cereus*, *P. flourescens* and *C. albicans.* ME and HF were least active against all the test microbes.

Discussion

It is widely accepted that the pharmacological effect of

Table 2. Zone of Inhibition of the extract/fractions against the test organisms (mm).

Test Organism	ME	EF	BF	CF	HF	Ciprofloxacin	Fluconazole
Streptococcus pyogenes	0	0	0	0	0	35	0
Streptococcus faecalis	16	27	21	24	20	37	0
Bacillus cereus	18	29	21	25	21	39	0
Corynebacterium ulcerans	0	0	0	0	0	30	0
Escherichia coli	17	26	20	23	21	37	0
Klebsiella pneumonia	0	0	0	0	0	32	0
Pseudomonas flourescens	18	29	23	24	22	0	0
Salmonella typhi	0	0	0	0	0	42	0
Candida albicans	16	28	21	23	20	0	35
Candida krusei	18	26	20	22	20	0	32

Standard Drugs concentration: Ciprofloxacin 5µg/ml, Fluconazole 5µg/ml.

Table 4. Minimum Inhibitory concentration of the fractions against the test organisms.

Extract	ME					EF					BF					CF					HF				
Concentration/Test Organism	**10mg/ml**	**5mg/ml**	**2.5mg/ml**	**1.25mg/ml**	**0.625mg/ml**	**10mg/ml**	**5mg/ml**	**2.5mg/ml**	**1.25mg/ml**	**0.625mg/ml**	**10mg/ml**	**5mg/ml**	**2.5mg/ml**	**1.25mg/ml**	**0.625mg/ml**	**10mg/ml**	**5mg/ml**	**2.5mg/ml**	**1.25mg/ml**	**0.625mg/ml**	**10mg/ml**	**5mg/ml**	**2.5mg/ml**	**1.25mg/ml**	**0.625mg/ml**
S. pyogenes																									
S. faecalis	-	A	B	C	D	-	-	-	A	B	-	-	A	B	C	-	-	A	B	C	-	-	A	B	C
B. cereus	-	A	B	C	B	-	-	-	A	B	-	-	A	B	C	-	-	A	B	C	-	-	A	B	C
C. ulcerans																									
E. coli	-	A	B	C	D	-	-	A	B	C	-	-	A	B	C	-	-	A	B	C	-	-	A	B	C
K. pneumonia																									
P. flourescens	-	A	B	C	D	-	-	-	A	B	-	-	A	B	C	-	-	A	B	C	-	-	A	B	C
S. typhi																									
C. albicans	-	A	B	C	D	-	-	-	A	B	-	-	A	B	C	-	-	A	B	C	-	-	A	B	C
C. krusei	-	A	B	C	D	-	-	A	B	C	-	-	A	B	C	-	-	A	B	C	-	-	A	B	C

Key -= no turbidity (no growth), A=MIC, B= Turbid (light growth), C= moderate turbidity, D= High turbidity.

Table 5. Minimum bactericidal/fungicidal concentration of the fractions against the test microbes.

Extract	ME					EF					BF					CF					HF				
Concentration/Test Organism	**10mg/ml**	**5mg/ml**	**2.5mg/ml**	**1.25mg/ml**	**0.625mg/ml**	**10mg/ml**	**5mg/ml**	**2.5mg/ml**	**1.25mg/ml**	**0.625mg/ml**	**10mg/ml**	**5mg/ml**	**2.5mg/ml**	**1.25mg/ml**	**0.625mg/ml**	**10mg/ml**	**5mg/ml**	**2.5mg/ml**	**1.25mg/ml**	**0.625mg/ml**	**10mg/ml**	**5mg/ml**	**2.5mg/ml**	**1.25mg/ml**	**0.625mg/ml**
S. pyogenes																									
S. faecalis	A	B	C	D	D	-	-	A	B	C	A	B	C	D	D	-	A	B	C	D	A	B	C	D	D
B. cereus	A	B	C	D	D	-	-	A	B	C	A	B	C	D	D	-	A	B	C	D	A	B	C	D	D
C. ulcerans																									
E. coli	A	B	C	D	D	-	A	B	C	D	A	B	C	D	D	-	A	B	C	D	A	B	C	D	D
K. pneumonia																									
P. flourescens	A	B	C	D	D	-	-	A	B	C	-	A	B	C	D	-	A	B	C	D	A	B	C	D	D
S. typhi																									
C. albicans	A	B	C	D	D	-	-	A	B	C	A	B	C	D	D	-	A	B	C	D	A	B	C	D	D
C. krusei	A	B	C	D	D	-	A	B	C	D	A	B	C	D	D	A	B	C	D	D	A	B	C	D	D

Key -= no colony growth, A=MBC/MFC, B= Scanty colonies growth, C= Moderate colonies growth, D= High colonies growth.

plants lie in the bioactive phyto-principles they contain (Volekobia et al., 2001). Bacteria and fungi such as *C. krusei*, *E. coli* and *B. cereus* are causative agents of several diseases and infections such as diarrhea, dysentery e.t.c (Cowan, 1999; WGOGG, 2012). MIC less than 100 µg/ml indicated that the extract/fractions have very strong resistance to these infective microbes (FP, 2014). The extract and fractions had broad spectrum antimicrobial activity due to their ability to inhibit growth of both gram negative and gram positive bacteria. It is noteworthy that *S. pyogenes, C. ulcerans, K. pneumoniae* and *S. typhi* were resistant to all the fractions at the tested concentrations. The extracts showed varying levels of activity probably due to different composition of phyto-constituents present in the plant (Tang et al., 2003). Antimicrobial potency of the extracts and fractions were in the order; EF> CF>BF>HF>ME. The highest activity was exhibited by the ethyl acetate fraction thereby suggesting that the components with antimicrobial activity in are the non-polar and/or moderately polar components. The phytochemical screening of the extract/fractions revealed the presence of saponins, alkaloids, flavonoids, phenols, steroids and triterpenes which varies in other fractions (Alebiosu and Yusuf, 20015). However, there has been established relationship between antimicrobial activity and these plants based constituents. Antimicrobial activity of saponins (Soetan et al., 2006), alkaloids (Daminito et al., 2005), flavonoids (Abdullahi et al., 2015), steroids and terpenes (Silvia et al., 2003) have been reported. Further work aimed at the isolation and elucidation of the bioactive principles is being pursued.

Conclusion

The result of this research suggests that the leaves of *C. lanatus* have a remarkable antimicrobial activity which validates the ethno-medicinal claim of the use of the leaves in management of microbial infections.

Conflict of interest statement

We declare that we have no conflict of interest.

REFERENCES

Yusuf AJ, Abdullahi MI, Haruna AK, Musa AM, Abdullahi MS, Ibrahim ZYY, Halilu ME and Odiba OJ (2015). Phytochemical and antimicrobial evaluations of the methanol stem bark extract of N*eocarya macropylla*. J. of Chem. and Pharm. Res., 7(1):477-481.

Gislene GF, Locatelli NJ, Paulo CF Giuliana LS (2000). Antibacterial activity of plant extracts and phytochemicals on antibiotic resistant bacteria. Braz. J. Microbiol., 31: 247-256.

Robinson RW, Decker-Walters RDS(1997). Cucurbits. NewYork Cab International, (Crop Production Science in, Horticulture nE.6), p.226.

Rahman AHMM, Anisuzzaman M, Ferdous A, Rafiul Islam AKM, Naderuzzama ATM (2008). Study of Nutritive Value and Medicinal Uses of Cultivated Cucurbits, J. of Appl. Sci. Res., 4(5): 555-558.

Grieve M, Leyel CF (1984). A modern herbal: Penguin Harmondsworth, 1984.

Chiej R (1984). Encyclopaedia of medicinal plants. MacDonald, 1984; ISBN 0-356-10541-5.

USA (2012). Watermelon extract supplementation reduces an [*Am J Hypertens*. 2012] - PubMed -NCBI". Ncbi.nlm.nih.gov; 2012. Retrieved 2012-08-13.

Duke JA, Ayensu ES (1985). Medicinal plants of China: Reference Publications, 1985.

Moerman, DE. Native American ethnobotany: Timber Pr, 1998.

Swapnil S, Sarvesh P, Jaya D, Amita T (2011). First report on laxative activity of *Citrullus lanatus*. Pharmacologyonline, 2: 790-797.

Loiy EAH, Hasnah MS, Sakina M, Ahemd Y, Waleed S, Koko S, and Siddig IA (2011). In vitro Antimicrobial activities of chloroformic, hexane and ethanolic extracts of *Citrullus lanatus* var. citroides (Wild melon). J. of Med. Plants Res., 5(8): 1338-1344.

Gill NS, Bansal RK, Garg M, Sood S, Muthuraman A, Bali M (2010). Evaluation of antioxidant, anti-inflammatory and analgesic potential of *Citrullus lanatus* seed extract in rodent model. The Internet J. of Nutr. and Wellness, 9(2): 12

Madhavi P, Maruthi R, Kamala V, Habibur RM and Chinna E (2012). Evaluation of Anti-Inflammatory Activity of *Citrullus lanatus* Seed Oil by In-vivo and In-vitro Models. Int. Res. J. Pharm. App Sci., 2012; 2(4): 104-10.

Naresh SG (2011). Evaluation of Antioxidant activity of Citrullus lanatus Seed extract in Rats. Latin American Journal of Pharmacy (formely Acta Farmaceutica Bonaerense), Lat. Am. J. Pharm., 30(3):429-34.

Alok BRK, Vivek D, and Niyaz A (2012). Evaluation of anti-ulcer activity of *Citrallus lanatus* seed extract in Wistar rats. J. of Pharm. and Pharm. Sci., 4S (5): 16

Okunrobo OL, Uwaya OJ, Imafidon EK, Osarumwense OP, Omorodion EJ (2012). Quantitative determination, Metal analysis and Antiulcer evaluation of Methanol seeds extract of *Citrullus lanatus* Thunb (Cucurbitaceae) in Rats. Asian Pacific J. of Tropical Dis., 2012; S1:261-S1265.

Madhavi P, Kamala V, Habibur R. Hepatoprotective Activity of *Citrullus Lanatus* Seed Oil on CCl4, Induced Liver Damage in Rats. Scholars Academic J. of Pharm. (SAJP), 1(1): 30-33.

Sevcan A, Göksel K, Murat K, Aydın K, Parvez IH (2011). Protective effect of Diyarbakır watermelon juice on carbon tetrachloride-induced toxicity in rats. Food and Chemical Toxicol., 49: 2433–2438.

Aruna P, Debra LR, Shubin KS, Sibu S, Alan D (2012). *Citrullus lanatus* 'sentinel' (watermelon) extract reduces atherosclerosis in LDL receptor-deficient mice. J. of Nutr. Biochem., 2012; (Article in Press).

Francis SO, Morufu EB and Adedeji GT (2013). Antisecretory Effects of Watermelon (*Citrullus lanatus*) Juice in Male Albino Rats. Annual Rev. & Res. in Biology, 2013; 3(4): 358-366.

Godwin CO, Olarewaju MO, Yetunde RO, Kayode EA, George OE, Reginah TO (2008). Compositional Studies of *Citrullus lanatus* (Egusi melon) Seed. The Internet J. of Nutr. and Wellness, 2008; ISSN: 1937-8297.

Adesanya AO, Olaseinde OO, Oguntayo OD, Otulana JO, Adefule AK (2011). Effects of Methanolic Extract of *Citrullus lanatus* Seed on Experimentally Induced Prostatic Hyperplasia. Euro. J. of Medicinal Plants. 1(4):171-179.

Volekobia A, Kostalova D, Sochorova R (2001). Isoquinoline alkaloid from *Mahonia aquifolium* stem bark is active against *Malsseizia* species. Folia Microbiol., 46, 107-111.

Cowan MM (1999). Plant Products as Antimicrobial Agents. Clin. Microbiol. Rev., pp. 564-582.

World Gastoroenterology Organization Global Guidelines (WGOGG) (2012). Acute diarrhea in adults and children: a global perspective. 2012, 1-24.

Food Poisoning (FP). www.foodsafety.gov/foodpoisoing. Retrieved 28 December 2014.

Tang S, Bremner P, Kortenkamp A, Schlage C, Gray AI, Gibbons S, Heinrich M (2003). Planta Med. 2003; 69: 247.

Alebiosu CO, Yusuf AJ (2015). Phytochemical Screening, Thin-layer

Chromatographic Studies and UV Analysis of Extracts of *Citrullus lanatus*. J of Pharm, Chemical and Biol., 3(2): 214-220.
Soetan K, Oyekunle MA, Aiyelaagbe OO, Fafunso MA (2006). Evaluation of the antimicrobial activity of saponins extract of *Sorghum Bicolor* L. Moench. Afr. J. Biotechnol., 5(23): 2405-2407.
Daminito K, Aly S, Antonella C, Saydou Y, Carla M, Jacques S, Vittoro C, Alfred ST (2005). Antibacterial activity of alkaloids from *Sida cuta*. Afr. J. Biotechnol., 4(12): 1452-1457.
Abdullahi MI, Musa AM, Haruna AK, Sule MI, Abdullahi MS, Akinwade Y, Abimiku AG, Iliya I (2011). Antimicrobial flavonoid diglycoside from the leaves extract of *Ochna schweinfurthiana* (*Ochnaceae*). Nig. J. of Pharm. Sci., 10(2): 1-7.
Silvia HT, Marcos JS, Evandio W, Izabel YI, Dioneia CR (2003). Antimicrobial activity of flavonoids and steroids isolated from two *Chromolaena* species. Braz. J. of Pharma. Sci., 39(4): 403-408.