PharmacologyOnLine August 30, 2012
Archives • 2012 • vol.2 • 98 - 103

ANTIMICROBIAL EFFECTS OF EXTRACTS AND DECUSSATIN FROM *Anthocleista vogelii* (PLANCH)

Alaribe CS [1*], Coker HAB [1], Shode F [2], Ayoola G[1], Adesegun S[3], Singh N[4], Anyim E[5] and Iwuanyanwu S[6]

[1] Department of Pharmaceutical Chemistry Faculty of Pharmacy, University of Lagos, Nigeria.
[2] School of Chemistry, University of Kwazulu Natal, Westvile, Durban, South Africa.
[3] Department of Pharmacognosy, Faculty of Pharmacy, University of Lagos, Nigeria.
[4] School of Biological Studies, University of Kwazulu Natal, Westvile, Durban,South Africa.
[5] Michael Okpara University of of Agriculture Umudike, Abia State.
[6] Dept of Mycology, College of Medicine, Lagos University Teaching Hospital, LUTH, Lagos.

*amachichi1@yahoo.com , *P.O.Box; 11661, Ikeja, Lagos. Tel: +234 - 8037263962, +234-8028968011*

Abstract

Anthocleista vogelii (Gentianaceae) has been claimed to be used traditionally in the treatment and management of fever and infectious related diseases. Investigation of pet.ether leaves (PEL) and stembark (PES) extracts of *A.vogelii* using chromatographic techniques led to isolation of xanthones, fatty acid and steroid; i) 1-hydroxy-3,7,8 trimethoxyxanthone (Decussatin); ii) 1,8-dihydroxy-3,7-dimethoxyxanthone, (Swertiaperennin); iii) hexadecanoic acid from the PEL extracts and iv) Stigmasta-5, 22-dien-3beta-ol and Decussatin from the PES extracts respectively. Spectral analysis were carried out with the aid of IR & UV (Perkin Elmer), GC – MS (Agilent 5973 Network Plus), 1D & 2D NMR(400MHz, 600MHz,TopsinBruker).

PEL, PES and decussatin, the main isolate from both extracts ,at various concentrations were screened *in vitro* for antibacterial and antifungal activities using Disk agar diffusion (DAD) and Brothmicrodilution (MHB) techniques against *Escherichia coli, Bacilus substilis*, *Klebsiela pneumonia*, *Staphylococcus aureus, Aspergillus fumigatus*, *Trichophyton mentagrophytes, Trichophyton rubrum* and *Candida albicans*. PES extracts showed potent antibacterial activities against *E. coli*, *B. subtilis* and *S. aureus* with MIC of 1 $mgml^{-1}$, 6 $mgml^{-1}$ and 7 $mgml^{-1}$ respectively while PEL extracts showed notable antibacterial activities against the four bacteria *E. Coli*, *K. Pneumonia* and *B. substilis*, at MIC of 1 $mgml^{-1}$ and 2 $mgml^{-1}$ resp. with the exception of *S. aureus.* PEL extracts was significantly active against *C. albicans* with MIC of 2 $mgml^{-1}$ and zone of inhibition 22.00 mm. However, Decussatin showed no appreciable activity at the maximum concentration (8 and 10 $mgml^{-1}$) used for antibacterial and antifungal activities respectively.

Decoctions prepared from the stembark and leaves of *A.vogelii* are used by the Southwestern Egun people of Nigeria in the management of "fevers" (translated in local language as *iba*) arising from infections. This has served as the driving impetus for the present investigational work on this plant. Ferrous Chelating activities (*FCA*) of were determined which is the lowest concentration (1$mgml^{-1}$) of PEL and decussatin at which maximum chelating activities [Fe^{2+} chelating ability (%)] comparative with EDTA, reference standard were observed. PEL extract demonstrated good ferrous chelating ability at 1$mgml^{-1}$ (89.48± 1.00). Decussatin demonstrated moderate ferrous chelating activities (77.80 ± 0.10). The leaves extract (PEL) is safe up till 2000 $mgkg^{-1}$.

Keywords: Gentinaceae, *Anthoclesita vogelii* , Decussatin, Antimicrobial, DAD, MHB

Introduction

Plants of West-Africa region of Africa have been biologically exploited especially those that are medicinally potent. Folks have over the years depended greatly on one part of plants for nutrition, healing and management of ailments, yet modest information on the single component bioactive constituents responsible for the biological action is sparingly known.

Quite a number of anti-microbial agents have originated from plants and their secondary metabolites. The fact that plants contain metabolites with inbuilt safety limit and a wide variety of different active principles is advantageous for drug discovery [1]. Despite the progress made in recent centuries in the fight against harmful microorganisms, incidents of epidemics due to drug resistant microorganisms and the emergence of hitherto unknown disease-causing microbes, pose enormous public health concerns [2]. Medicinal chemists are still actively seeking new and improved anti-microbial agents to combat this worrisome menace. The quest to join in the search for new anti-microbial agents has instigated this research study.

Some plants and herbal remedies are known to be used in the treatment or fight against microbial infections. Such plants include extracts and decoctions of *Hydrastis Canadensis* (goldenseal), *Garcinia kola* (bitter kola), *Aframomum melegueta* (grains of paradise), *Xylopia aethiopia*, to mention but a few.

Anthocleista vogelii (Planch), a local plant was selected following an ethno-pharmacological investigation and indigenous claim that the plant is used to treat fever, malaria and infectious related diseases. All the parts are noted to be used therapeutically [3].

Our research study is focused on the identification, isolation, elucidation and anti-microbial evaluation of the crude extract and isolate(s) from the leaves (PEL) and stembark (PES) of *A.vogelii*.

Materials And Methods

Plant material: Leaves and stembark of *A. vogelii* were collected in August, 2007 from uncultivated land in University of Lagos, Akoka, identified and authenticated by Mr. M. O. Onadeji, Herbarium Officer at the Forest Research Institute of Nigeria (FRIN), Ibadan and voucher specimen (FHI NO 107844) was deposited at same institute. The leaves and stembark were air dried at room temperature, pulverised using electric grinder and kept at room temperature prior to use.

***Extraction, Isolation and Structure Elucidation*:** The dried pulverized leaves and stembark of *A. vogelii* (350g each) were extracted with petroleum ether [PE] for 72h at 40°C using soxhlet extractor, evaporated in vacuo to yield semi solid brown and gummy extract; PEL (50 g) and PES (35 g) respectively. The pet ether leaves [PEL] fraction (20g) was chromatographed on a column of silica gel (200 g) and eluted with hexane-EtOAC (10:1; 1–10, step gradient) to give fourteen fractions (A – N) of 20 mL each. The fractions thus obtained were compared using TLC fluorescence silica gel plates with hex-EtOAc {8:2} as solvent. Those giving similar spots were combined. Fractions G, left in the vial overnight formed solid deposit which was decanted and the residues recrystallised using MeOH. The recrystallized product afforded Compound **1**. Fraction H subjected to LC-MS and GC-MS afforded compounds **2** and **3**.

PES fraction was concentrated in vacuo, subjected to vaccum column chromatography (vcc) and further eluted with hex—EtOAc {7: 3}. Like fractions (1- 6) were collected in vials, fraction 5 was found with deposits of impure compound. The deposits was decanted from supernants and recrystallised in MeOH to yield compound **4**. The supernant was further subjected to vcc to yield fraction 1- 19. Fraction 2 gave compound **5**.

The isolated compounds were successively subjected to ^{1}H NMR, infra red (IR), UV, MS and ^{13}C NMR analysis for structural elucidation. The ^{1}H-NMR and ^{13}C-NMR were recorded in $CDCl_3$ with TMS as internal standard on Bruker AMX-400 Ultrashield

NMR spectrometer operating at 400 and 600 MHz. The infrared (IR) was recorded on the Perkin Elmer-Universal ATR sampling Accessory, FT-IR spectrometer. LC-MS of 150mm x 3.0mm i.d.; 3.5 µm C18 XTerra column (phenomenex) was used and the eluent consisted of acidified methanol and water. The column temperature was 30^{O}C. The eluent flow rate was 1.0 ml /min and the injection volume used was 20 µL. The LC /MS data was acquired using a LC - Agilent MS-Ion Trap Quadrupole analyzer. The ultraviolet (UV) spectra were taken in DCM and recorded on Lambda 18 UV/VIS Spectrometer. Si gel refers to Merck Kieselgel 60 (70–230 and 230–400 mesh ASTM). All other reagents and chemicals used were of analytical grade.

Microdilution Bioassay: Microdilution bioassay method was carried using the microplate method of Eloff [4] with modifications. PEL, PES extracts and decussatin were screened against ATCC culture strains of *Escherichia coli* (ATCC 11775), *Bacillus subtilis* (ATCC 6051), *Klebsiella pneumoniae* (ATCC 13883) and *Staphylococcus aureus* (ATCC 12600) obtained from The School of Biological and Conservation Sciences, University of Natal, Pietermaritzburg Campus, South Africa. PES, PEL extracts (1–8 mgmL^{-1}, 100 µL) and decussatin (8 mgmL^{-1},100µL) were introduced separately into 96 well microplates and 100 µL bacterial cultures were added to each well. Neomycin (100 ìgmL^{-1}) was used as positive control, 10% tween 80, a suspending medium for extracts was used as negative control and bacterial free wells used as blank controls. The microplates were covered with parafilm and incubated for 24 h at 37°C Forty (40) µL of 0.2 mgmL^{-1} of p-iodonitrotetrazolium violet (INT) were added to the wells to serve as an indicator of bacterial growth and incubated at 37 °C for 30 min. The minimum inhibitory concentration (MIC) was taken as the lowest concentration of the extract or compound that completely inhibited bacterial growth. All determinations were in triplicates.

Antifungal Assay: Disk Agar Diffusion (DAD) method [5] was used in this study. The plates were prepared by pouring 15 mL of molten Sabouraud dextrose agar into sterile petri dishes. The plates were allowed to solidify and 0.1% inoculums suspension of four fungi clinically isolated from the dept of Mycology, College of Medicine, University of Lagos. The fungi; *A.fumigatus*, *T.rubrum, T.mentagrophytes* and *C. albicans* were separately swabbed uniformly and incubated at 25–30 °C. The cultures were examined every 2–3 days under the microscope for evidence of growth for 6 weeks. Various concentrations of PEL, PES extracts (1–5 mg/mL) and decussatin (10 mg/mL) were loaded separately on individual discs. The discs were placed on the surface of the medium and the plates were incubated at 30 °C for 24 h. Nystatin (100 ìg/mL) and canesten (100 ìg/mL) were used as positive controls. Negative controls were prepared using 10% tween 80 and distilled water. The zones of inhibition around the disc were measured in millimeter. All determinations were in triplicates.

Acute Toxicity Studies: The medial lethal dose of the PEL that would kill 50 % of the animals in a population (LD_{50}) was determined intraperitoneally. Albino mice were divided into five groups of six (6) animals each weighing between (18 - 20g). The mice were subjected to 24 hrs fasting (with only water) before administering PEL. PEL was suspended in medium (Tween 80 in distilled water, 3% v/v) and administered in dose of 1,000, 2000, 4,000, 6,000 and 8,000 mg/kg i.p. The sixth group served as control received only 3% Tween 80. All animals were kept at room temperature in cross ventilated room without illumination at night. The mice were then observed for toxicity and fatalities over 48hrs.

Ferrous chelating ability (FCA) of Extract: A slightly modified method developed by Dinis [6] was adopted. PEL and Decussatin (5.0 ml each; 1 mg/ml) including EDTA Solution (reference standard) were spiked with 0.1 ml of 2 mM $FeCl_2$ and 0.2 ml of 5 mM ferrozine solution. After reaction for 10 mins, the absorbance (at 562 nm) of the resulting solutions was recorded. The higher the ferrous chelating ability (%) of the test sample, the lower was the resulting absorbance.

The percentage of ferrous ion chelating ability was expressed by:

$\% = [(A_o - A_S) / A_S] \times 100$

A_o = the absorbance of the control; A_s = the absorbance of the extract

Statistical analysis: The statistical analysis of all the analyses was carried out using GraphPad Prism 5 Demo. The values are represented as mean ± SD. One way Anova was used for comparison between groups. *P* values less than 0.05 and in some cases less than 0.01 were judged significant.

Results And Discussion

The present study was aimed to characterize the phytochemicals isolated from the pet-ether leaves(PEL) and stembark (PES) extracts of *A.vogelii*, to evaluate the in-vitro anti-microbial activities of the extracts and isolates upon a representative wide range of pathogenic micro-organisms and to determine the safety of the plant extracts. The use of plants, herbs and all forms of natural product in the treatment of ailments cannot be over used, particularly in local health care systems and developing countries where cultural and economic reasons play vital roles.

Phytochemical results revealed four compounds from PEL and PES extract of *A.vogelii*.

The UV, IR, 1D and 2D NMR spectral data of compound **1** and **4** from PEL and PES were consistent with the presence of a xanthone nucleus. Comparison of their spectroscopic and physical data with those published allowed us to establish the structure as 1- hydroxyl -3, 7, 8 –trimethoxyxanthones – Decussatin. Compound **2** and **3**; 1, 8 - dihydroxy - 3, 7- dimethoxyxanthone (swertiaperennin) and hexadecanoic acid respectively were isolated from PEL while Compound **5**; Stigmasta-5, 22- dien-3-ol (stigmasterol) was isolated from PES. Figure 1. Two of the compounds; decussatin and swertiaperenin have been previously isolated from the leaves of *A.vogelii* [7].

The result of the MIC determination of PEL, PES, decussatin and positive controls against the strains of microorganisms tested is shown in Table 1. PES showed potent antibacterial activity against three out of the four tested bacterial; *E. coli*, *B. subtilis* and *S.aureus* with MIC of 1 $mgmL^{-1}$,6 $mgmL^{-1}$ and 7 $mgmL^{-1}$ respectively. PEL showed more potent antibacterial activity against the tested bacteria; *E. coli*, *B. subtilis* and *K.pneumoniae* with MIC 1 $mgmL^{-1}$, 1 $mgmL^{-1}$ and 2 $mgmL^{-1}$ respectively. The antifungi studies on *A.vogelii* showed that both PEL and PES were strongly active against *C. albicans* with MIC of 1 $mgmL^{-1}$. Decussatin showed no significant antibacterial activity against any of the tested strains of microorganism at the highest concentration; 8 $mgmL^{-1}$ and 10 $mgmL^{-1}$ for antibacterial and antifungi respectively. Decussatin and Swertiaperennin belong to a class of secondary metabolites known as Xanthones commonly occurring in a few higher plant families, in fungi and lichens [8]. Recently, various bioactivities of xanthones including cytotoxic, antitumor, anti-inflammatory, antifungal, enhancement of choline acetyltransferase activities and inhibition of lipid peroxidase have been described [9]. The anti-infective activities of some exudates of *A.vogelii* may well be attributed to these existing isolated compounds.

The ferrous chelating ability (*FCA*) for PEL, PES and decussatin indicated by the lowest concentration IC ($1mgml^{-1}$) as shown in table 2. The Ferrous chelating ability (%) values revealed that PEL and PES showed appreciable and promising ferrous chelating activities (89.48 ± 1.00) and (88.32 ±0.52) comparable to EDTA (118.5 ± 0.50). decussatin showed moderate *FCA* activity. (77.80 ± 0.10).

The emergence of drug resistant micro-organism has intensified the search for new therapeutic approaches such as drug combinations. One of such new approach under investigation is the administration of ferrous chelating agents [10, 11]. It is possible that ferrous chelators can chelate a metal ion (e.g iron) needed for normal functioning of an enzyme system in targeted micro-organism (fungi, bacteria,

virus) and consequently destroying the microorganism. This is one of the postulated initiatives of possible mechanism of action of the plant extracts.

The acute toxicity test indicated non-toxicity of PEL up till 2000mgkg^{-1}.

The present study revealed the presence of xanthones, steroids and palmtic acid which synergistically revealed the anti-microbial activities of *A.vogelii* as showed by PEL and PES. The single component – decussatin subjected though concentration controlled was insignifically effective in the anti-microbial assay. The plant, *A.vogelii* can be seen as a potential source of drug. This result supports to an extent the ethno medicinal uses of *A.vogelii* in the treatment of infections and related diseases.

Structural and spectral information

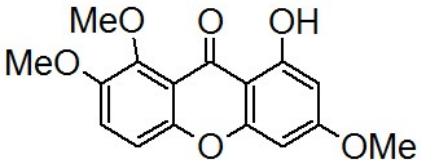
*Compound **1 & 4**:* 1 hydroxy - 3, 7, 8 trimethoxyxanthone (Decussatin); Yellow - whitish crystals, $C_{16}H_{14}O_6$; M.pt; 150°C–154°C . M^+ : m/z 302, runtime: 11.6mins; 287, 259, 227, 201, 171, 143, 122, 100, 79, 51. IR ν_{max} cm^{-1} : 3100, 2919, 2849, 1743, 1658, 1596, 1571, 1480, 946 for major peaks. ^{1}H NMR (600 MHz, $CDCl_3$): 13.24 (1H,s ArOH), 7.31 (1H,d, J 9.18Hz), 7.15 (1H,d, J 9.18Hz), 6.31 (1H, d, J 2.20Hz), 6.20 (1H,d, J 2.20Hz), 3.98 (3H, s, OMe), 3.91 (3H,s,OMe), 3.86 (3H,s,OMe); ^{13}C NMR (600 MHz, $CDCl_3$): δ; 181.2(C-9), 166.4(C-3), 163.9(C- 1), 157.1(C-4a), 151.0 (C-5a), 149.3(C- 7), 148.9 (C-8), 120.5(C-6), 115.8 (C-8a), 112.8 (C-5), 104.1 (C-9a), 96.9 (C-2), 92.1 (C-4), 61.8 (OCH_3 – 8), 57.2 (OCH_3 – 7, 55.8 (OCH_3 – 3) . ^{13}C and ^{1}H- NMR are in agreement with literature [12].

*Compound **2**:* 1, 8 - dihydroxy - 3, 7- dimethoxyxanthone (Swertiaperennin); Pale yellow crystals; $C_{15}H_{12}O_6$; M.pt; 150 ° -153 °C . M^+ : m/z 288.; IR ν_{max} cm^{-1}: 2900.00, 2849.34, 1704.62, 1667.67, 1641.30, 1603.59, 1575.86, 1505.88, 1464.27, 1436.83, 1378.45 for major peaks. ^{1}H NMR: (600 MHz, $CDCl_3$); 12.00 (1H,s, ArOH), 11.40 (1H,s), 7.20 (1H,d,J=7.24Hz), 6.78 (1H,d,J=9.0Hz), 6.33 (1H,d,J=2.20Hz), 6.27 (1H,d,J=2.24Hz), 3.87 (3H, s,OMe), 3.83 (3H,s,OMe). ^{13}C NMR : (600 MHz, $CDCl_3$); δ ; 185.1 (C-9), 167.5 (C-3), 163.0 (C-1), 158.1 (C-4a), 150.18 (C-5a), 149.6 (C-7), 142.9 (C -8), 120.4 (C-6), 107.8 (C-8a), 105.6 (C-5), 102.4 (C-9a), 97.2 (C-2), 93.0 (C-4), 57.1 (OCH_3 – 7), 55.9 (OCH_3 – 3). ^{13}C and ^{1}HNMR are in agreement with literature [13]

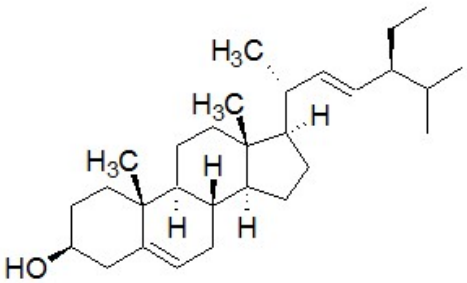
*Compound **3**:* Hexadecanoic acid (Palmitic acid); $C_{16}H_{32}O_2$ M^+: m/z 270 (fragments at; 227,185, 143, 121, 97, 74, 43). Run time: 3.43 min. IR ν_{max} cm^{-1} : 3076.72, 1704.62, 1667.67, 978.67, 958.72 for major peaks. ^{1}H NMR: (600 MHz, $CDCl_3$); δ 2.3 – 2.5 (m, 31H), ^{13}C NMR: (600 MHz, $CDCl_3$); δ 33.9, 31.9, 29.7, 29.7, 29.76, 29.6, 29.6, 29.4, 29.4, 29.3, 29.1, 28.8, 24.7, 24.6, 22.7, 14.1.

*Compound **5**:* Stigmasta-5,22- dien-3β-ol (Stigmasterol); colorless pleasant odour gel; $C_{29}H_{48}O$; M. pt 161 °C - 170 °C M^+ : m/z 413, IR ν_{max} cm^{-1} : 2954.08, 2900.00, 2852.55, 1460.10, 1377.16, 909.03, 721.63, 438.18 for major peaks. ^{1}H NMR: (400 MHz, $CDCl_3$); δ3.50 (H-3), δ 5.85 (m, H- 6), δ 4.60 (m, H-22, H-23), multiple signals at δ 1.50 - 2.00. ^{13} C NMR: (400 MHz, $CDCl_3$); δ; 1 C 42.2 (C-1), 56.4 (C - 2), 28.9 (C-3), 24.4 (C- 4), 57.0 (C- 5), 31.9 (C-6), 50.1 (C-7), 21.1(C-8), 39.8 (C- 9), 31.5 (C-10),121.7 (C-11), 140.7 (C-12), 36.3 (C- 13), 37.4 (C- 14), 31.7 (C-15), 76.7 (C-16), 42.4 (C-17), 12.2 (C-18), 40.5 (C-19), 21.1 (C- 20), 19.4 (C-21), 138.3 (C-23), 129.3 (C-24), 51.2 (C-25), 31.6 (C-26), 21.6 (C-27), 25.4 (C-28), 12.2 (C-29). ^{13}C and ^{1}H-NMR are in agreement with literature [14].

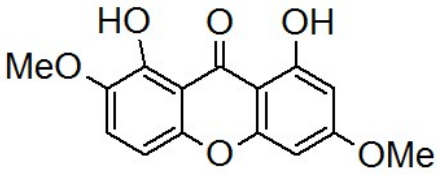
Figure 1:



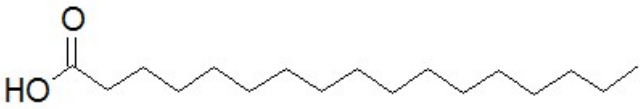
cpds 1, 4 (1-Hydroxy-3,7,8-trimethoxyxanthone) (Decussatin)



cpd **5**. Stigmasta-5, 22-dien-3-beta-ol (Stigmasterol)



cpd 2. (1,8- Dihydroxy- 3,7-Dimethoxyxanthone)
(Swertiaperennin)



cpd 3. Hexadecanoic acid

see Table 1.

Table 2

Compounds	FCA %
PEL	89.48 ± 1.00
PES	88.32 ± 0.52
Decussatin	77.80 ± 0.10
EDTA	118.50 ± 0.50

Acknowledgements

The authors are grateful to grateful to Mr. James Ofem and Mr. Patrick for harvesting the plants.

References

1. Graham, L.P. (2005). An Introduction to Medicinal Chemistry. Third Edition. Oxford University Press. p. 173-74, 685 - 86.
2. Iwu, M. M., Duncan A.R. and Okunji C. O. (1999). New Antimicrobials of Plant Origin. Perspectives on new crops and new uses. ASHS Press, Alexandria,VA. p. 457- 4
3. Burkill, H.M. (1997). The Useful Plants of West Tropical Africa, Royal Botanic Gardens.Kew. 4. p. 527 – 8.
4. Eloff, J.N. (1998). A sensitive and quick microplate method to determine the minimal inhibitory concentration of plant extracts for bacteria. Planta Medica 64. p.711 713Reference for DAD
5. Utz, C., Shadomy, S. Antifungal activity of 5-fluorocytosine as measured by disc diffusion susceptibility testing. J. Infect. Dis. 1977, 135, 970–974.
6. Dinis, T.C.P., Madeira, V.M.C and Almeida M.L.M (1994).Action of Phenolic derivates (acetoaminophen, salycilate and 5-aminosalycilate) as inhibitors of membrane lipid peroxidation and as peroxyl radical scavengers. Arch. Biochem. Biophys. 315. p.161-169.
7. Okorie, D. A New Phthalide and Xanthones from Anthocleista Djalonensis and Anthocleista vogelii. J. phytochem. 1776, 15, 1799 –1800.
8. Cardona, M.L., Fernandez,M.I., Pedro, J.R. and Serrano, A., (1990). Xanthones from Hypericum reflexum, Journal of Phytochemistry 29, p. 3003 - 3006.
9. Peres, V., Nagem, T.J., Oliveria, F.F. Tetraoxygenated naturally occurring Xanthones. J. Phytochem. 2000, 55, 683–710.
10. Cabantchik Z.I., Golenser J, Loyevsky M, Tsafack A. (1996). Iron chelators. Mode of action as antimalarials. Journal of Acta Haematol 95:70-77.
11. Van Zyl, R. L. (1992) Journal of Antimicrobial Chemotheraphy 30. p. 273-278.
12. Peres, V., Nagem, T.J. Naturally occurring pentaoxygenated, hexaoxygenated and dimeric xanthones : A literature survey. Química Nova 1997, 20, 388-397.
13. Valento, P., Andrade P.B., Silva, E.; Vicente, A., Santos, H., Bastos, M.L., Seabra, R.M. Methoxylated Xanthones in the quality control of small centaury (Centarium erythraea) flowering tops. J. Agr. Food Chem. 2002, 50, 460–463.
14. Li, C., P.B. BU, D.K. Yue and Y.F. Sun. (2006). Chemical Contituents from roots of Ficus hirta, Zhongguo Zhong. Yao. Za. Zhi., 31. p. 131 -133.

— § —

Table 1. Minimum Inhibitory Concentration (MIC) of PES, PEL and Decussatin against the eight strains of micro-organisms.

Test drugs/agents	E.c	K.p	B.s	S.a	T.m	T.r	A.f	C.a
PES	1.00	NOI	6.00	7.00	5.00	4.00	NOI	3.00
PEL	1.00	2.00	1.00	ND	5.00	4.00	NOI	3.00
decussatin	ND	ND	ND	ND	ND	ND	ND	ND
neomysin	0.09	1.56	0. 19	0.39	-	-	-	-
nystasin	-	-	-	-	0.1	0.1	0.1	0.1
canesten	-	-	-	-	0.1	0.1	0.1	0.1

Test strain

→ * MIC values in mgml^{-1}

* MIC values in mgml^{-1}

** All MIC values in mg/mL with the exception of Neomycin in µg/mL;*
ND: Not detected; within the tested concentration (8 mg/mL).
PES. A.vogelii stembark
PEL. A.vogelii leaves