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REGULAR ARTICLE

PRELIMINARY ANTIMICROBIAL AND PHYTOCHEMICAL STUDY OF THE AQUEOUS, ALCHOLIC AND CHLOROFORM EXTRACTS OF THE LEAVES OF *NAPOLEONAEA VOGELLI* HOOK. AND PLANCH. (*LECYTHIDIACEAE*)

Iriagbonse Asowata[1], *Joseph Omorogiuwa Erhabor*[1]*, *MacDonald Idu* [1], *Odaro Timothy*[1] *and Nosa Omoregbe Obayagbona*[2]

Address: Joseph Omorogiuwa Erhabor

[1] University of Benin, Faculty of Life Sciences, Department of Plant Biology and Biotechnology, P.MB.1154, 300001 Benin City, Edo State, Nigeria. +2348077979390

[2] Edo Environmental Consults and Laboratory, Microbiology laboratory, Palm House Annex, Sapele road, 300001, Benin City, Edo State, Nigeria

*Corresponding author: erhaborjoseph@yahoo.com

ABSTRACT

The antimicrobial activity and phytochemical analysis of *Napoleonaea vogelli* P. Beauv. {*Lecythidiaceae*} was done using aqueous, ethanol, methanol and chloroform leaf extracts to determine its antimicrobial and phytochemical constituents. The antimicrobial activities of the extracts were tested against bacteria and fungi isolates using the agar well diffusion method. Commercial antibiotics were used as positive reference standards to determine the sensitivity of the isolates. The leaf extracts of the plant were subjected to phytochemical analysis using standard experimental procedures. The extracts showed significant inhibitory activity against the test microbial isolates: *Escherichia coli, Bacillus subtilis, Staphylococcus aureus, Klebsiella pneumoniae, Proteus mirabilis, Pseudomonas aeruginosa, Penicillum notatum, Aspergillus niger, Fusarium oxysporum, Saccharomyces cerevisiae* and *Candida albicans*. The MIC values obtained using the Agar-dilution test ranged from 0.5-10mg/ml. The results demonstrated that the extracts of the leaves (*N. vogelli*) possess broad spectrum antimicrobial activity. These results suggest that it will be useful in the treatment of microbial infections.

Keywords: *Napoleonaea vogelli,* aqueous extract, ethanol extract, methanol extract, chloroform extract, antimicrobial activity, phytochemical analysis

INTRODUCTION

Medicinal plants are plants in which one or more of their organs contain substances that can be used for therapeutic purposes or which can be precursors for the synthesis of useful drugs (**WHO., 1977; Sofowora, 1982**). It is often said that every plant is a potential medicine for one disease or the other. Traditional healers have put forward many claims that are being subjected to scientific investigation (**Idu *et al.*, 2006**). Medicinal plants are now widely used in the treatment of microbial infections because of the problems of resistance to modern antimicrobial drugs (**Idu *et al.*, 2006**). Furthermore, it has been found that some drugs are synthesized from plants (**Idu *et al.*, 2007**). The plant *Napoleonaea vogelli* Hook. and Planch. belong to the family *Lecythidiaceae*. It is a tree that grows often more than 6m high. The fruit of *Napoleonaea vogelli* is yellowish or reddish when ripe and is often slightly warted. The fruits of *Napoleonaea vogelli* has been recommended for diabetes patients. The decoction of the leaves, bark and fruit rind is given for fever, cough and catarrh. The young shoots are used as chewing sticks for cleaning teeth.

The objective of this study was to investigate the antimicrobial and phytochemical properties of the alcoholic extracts of *Napoleonaea vogelli*.

MATERIALS AND METHODS

Plant materials

The leaves of *Napoleonaea vogelli* was collected from farmlands located around University of Benin and environs. It was identified by Professor M. Idu of the Department of Plant Biology and Biotechnology, University of Benin, Benin City. The procedure was described by **Onwuliri (2004)** and **Onwuliri and Wang (2005).** The leaves were dried in an open air under shade to prevent ultra violet rays from inactivating the chemical constituents. The leaves were grounded to fine particles. 50g of the leaves were soaked in 300ml of sterile water, 50g of grounded leaves were also soaked in 250ml ethanol, methanol and chloroform, all in bottles. They were left for 24hours at room temperature with occasional stirring. The

extracts were filtered and concentrated to dryness by evaporation and was stored in refrigerator until required for use.

Test organisms

The microbial isolates used for the study were: *Staphylococcus aureus, Bacillus subtilis, Escherichia coli, Pseudomonas aeruginosa, Klebsiella pneumoniae, Proteus mirabilis, Aspergillus niger, Penicillum notatum, Saccharomyces cerevisiae, Fusarium oxysporum* and *Candida albicans*. Pure cultures of the bacterial isolates and *Candida albicans* were obtained from the Department of Medical Microbiology, University of Benin Teaching Hospital, Benin City. Pure cultures of the molds and *Saccharomyces cerevisiae* were collected from the Microbiology laboratory, Edo Environmental Consults and Laboratory, Benin City, Nigeria.

Standardization of the microbial inoculum

All the test bacterial isolates and fungi isolates were sub-cultured on freshly prepared nutrient agar plates and malt extract agar plates and incubated for 24h and 48h respectively. Portions of the streaked colonies were transferred into test tubes containing 5ml of sterile nutrient broth and incubated for 3h at 37 ^{o}C. The growth of bacteria and fungi suspension obtained was compared to that of freshly prepared Barium sulphate solution {0.5ml of 1% Barium in Chloride to 99.5ml of 1% H_2SO_4 (0.36 Normal) **(Vandepitte *et al*., 2003).** The turbidity was adjusted by adding more sterile nutrient broth to match 0.5 McFarland standard (10^6 cfu/ml).

Test for antimicrobial activity

The diluted aqueous, ethanol, methanol and chloroform extracts of the leaves of the plant were tested for their antimicrobial properties using agar well diffusion method **(Pelezar *et al*., 1993**; **Barry and Thornsberry, 1995).** Also**,** commercially available standard antibiotic disc was placed on the agar plates as positive control after which the plates were incubated overnight at 37 ℃ and 25 ℃ for bacteria and fungi cultures respectively. At the end of the incubation period, the diameter of inhibition zone(s) were measured and recorded.

Minimum Inhibitory Concentration (MIC) Determination

The Minimum Inhibitory Concentration (MIC) of the aqueous and ethanolic leaf extracts of *N. vogelli* was determined by broth dilution method (Cheesebrough *et al.*, 2000). In it, a two-fold serial dilution method (double dilution) was used. The tubes were incubated at 37 ℃ for 24hours for bacteria and 25 ℃ for 48hours for fungi. The least concentration of the extract which inhibited the growth of the inoculum was considered as the minimum inhibitory concentration.

Minimum Bactericidal Concentration (MBC) Determination

The minimum bactericidal concentration (MBC) of the plant extracts was determined by a modification of the method of **Spencer and Spencer (2004).** Aliquots were taken from tubes with no visible growth in the MIC assay and sub-cultured on freshly prepared nutrient agar plates and later incubated at 37 ℃ for 24hours. The MBC was taken as the concentration of the extract that did not show any growth on a new set of agar plates.

Phytochemical screening of the extracts of the leaf

The aqueous, ethanol, methanol and chloroform leaf extracts were tested for the presence of saponins, flavonoids, tannins, phlobatannins, steroids, terpenoids, cardiac glycosides, alkaloids, anthracene and reducing sugar using standard procedures to identify the constituents as described by **Sofowora (1993)**, **Trease and Evans (1985)** and **Harborne (1973).** Quantitative determination was also carried out.

Statistical analysis

Results were expressed as means± standard error of means [S.E.M] and level of significance between means were computed by student's t-test using SPSS 14.00 computer software package. The level of significance was determined at 0.05.

RESULTS

The test microbial isolates exhibited varying degrees of sensitivity towards various concentrations of *Napoleonaea vogelli* aqueous leaf extract. The differences in the mean diameter of inhibitory zones exhibited by *B. subtilis*, *S. aureus*, *K. pneumoniae*, *P. notatum*, *A. niger*, *F. oxysporum* and *S. cerevisiae* was significant (P<0.05) (Table 1).

Table 1 The effect (zone of inhibition in mm) of *Napoleonaea vogelli* aqueous leaf extract at various concentrations on test organisms

Test organisms	Zone of inhibition (mm)			
	10mg/ml	5mg/ml	0.5mg/ml	Sterile distilled water
Bacillus subtilis	$14.5^{a} \pm 1.5$	$10.0^{ab} \pm 2.0$	$7.5^{b} \pm 0.5$	-
Escherichia coli	$10.5^{a} \pm 1.5$	$5.0^{a} \pm 2.0$	$4.0^{a} \pm 2.0$	-
Proteus mirabilis	$8.0^{a} \pm 2.0$	$5.0^{a} \pm 0.0$	$3.0^{a} \pm 1.0$	-
Pseudomonas aeruginosa	$3.0^{a} \pm 3.0$	$6.0^{a} \pm 1.0$	$4.0^{a} \pm 1.0$	-
Staphylococcus aureus	$13.0^{a} \pm 1.0$	$9.5^{b} \pm 0.5$	$0.0^{c} \pm 0.0$	-
Klebsiella pneumoniae	$10.5^{a} \pm 2.5$	$6.5^{ab} \pm 0.5$	$0.0^{b} \pm 0.0$	-
Penicillum notatum	$5.5^{a} \pm 0.5$	$3.5^{ab} \pm 0.5$	$1.0^{b} \pm 1.0$	-
Aspergillus niger	$11.0^{a} \pm 1.0$	$3.5^{b} \pm 0.5$	$2.0^{b} \pm 0.0$	-
Fusarium oxysporum	$15.0^{a} \pm 1.0$	$4.5^{b} \pm 2.5$	$0.0^{b} \pm 1.0$	-
Saccharomyces cerevisae	$12.5^{a} \pm 2.5$	$3.0^{b} \pm 1.0$	$1.0^{b} \pm 1.0$	-
Candida albicans	$3.0^{a} \pm 1.0$	$2.0^{a} \pm 1.0$	$0.0^{a} \pm 0.0$	-

Legend: Values are means ± S.E.M of two measurements across each zone of inhibition.
Means ± S.E.M with different superscript within a row are significantly different (P < 0.05).

P. notatum exhibited the highest mean inhibition zone (24.0 mm) whilst the least inhibitory zone (1.0 mm) was displayed by *S. cerevisiae* against a 0.5 mg/ml concentration of *Napoleonaea vogelli* ethanolic extract. The differences in the mean inhibitory zones ellicted by *B. subtilis, E. coli, P. aeruginosa, K. pneumoniae, A. niger, F. oxysporum, S. cerevisiae* and *C. albicans* was significant (P<0.05) (Tab 2).

Table 2 The effect (zone of inhibition in mm) of *Napoleonaea vogelli* ethanolic leaf extract at various concentrations on test organisms

Test organisms	Zone of inhibition (mm)		
	10mg/ml	5mg/ml	0.5mg/ml
Bacillus subtilis	$13.0^{a} \pm 1.0$	$6.0^{b} \pm 1.0$	$4.0^{b} \pm 1.0$
Escherichia coli	$8.0^{a} \pm 1.0$	$5.5^{ab} \pm 0.5$	$4.0^{b} \pm 1.0$
Proteus mirabilis	$8.5^{a} \pm 0.5$	$4.5^{b} \pm 0.5$	$3.0^{b} \pm 0.0$
Pseudomonas aeruginosa	$8.5^{a} \pm 1.5$	$5.0^{a} \pm 1.0$	$2.0^{b} \pm 2.0$
Staphylococcus aureus	$12.0^{a} \pm 5.0$	$6.0^{a} \pm 1.0$	$4.5^{a} \pm 0.5$
Klebsiella pneumoniae	$8.0^{a} \pm 0.0$	$5.5^{ab} \pm 0.5$	$2.0^{b} \pm 2.0$
Penicillum notatum	$24.0^{a} \pm 5.0$	$12.0^{a} \pm 3.0$	$8.0^{a} \pm 4.0$
Aspergillus niger	$12.5^{a} \pm 1.5$	$4.5^{b} \pm 1.5$	$2.5^{b} \pm 0.5$
Fusarium oxysporum	$19.0^{a} \pm 1.0$	$8.5^{b} \pm 0.5$	$3.0^{c} \pm 0.0$
Saccharomyces cerevisae	$11.5^{a} \pm 1.5$	$4.0^{b} \pm 0.0$	$1.0^{b} \pm 1.0$
Candida albicans	$16.0^{a} \pm 2.0$	$9.0^{b} \pm 1.0$	$5.5^{b} \pm 1.5$

Legend: Values are means ± S.E.M of two measurements across each zone of inhibition. Means ± S.E.M with different superscript within a row are significantly different, $P < 0.05$.

P. notatum exhibited the highest mean inhibition zone (18.5 mm) against 10mg/ml plant methanolic extract concentrate. The least inhibitory zone (0.0 mm) was displayed by *S. cerevisiae* against a 5 mg/ml concentration of *Napoleonaea vogelli* methanolic extract. The differences in the mean inhibitory zones ellicted by *B. subtilis, E. coli, S. aureus, K. pneumoniae, A. niger, F. oxysporum, S. cerevisiae* and *C. albicans* was significant ($P<0.05$) (Tab 3).

Table 3 The effect (zone of inhibition in mm) of *Napoleonaea vogelli* methanolic leaf extract at various concentrations on test organisms

Test organisms	Zone of inhibition (mm)		
	10mg/ml	5mg/ml	0.5mg/ml
Bacillus subtilis	$16.5^{a} \pm 2.5$	$9.5^{b} \pm 0.5$	$3.5^{b} \pm 0.5$
Escherichia coli	$16.0^{a} \pm 1.0$	$5.5^{b} \pm 0.5$	$2.5^{b} \pm 0.5$
Proteus mirabilis	$13.0^{a} \pm 2.0$	$7.0^{ab} \pm 1.0$	$4.5^{b} \pm 1.5$
Pseudomonas aeruginosa	$12.0^{a} \pm 6.0$	$5.5^{a} \pm 0.5$	$4.0^{a} \pm 1.0$
Staphylococcus aureus	$16.5^{a} \pm 3.5$	$8.0^{ab} \pm 1.0$	$6.5^{b} \pm 0.5$
Klebsiella pneumoniae	$9.0^{a} \pm 1.0$	$6.0^{b} \pm 0.0$	$3.5^{b} \pm 0.5$
Penicillum notatum	$18.5^{a} \pm 1.5$	$7.5^{b} \pm 1.5$	$0.0^{c} \pm 0.0$
Aspergillus niger	$17.0^{a} \pm 1.0$	$5.0^{b} \pm 1.0$	$2.5^{b} \pm 0.5$
Fusarium oxysporum	$12.5^{a} \pm 0.5$	$6.5^{b} \pm 0.5$	$4.5^{b} \pm 0.5$
Saccharomyces cerevisiae	$6.0^{a} \pm 1.0$	$1.0^{b} \pm 1.0$	0.0^{b}
Candida albicans	$20.0^{a} \pm 2.0$	$12.5^{b} \pm 0.5$	$7.0^{b} \pm 2.0$

Legend: Values are means ± S.E.M of two measurements across each zone of inhibition. Means ± S.E.M with different superscript within a row are significantly different, $P < 0.05$.

E. coli displayed the highest mean inhibition zone (11.0 mm) against 10mg/ml *Napoleonaea vogelli* methanolic extract concentrate. The least inhibitory zone (0.0 mm) was displayed by *P.mirabilis, P. aeruginosa, S. aureus, K. pneumoniae, P. notatum, A. niger, F. oxysporum, S. cerevisiae* and *C. albicans* against 5 mg/ml and 0.5mg/ml concentration of *Napoleonaea vogelli* methanolic extract. The differences in the mean inhibitory zones elicited by *B. subtilis, S. aureus, P. mirabilis, A. niger, S. cerevisiae* and *C. albicans* was significant ($P<0.05$) (Tab 4).

Table 4 The effect (zone of inhibition in mm) of *Napoleonaea vogelli* chloroform leaf extract at various concentrations on test organisms

Test organisms	zone of inhibition (mm)		
	10mg/ml	5mg/ml	0.5mg/ml
Bacillus subtilis	$4.0^{a} \pm 1.0$	$0.0^{b} \pm 0.0$	$0.0^{b} \pm 0.0$
Escherichia coli	$11.0^{a} \pm 1.0$	$3.5^{a} \pm 3.5$	$1.5^{a} \pm 1.5$
Proteus mirabilis	$3.0^{a} \pm 1.0$	$0.0^{b} \pm 0.0$	$0.0^{b} \pm 0.0$
Pseudomonas aeruginosa	$6.0^{a} \pm 1.0$	$2.0^{ab} \pm 2.0$	$0.0^{b} \pm 0.0$
Staphylococcus aureus	$8.5^{a} \pm 0.5$	$5.0^{b} \pm 1.0$	$0.0^{c} \pm 0.0$
Klebsiella pneumoniae	$3.0^{a} \pm 3.0$	$0.0^{a} \pm 0.0$	$0.0^{a} \pm 0.0$
Penicillum notatum	$3.5^{a} \pm 3.5$	$0.0^{a} \pm 0.0$	$0.0^{a} \pm 0.0$
Aspergillus niger	$3.0^{a} \pm 1.0$	$0.0^{b} \pm 0.0$	$0.0^{b} \pm 0.0$
Fusarium oxysporum	0.0	0.0	0.0
Saccharomyces cerevisae	$4.5^{a} \pm 0.5$	$0.0^{b} \pm 0.0$	$0.0^{b} \pm 0.0$
Candida albicans	$6.5^{a} \pm 0.5$	$3.0^{b} \pm 1.0$	$0.0^{c} \pm 0.0$

Legend: Values are means ± S.E.M of two measurements across each zone of inhibition. Means ± S.E.M with different superscript within a row are significantly different, $P < 0.05$.

P. mirabilis exhibited the highest inhibition zone (24 mm) against 10 µl gentamicin disc. *K. pneumoniae* and *E. coli* elicited the highest zone of inhibition (20 mm) against 10 µl ciprofloxacin disc. *Candida albicans* displayed the most sensitivity (22 mm) against 200 mg/ml ketoconazole disc (Tab 5).

Table 5 Antibiotic suspectibility patterns of the test organisms

Test organisms	Zone of inhibition (mm)		
	Gentamicin (10 µl)	Ciprofloxacin (10 µl)	Ketoconazole (200 mg/ml)
Proteus mirabilis	24		
Pseudomonas aeruginosa	20		
Klebsiella pneumoniae		20	
Staphylococcus aureus		18	
Escherichia coli		20	
Bacillus subtilis		15	
Penicillum notatum			20
Aspergillus niger			20
Fusarium oxysporum			18
Saccharomyces cerevisiae			16
Candida albicans			22

B. subtilis, E. coli, P. mirabilis, P. aeruginosa, S. aureus, K. pneumoniae, P. notatum and *C. albicans* had nil MIC against *N. vogelli* aqueous extract. *B. subtilis, P. mirabilis, S. aureus, K. pneumoniae* and *S. cerevisiae* displayed a MIC value of 10 mg/ml against *N. vogelli* chloroform extract (tab 6).

Table 6 Minimum inhibitory concentration of aqueous, ethanol, methanol and chloroform leaf extracts of *Napoleonaea vogelli* on test organisms

Test organisms	Concentration of extracts (mg/ml)			
	Aqueous	Ethanol	Methanol	Chloroform
Bacillus subtilis	0	0	0	10
Escherichia coli	0	5	5	5
Proteus mirabilis	0	5	0	10
Pseudomonas aeruginosa	0	5	0	5
Staphylococcus aureus	0	5	0	10
Klebsiella pneumoniae	0	5	0	10
Penicillum notatum	0	10	0	0
Aspergillus niger	10	0	0	0
Fusarium oxysporum	10	10	0.5	0
Saccharomyces cerevisiae	5	0.5	5	10
Candida albicans	0	0	10	0

All the bacterial isolates elicited nil MBC values against *N. vogelli* aqueous leaf extract. *E. coli, P. mirabilis, S. aureus* and *K. pneumoniae* exhibited a MBC value of 10 mg/ml against *N. vogelli* chloroform extract (tab 7).

Table 7 Minimum bactericidal concentration of aqueous, ethanol, methanol and chloroform leaf extract of *Napoleonaea vogelli* on test organisms

Test bacteria	Concentration of extracts (mg/ml)			
	aqueous	ethanol	methanol	Chloroform
Bacillus subtilis	0	0	0	0
Escherichia coli	0	5	5	5
Proteus mirabilis	0	5	0	10
Pseudomonas aeruginosa	0	10	0	5
Staphylococcus aureus	0	5	0	10
Klebsiella pneumoniae	0	5	0	10

Legend: 0 = Not bactericidal

The presence of saponins, tannins, steroids, flavonoids, terpenoids, cardiac glycosides and reducing sugars were detected in the aqueous extract of *N. vogelli*. Phlobatannins and alkaloids was absent from the analysed aqueous and alcoholic leaf extracts of *N. vogelli* (tab 8).

Table 8 Qualitative determination of leaf extracts of *Napoleonaea vogelli*

Phytochemical constituents	**Aqueous extract**	**ethanol extract**	**chloroform extract**	**methanol extract**
Saponins	+	+	-	+
Tannins	+	+	-	+
Phlobatannins	-	-	-	-
Steroids	+	+	+	+
Flavonoids	+	+	+	+
Terpenoids	+	-	+	+
Cardiac glycosides	+	+	-	+
Alkaloids	-	-	-	-
Reducing sugars	+	+	-	+
Anthracene	-	-	-	-

Legend: - = Absent, + = Present

Saponin (13.47%) and terpenoid (0.10%) were found to be present in the analysed *N. vogelli* leaves (Table 9)

Table 9 Quantitative determination of the phytochemical constituents of the leaves of *Napoleonaea vogelli*

Plant	**Tannin (%)**	**Saponin (%)**	**Terpenoid (%)**	**Flavonoid (%)**	**reducing sugar (%)**
Napoleonaea vogelli	*0.21± 0.02	13.47±3.66	0.10±0.04	1.53±0.23	9.75±2.08

Legend: * mean ±Standard error of mean (SEM)

DISCUSSION

The aqueous, ethanol and methanol leaf extracts of *N. vogelli* showed strong antimicrobial activities against all test organisms than the chloroform extract. It was observed that sensitivity increased with increasing concentration of extracts. *N. vogelli* exhibited broad spectrum inhibitory activities against both Gram positive and Gram negative test bacterial isolates as well as the fungal isolates. *Napoleaneae vogelli* can be used for gastrointestinal

disorders. *Staphyloccocus aureus* is known to play a significant role in skin diseases including superficial and deep follicular lesion, so the strong activity of the aqueous, ethanol, methanol and chloroform extracts of *N. vogelli* indicated that they can be effective against skin infections. *Proteus mirabilis, Escherichia coli* and *Klebsiella pneumoniae* are the most cause of urinary tract infection.

Considering the minimum inhibitory concentration (MIC) values of the aqueous, ethanol, methanol and chloroform extracts of the plant, the most potent activities were against *Staphylococcus aureus, Proteus mirabilis, Pseudomonas aeruginosa, Klebsiella pneumoniae, Escherichia coli, Saccharomyces cerevisiae* and *Fusarium oxysporum*. This confirms its use by traditional medical practitioners for the treatment of stomach disturbances, urinary tract infections, skin infections and fungal infections. These results agreed with the findings of **Kurosaki and Nishi (1983)** that higher concentration of antimicrobial substances of the same extracts could show appreciably more growth inhibitions in being both bacteriostatic and bacteriocidal.

The plant being bacteriocidal is of great value. This is because as drug extract, administered for treatment, there would be no form of re-occurrence of the infection. Considering the aqueous, ethanol, methanol and chloroform extracts of *N. vogelli*, it was found that they do not contain alkaloids as against the extracts of *N. imperialis* in which only the aqueous extract contain alkaloid as reported by **Idu *et al*. (2011).** The methanol extract exhibited a stronger antimicrobial effect on all test organisms as compared with all other extract of *N. vogelli* (aqueous, ethanol and chloroform). The highest zone of inhibition was displayed by the test yeast: *Candida albicans* (20.0$\pm$ 2.0mm at 10mg/ml) and the test bacterium; *Bacillus subtilis* (16.5$\pm$2.5mm at 10mg/ml). Identifying the phytochemical constituents can help one to speculate on the medicinal value of the leaves of *N. vogelli.* Tannins have antimicrobial **(Ya *et al*., 1988)** and antioxidant properties. **Parekh and Chanda (2007**) reported that tannins are known to react with proteins to provide the typical tanning effect which is important for the treatment of inflamed or ulcerated tissues. These observations therefore support the use of *N. vogelli* in herbal remedies. The presence of tannins also showed that the leaves of the plant could be used as purgative, for cough, asthma and hay fever according to **Gill (1992).** Saponin was found to be present in *N. vogelli* and supported the usefulness of these plants in managing inflammation. Saponins have been shown to have immense significance as antihypercholesterol, hypotensive and cardiac depressant properties **(Trease and Evans, 1985; Price, 1987).** Steroidal compounds present

in *N. vogelli* extracts are of importance and interest due to their relationship with various anabolic hormones including sex hormones **(Okwu, 2001).** Cardiac glycosides have been used for over two centuries as stimulants in cases of cardiac failure **(Trease and Evans, 1985; Olayinka *et al*., 1992).** This perhaps justifies the functions of the plant in treatment and management of hypertension.

CONCLUSION

N. vogelli extracts are potentially useful antimicrobial agents. Bioactive substances from this plant can therefore be employed in formulation of antimicrobial drugs for the treatment of various bacterial and fungal infections and can be used for treatment of chronic and degenerative diseases such as cardiovascular diseases.

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