Antimicrobial and Physical Properties of Herbal Ointments Formulated with Methanolic extracts of *Persea americana* seed and *Nauclea latifolia* stem bark

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**ABSTRACT**

**Aims:** In this study the antimicrobial and physical properties of herbal ointments formulated with methanolic extracts of *Persea americana* seeds and *Nauclea latifolia* stem bark were evaluated.

**Methodology:** The preliminary in-vitro antimicrobial activity of the methanolic extracts *Persea americana* seeds and *Nauclea latifolia* stem bark was determined using Mueller Hinton agar for bacteria isolates (*Staphylococcus aureus*, *Bacillus subtilis*, *Escherichia coli* and *Pseudomonas aeruginosa*) and Sabouraud Dextrose agar for fungi isolates (*Candida albicans* and *Aspergillus niger*). Five formulations of the herbal ointment were prepared by mixing varying concentrations of the extracts with Emulsifying ointment B.P. The results from antimicrobial evaluation of the formulated ointments were compared to those of commercial brands [Gentamicin cream and Clotrimazole cream (Ytacan®)].

**Results:** The diameter of zones of inhibition (mm) of the Persea extract (400mg/ml) on the growth of microorganism were 19 ± 0.3 (*Pseudomonas aeruginosa*), 20 ± 0.5 (*Escherichia coli*), 12 ± 0.2 (*Bacillus subtilis*), 20 ± 0.3 (*Staphylococcus aureus*) and 20 ± 0.4 (*Candida albicans*) while that of Nauclea extract (400mg/ml) were 18 ± 0.2 (*Pseudomonas aeruginosa*), 13 ± 1.1 (*Bacillus subtilis*) and 14 ± 0.1 (*Candida albicans*). No growth was observed for all the extracts against *Aspergillus niger*. The Minimum inhibitory concentration (mg/ml) values of Persea and Nauclea extracts were *Pseudomonas aeruginosa* 50, 200; *Staphylococcus aureus* 200, nil; *Escherichia coli* 50, nil; *Bacillus subtilis* 200, 400; *Candida albicans* 200, 400. The formulation containing only *P. americana* exhibited better antimicrobial activity than that of *N. latifolia*. The formulated ointments possessed good physical characteristics and comparably lesser antimicrobial properties to that of Gentamicin and Ytacan®.

**Conclusion:** This study showed that the methanolic extracts of *Persea americana* seeds and *Nauclea latifolia* stem bark possess antimicrobial activities and also has high potential as antimicrobial agent when formulated as ointment. The herbal ointment containing *P. americana* extract compared favourably with commercial brand for bacteria but was less potent for fungi.

**KEYWORDS:** *Persea americana*, *Nauclea latifolia*, herbal ointments, antimicrobial activity

**INTRODUCTION**

Bacterial and fungal pathogens have evolved numerous defence mechanisms against antimicrobial agents, and resistance to old and newly produced drugs are on the rise1. Most antibiotics are no longer active against the targeted organisms leading to occurrence of antibiotic resistant organisms1. In addition, majority of the orthodox drugs are both expensive and display numerous side effects on the users. As a result, managing patients especially in developing countries is rather expensive. Discovering and identifying new safe drugs without severe side effects has become an important goal of research in biomedical science. Plants are rich in a wide variety of secondary metabolites, such as tannins, terpenoids, alkaloids, and flavonoids; which have been found in-vitro to possess antimicrobial properties1. *Persea americana* Mill. (Lauraceae) is a plant from Central America but it has shown easy adaptation to other tropical regions. Its fruits are commonly known as avocados. The by-products like avocado seeds are normally discarded which globally can lead to increased numbers of insects and rodents and also incur economic losses due to the high cost of transporting these by-products to disposal areas. Studies to investigate the benefits of these by-products as sources of food supplements or medicinal...
products are needed. Many parts of the avocado tree have a use in herbal medicine. The leaves and bark are effective remedies for digestive problems, coughs, promotion of menstrual flow, clearing high uric acid levels which cause gout and treatment of liver obstructions. The leaves and bark of *Persea americana* were found to possess antibacterial properties. The leaves and bark are effective remedies for digestive problems, coughs, promotion of menstrual flow, clearing high uric acid levels which cause gout and treatment of liver obstructions. The fruit is extremely nutritious and has a wide range of medicinal use by serving as emollient and carminative. The oil is used when expressed from the avocado seed as nourishment for the skin. It softens dry or flaking skin and, massaged in to the scalp, it improves hair growth. Guzmán-Rodríguez et al. reported that avocado fruit extracts from *P. americana* are known to exhibit antimicrobial properties. Among other uses, *Persea americana* seeds are widely used to treat diarrhoea and dysentery caused by intestinal parasites.

*Nauclea latifolia* Smith (Rubiaceae) is a straggling, evergreen, multi-stemmed shrub or small tree native to tropical Africa and Asia. The plant has rough bark; leaves are 7 by 4 to 5 inches and are glabrous obovate. Extracts of different parts of the plant *Nauclea latifolia* (e.g. fruits, leaves, stem bark and roots) in hot water or alcohol are used in form of infusions, decoctions or concoctions. The plant, *Nauclea latifolia* is used in many African countries for the treatment of various ailments including bacterial diseases. *Nauclea latifolia* stem bark has been found to have antimicrobial properties.

Most of these studies have confirmed that the nature of the extracting solvent and the extraction processes employed play very crucial roles in the phytochemicals present in the extracts and hence their medicinal properties. However, there are still limited studies on their formulation into dosage forms. Formulating the extracts into dosage forms will not only improve their acceptability but also stability. The aim of this study is to evaluate the antimicrobial properties of methanolic extracts of seeds of *Persea americana* and stem bark of *Nauclea latifolia*, their combinations and ointment formulations.

**MATERIAL AND METHODS**

**Materials**

*Persea americana* seeds and *Nauclea latifolia* stem bark were purchased on 11th of August 2014 from a local store in Mushin, Lagos Nigeria. They were identified and authenticated at the herbarium of the Department of Botany, University of Lagos, Nigeria with voucher number LUH 6211 for *Persea americana* and LUH 6179 for *Nauclea latifolia*.

The test organisms which include bacteria: *Staphylococcus aureus*, *Bacillus subtilis*, *Escherichia coli*, *Pseudomonas aeruginosa* and fungi: *Candida albicans* and *Aspergillus niger* were clinical isolates obtained from Lagos University Teaching Hospital (LUTH) Nigeria. The bacterial strains were maintained in Muller Hinton (MHA, pH 7.2) at 37±1 °C and fungi were maintained in Sabouraud dextrose agar (SDA, pH 5.4) at 25±1 °C.

Commercial brands of ointment used include Gentamicin ointment (Gentamicin 1%w/w in the form of Gentamicin Sulphate manufactured by Drugfield Pharmaceuticals Limited, Lynson Chemical Avenue, KM 38, Lagos-Abeokuta Expressway, Sango-Otta, Ogun State, Nigeria) and Ytacan® cream (Clotrimazole U.S.P. 1% w/w, manufactured by Strides, off Oba Akran Avenue, Ikeja Industrial Area, Lagos, Nigeria) were purchased from a registered pharmacy, while methanol and Emulsifying ointment B.P. were obtained from Department of Pharmaceutics and Pharmaceutical Technology, Faculty of Pharmacy, University of Lagos.

**Methods**

**Preparation of Persea americana and Nauclea latifolia extracts**

The maceration method was employed for the two extractions. The barks of the seeds collected from the matured plant of *Persea americana* were peeled and chopped into smaller pieces and oven-dried at 40°C for 7 days. After grinding the weight recovered from the seeds was 1300g and 800g was weighed and macerated using 100% methanol as solvent. The extracts gotten were concentrated using a rotary evaporator at 40°C and speed of 85rpm (revolution per minute) and a dark brown extract was obtained from the seeds. The extract was further dried in the oven to obtain solid form of the extract. The stem bark of the mature plant of *Nauclea latifolia* was extracted in a similar way as barks of the seeds the matured plant of *Persea americana*. The extracts were stored in air tight containers at 4°C for further use.

**Phytochemical Screening**

The two plants extracts were screened for the presence of alkaloids, saponins, tanins, anthraquinones, cardiac glycosides, reducing sugar and flavonoids using methods reported elsewhere.

**Determination of antimicrobial activity of the extracts**

The antimicrobial activity was carried out as detailed in an earlier study. Briefly, 1ml of the calibrated organisms (*Staphylococcus aureus*, *Bacillus subtilis*, *Escherichia coli* and *Pseudomonas*...
aeruginosa, Candida albicans and Aspergillus niger) each was transferred into different sterile petri dish, and then the melted 25mls of agar was poured into it and allowed to solidify. Four wells were bored on each petri dish using sterile cork borer and all these were done in aseptic zone to prevent invasion of foreign microorganisms. Then 0.2mls of concentration of 400mg/ml, 200mg/ml and 100mg/ml of the extract were poured into separate wells and the 2% methanol as control in the last well. The bacteria were incubated for 24hrs at 37°C and 27°C for fungi, after which the diameter of zones of inhibitions were determined.

Minimum Inhibitory Concentration (MIC) of extracts

Four concentrations (25mg/ml, 50mg/ml, 100mg/ml, 200mg/ml) of Persea americana and four concentrations (50mg/ml, 100mg/ml, 200mg/ml, 400mg/ml) of Nauclea latifolia extracts were prepared by dissolving the extracts in prepared agar and were mixed thoroughly and transferred into sterile petri dishes to which the organisms were added the second day. This was done by incorporating a loopful of the test isolates and streaking, the plates were incubated for 72 hours and the results obtained were recorded.

Formulation of Ointment

Five different ointment formulations (A, B, C, D and E) were prepared by with emulsifying ointment B.P (Table 1). Emulsifying ointment of 20g was prepared by melting 6g of emulsifying wax, 10g of white soft paraffin and 4.8mls of liquid paraffin in a melting pan on water bath at a temperature of 70°C. Then the extracts were dissolved in sterile distilled water, after which they were added to the emulsifying ointment with continuous stirring. This mixture was stirred continuously until a homogenous mixture was obtained and transferred into clean cream jars.

Physical Evaluation of Formulated Ointments

The evaluation of the ointments was carried out over a period of 10days, the following parameters such as odour; colour, appearance, texture and pH were noted.

For the determination of spreadability, excess sample was applied in between two glass slides and was compressed to form a thick by placing 300mg weight for 5minutes. Weight was added to the pan. The time required to separate the two slides (i.e. the time in which the upper slide moves over the lower plate was taken as the spreadability) was noted. Spreadability was calculated using equation 1.

$$ S = \frac{ML}{T} $$

Equation 1

Where S is spreadability, M is mass tide to the upper slide, L is length move on the glass slide and T is time taken.

Determination of antimicrobial activity of formulated ointments

The antibacterial activities of various combinations of formulated ointments of Persea americana and Nauclea latifolia against various microorganisms were evaluated by agar diffusion method. Mueller Hinton agar was the medium used for bacterial organisms (Bacillus subtilis, Staphylococcus aureus, Escherichia coli, Pseudomonas aeruginosa) while Sabouraud dextrose agar was used for fungal organisms (Candida albicans and Aspergillus niger). The bacteria were incubated at 37°C while the fungi at 25°C for 24 hours.

Statistical Analysis

Data obtained was expressed as mean ± standard deviation. The ANOVA test was employed to assess if there were any difference in the obtained data. p-values less than 0.05 were considered statistically significant.

RESULTS

Persea extract gave a dark brown colour with percentage yield of 38.6% while Nauclea extract gave a brownish orange extract with percentage yield of 24.8%.

Phytochemical Screening

The phytochemical screening of the seeds of Persea americana revealed the presence of alkaloids, saponins, tannins, cardiac glycoside, reducing sugar, flavonoids and absence of anthraquinones while that of the extract of the bark of Nauclea latifolia revealed the presence of alkaloids, saponins, tannins, cardiac glycoside, reducing sugar and anthraquinones and absence of flavonoids.

Antimicrobial activity of the extracts

The antimicrobial susceptibility results of the test microorganisms to the extracts of Persea americana seeds and Nauclea latifolia stem bark are presented in table 2 while results of the minimum inhibitory concentrations (MIC) are presented in table 3.
Antimicrobial and Physical Properties of Herbal Ointments Formulated with Methanolic ointment base used in the formulation of the extracts did not show any inhibition zone.

All the five formulated ointments (A to E) had good appearance and fine texture. While formulation A containing only Persea americana extract had dark brown colour, characteristic smell, pH of 6.11 ±0.24, formulation E containing only Nauclea latifolia extract had reddish orange colour, pungent smell and pH of 5.47 ±0.25. Formulations B, C and D containing the mixture of the two extracts in different proportions showed pH within a broad range of 5.25 to 6.50.

Table 1: Composition of the extracts of Persea americana and Nauclea latifolia in the formulated ointments

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Formulation A(%w/w)</th>
<th>Formulation B(%w/w)</th>
<th>Formulation C(%w/w)</th>
<th>Formulation D(%w/w)</th>
<th>Formulation E(%w/w)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Persea americana</td>
<td>24.2</td>
<td>18.1</td>
<td>12.1</td>
<td>6.1</td>
<td>0</td>
</tr>
<tr>
<td>Nauclea latifolia</td>
<td>0</td>
<td>6.1</td>
<td>12.1</td>
<td>18.1</td>
<td>24.2</td>
</tr>
<tr>
<td>Emulsifying oil</td>
<td>75.8</td>
<td>75.8</td>
<td>75.8</td>
<td>75.8</td>
<td>75.8</td>
</tr>
<tr>
<td>Total weight</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
</tbody>
</table>

Table 2: Antimicrobial susceptibility of test organisms to the extracts of Persea americana and Nauclea latifolia (diameter of zone of inhibition, mm)

<table>
<thead>
<tr>
<th>Extract/ Solvent</th>
<th>Concentration (mg/ml)</th>
<th>Pseudomonas aeruginosa</th>
<th>Escherichia coli</th>
<th>Bacillus subtilis</th>
<th>Staphylococcus aureus</th>
<th>Candida albicans</th>
<th>Aspergillus niger</th>
</tr>
</thead>
<tbody>
<tr>
<td>Persea americana</td>
<td>100</td>
<td>15±0.1</td>
<td>15±1.1</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>200</td>
<td>17±0.2</td>
<td>19±0.4</td>
<td>11±1.3</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>400</td>
<td>19±0.3</td>
<td>20±0.5</td>
<td>12±0.2</td>
<td>20±0.3</td>
<td>20±0.4</td>
<td>-</td>
</tr>
<tr>
<td>Nauclea latifolia</td>
<td>100</td>
<td>12±0.5</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>200</td>
<td>16±0.1</td>
<td>-</td>
<td>13±1.1</td>
<td>-</td>
<td>14±0.1</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>400</td>
<td>18±0.2</td>
<td>-</td>
<td>13±1.1</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>2% Ethanol</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

- indicates no zone of inhibition, Values are the means of triplicates with standard deviations

Table 3: Minimum inhibitory concentration (MIC) of Persea and Nauclea extracts on the test organisms.

<table>
<thead>
<tr>
<th>Extract</th>
<th>Pseudomonas aeruginosa</th>
<th>Escherichia coli</th>
<th>Bacillus subtilis</th>
<th>Staphylococcus aureus</th>
<th>Candida albicans</th>
</tr>
</thead>
<tbody>
<tr>
<td>Persea americana</td>
<td>50</td>
<td>50</td>
<td>200</td>
<td>200</td>
<td>200</td>
</tr>
<tr>
<td>Nauclea latifolia</td>
<td>100</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>200</td>
</tr>
</tbody>
</table>

ND means not detectable

Table 4: Antimicrobial susceptibility of test organisms to the formulated ointments and the standards (diameter of zone of inhibition, mm)

<table>
<thead>
<tr>
<th>Organisms</th>
<th>Formulations / Ratio</th>
<th>Strength</th>
<th>Commercial brands</th>
</tr>
</thead>
<tbody>
<tr>
<td>Staphylococcus aureus</td>
<td>A P:100%</td>
<td>B P:N(3:1)</td>
<td>C P:N(1:1)</td>
</tr>
<tr>
<td>Bacillus subtilis</td>
<td>20±0.3</td>
<td>18±0.1</td>
<td>12±0.1</td>
</tr>
<tr>
<td>Pseudomonas aeruginosa</td>
<td>11±0.2</td>
<td>11±0.4</td>
<td>-</td>
</tr>
<tr>
<td>Escherichia coli</td>
<td>18±0.2</td>
<td>20±0.3</td>
<td>20±0.3</td>
</tr>
<tr>
<td>Candida albicans</td>
<td>19±0.2</td>
<td>16±0.1</td>
<td>12±0.2</td>
</tr>
<tr>
<td>Aspergillus niger</td>
<td>19±0.2</td>
<td>18±0.4</td>
<td>13±0.2</td>
</tr>
</tbody>
</table>

Note: The amount strength of Formulations A to E used was 640mg/ml of the single or combined extract while 40mcg/ml was used for the commercial brands. Values are the means of triplicates with standard deviations.
P = Persea americana, N = Nauclea latifolia, GM = Gentamycin and YC = Ytacan®, -= No growth.
DISCUSSION

Phytochemical Screening
The presence of phytochemical substances has been reported to exhibit varied biochemical and pharmacological effects in animals and microorganisms when ingested\(^5\). The presence of saponins and tannins could be the basis for the antimicrobial activity which has been associated with antimicrobial effects. Tannins act by coagulating the cell wall proteins\(^1,3\). while saponins cause the lysis of the bacterial cell. Tannins are also reported to have various physiological effects like anti-irritant, anti-secretolytic, anti-phlogistic, antimicrobial and anti-parasitic effects. Phytotherapeutically, tannin-containing plants are used to treat nonspecific diarrhoea, inflammations of mouth and throat and slightly injured skins. Thus, this may therefore explain the demonstration of antimicrobial activity by the methanolic extracts of stem bark of \textit{Nauclea latifolia} and seeds of \textit{Persea americana}. De and Ifeoma\(^4\) reported that the phytochemical components also offer plants themselves protection against infection by pathogenic microorganisms. These results obtained in the phytochemical screening correlate with the work done by Hotellier et al.\(^5\) and Morah\(^6\) who reported that \textit{Nauclea latifolia} contains terpenes, alkaloids, glycoalkaloids and tannins. Flavonoids are known to be synthesized by plants in response to microbial attack. Hence, it should not be surprising that they have been found to be effective antimicrobial substances against a wide array of microorganisms, when tested \textit{in-vitro}. Their activity is probably due to their ability to react with extracellular and soluble proteins and to complex with bacterial cell walls leading to the death of the bacteria\(^7\). The inhibitory activity exhibited by the secondary metabolites tends to agree with the reports of Adeyemi et al.\(^8\) which linked the antimicrobial properties of plants to the presence of secondary metabolites. The differences in the antimicrobial properties of the extracts from \textit{Persea americana} and \textit{Nauclea latifolia} might be attributed to the differences in the composition and concentration of the phytochemicals.

Antimicrobial properties of Extracts
The findings in this study revealed that the extract of the seeds of \textit{Persea americana} possessed \textit{in-vitro} antimicrobial activity against \textit{Staphylococcus aureus}, \textit{Escherichia Coli}, \textit{Pseudomonas aeruginosa}, \textit{Bacillus subtilis} and \textit{Candida albicans} with MIC (mg/ml) of 200, 50, 50, 200 and 200 respectively while \textit{Nauclea latifolia} possessed \textit{in-vitro} antimicrobial activity against \textit{Pseudomonas aeruginosa} and \textit{Candida albicans} with MIC (mg/ml) of 100 and 200 respectively (Table 3). The activity exhibited by the extracts against the clinical bacterial and fungal isolates that are associated with various infections, may provide scientific justification for the ethnomedicinal uses of the seeds of the plant. The inhibitory effect observed on \textit{Pseudomonas aeruginosa} by both indicates that the plants might possess some wound healing property if further purified. The antimicrobial activity is concentration dependent, the higher the concentration, the greater is the rate at which inhibition zone diameter increases\(^4\). From this study \textit{Persea americana} extract generally showed better antimicrobial activity and larger diameter of zone of inhibition and lower MIC (Tables 2 and 3) than \textit{Nauclea latifolia} extract to the tested organisms. The difference might be due to differences in their phytochemical evaluations as reported earlier. This study is in line with previous studies\(^4,7,8\) which have claimed that \textit{Persea americana} plant parts such as leaves, seeds and stem possess antimicrobial activity. Various factors affect the potency of the extracts such as age of plant, solvent used for extraction, method of extraction and drying, method and medium of storage of extract. The poor antimicrobial activity of the \textit{Nauclea latifolia} extract is not in line with earlier study\(^5\) which has claimed that it possesses activity against \textit{Escherichia coli}, \textit{Staphylococcus aureus} and \textit{Aspergillus niger} and may be attributed to the extraction procedures.

Antimicrobial and physical properties of the formulated ointments
The herbal ointments containing the extracts of \textit{Persea americana}, \textit{Nauclea latifolia} and combination of the two extract in different concentrations demonstrated some antimicrobial activity against some tested organism (Table 4). Generally, there is a significant difference between formulation A (containing only the extract of \textit{Persea americana}) or B (containing \textit{Persea americana} 75% and \textit{Nauclea latifolia} 25%) and formulations C, D and E containing equal or more concentration \textit{Nauclea latifolia} extracts. Formulation A with only \textit{P. americana} extract possessed more antimicrobial activity when compared with that containing only \textit{N. latifolia} extract. There is no significant difference between the antibacterial activity of formulation A and B against \textit{Bacillus subtilis} and \textit{Candida albicans} but Formulation B had a better activity against \textit{Pseudomonas aeruginosa}. All the formulated ointments were inactive against \textit{Aspergillus niger} as expected. The pathogens were sensitive to the commercial brands, Gentamicin (antibacterial agent) and Ytacan\(\textregistered\) (antifungal agent) which gave larger zones of mean diameter inhibition than the formulated ointments and extracts even at lower concentration (40mcg/ml) used for the study (Table 4). The commercial brands, (Gentamicin and Ytacan\(\textregistered\))
showed higher activity on the test microorganisms when compared to the crude extracts of *Nauclaea latifolia* and *Persea americana* their formulations. This could be due to their higher degree of purity of Gentamicin and Ytacan®. Impurities or contaminants in the extracts and lack of standardization could have been responsible for the reduced activities of the extracts on the test clinical isolates.

However, the crude extracts exhibited better antimicrobial activity than the ointment formulations containing the corresponding extracts. This may be attributed to the various ingredients incorporated in the formulation of the ointments and the formulation process employed. For the combined extracts, the formulations containing equal or higher proportion of the Persea extract showed better antimicrobial activity than the formulation containing higher proportion of the Nauclea extract and only Nauclea. Formulations B and C showed relatively better activity against *Pseudomonas aeruginosa* than other formulations. The activity of *Persea americana* against *Candida albicans* and *Staphylococcus aureus* is of significant interest because it is commonly found and widely encountered. *Staphylococcus aureus* is not easily eliminated from deep skin layer, sweat gland, sebaceous gland and hair follicle by routine washing and scrubbing even with antiseptic soap11. Also *Candida albicans* which causes vagina candidiasis is not easily washed away and these extracts can be formulated into pessaries for the treatment.

All the prepared ointments showed smooth appearance with good spreadability. *P. americana* extract and formulated ointment exhibited good antimicrobial activity on *Escherichia coli* and would provide suitable means of treating skin infections related to the organism.

The pH values of all the formulated ointments ranged from 5.25 to 6.50, which are considered acceptable to avoid the risk of irritation upon application to the skin. The pH values of formulated ointments fall within the range of human skin and are therefore compatible with the human skin.

**CONCLUSION**

This study shows that methanolic extracts of *Persea americana* seeds and *Nauclea latifolia* stem bark possess antimicrobial properties and have potency against some bacteria and fungi when formulated as ointment for topical use for the treatment of common skin diseases. However, both extracts had no activity against *Aspergillus niger* while *P. americana* showed more activity over most of the test organisms than *N. latifolia*. The antimicrobial activity of the formulated ointments was lower than those of the commercial brands. However on *Escherichia coli* and *Pseudomonas aeruginosa*, *P. americana* extract and formulated ointments containing more proportion of *P. americana* extract showed a remarkable activity. The formulation can therefore be employed in treating skin infections caused by any of the susceptible organisms. The physical properties of the formulated extracts were similar to those of commercial brands used in the study.

**REFERENCES**


