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Antimicrobial analysis of *Persia Americana* leaf

Amuda MO[1], Edewor TI[1], Oderinlo OO[2*]

1.Ladoke Akintola University of technology, Ogbomoso

ABSTRACT

The pulvurised leaves of *P. americana* was extracted with methanol and the crude extract obtained was subjected to antimicrobial screening against clinically isolated organisms from patients with infectious wounds, the organisms include; *E colli spp, Klebsellia spp, and Pseudomona spp.*, *Klebsiellia spp* only produce inhibitory activity against the extract with minimum inhibitory concentration (MIC) ranging from 0.05-0.15mg/ml. The methanolic extract showed no zone of inhibition with *E. coli* and *Psuedomonas spp* indicating that it does not have any inhibition activity against such bacteria with minimum inhibitory concentration (MIC) ranging from 0.05-0.15mg/ml.

Keywords: Antimicrobial, *Persia americana*, methanolic extract.

*Corresponding Author Email: talk2mt@yahoo.com

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INTRODUCTION

Antimicrobial agents are essentially important in reducing the global burden of infectious diseases (Bhatia and Narain, 2010)[1]. Antibiotics resistance is worldwide problem which is rapidly increasing in both hospitals and the community involved in morbidity and health care (Mill *et al*, 2015)[2]. Antibiotic resistance is problem that continues to challenge the health care sector in a large part of the world in both developing and developed countries (Sarita *et al*, 2019)[3].

The drug metabolites produced from synthetic drug sources exert fewer therapeutic effects with adverse side effects. However, therapeutic agents formed from natural sources may avoid the side effects, as they produce physiological and pharmacological effects within living cells. Plants remain important as a primary healthcare mode for approximately 85% of the world's population (Peršić , 2015)[4], and as a resource for drug discovery, with 80% of all synthetic drugs deriving from them (Bauer and Brönstrup, 2014)[5].

However, emergence and dissemination of multidrug resistant (MDR) strain in pathogenic bacteria have become significant public health threat as there are fewer, or even sometimes no, effective antimicrobial agents available for the infection caused by pathogenic bacteria (Boucher *et al*, 2009; Giamazellou, 2010)[6,7]. *Persea americana*, commonly called avogado, is a flowering plant of lauraceae family and a native to central Mexico. It is well known throughout the world for its fruit which is highly consumed (Rahman, 2018)[8] because of its peculiar nutritional composition, antioxidant content, and biochemical profile. These study, estimate the antibacterial activity of methanol extract of *persea ameriacana* leaf against sensitive bacteria strain isolated from patients having wound infections. Plant extract contained a very complex structure with the active ingredients present in the form of natural organic compounds. The process of extraction for a particular compound is dependent on the solubility of component in the solvent (water or organic solvent) (Masounmian and Zandi, 2017)[9].

Persia Americana Plant

Persea Americana is a native plant of central mexico, classified in the flowering plant family lauraceae. They are commonly valuable and are cultivated in tropical and mediteranean climates throughout the world. The tree grows to 20m, with alternately arranged leaves 12-25cm long, the flowers are inconspicuous, greenish-yellow 5-10mm wide. The plant is remarkably versatile as to soil adaptability, doing well on such diverse types as red clay, sand, volcalnic loam, or lateritic soil.it has been found healthier on nearly neutral or slightly alkaline soil than on moderately or highly acid soil.

Taxonomy

Synonyms(s): *Laurus persea L, Persea drymifolia Schlecht, and cham,Persea gatissima Gaertn.f.,*

Common names

Amharic: *avocado*

English: *alligator pear, avocado-pear, butter fruit*

Filipino*: avocado*

French: *avocet, avocatier, zabelbok, zaboka*

Current name: *Persea Americana*

Authority: *Miller*

Filipino*:* *avocado*

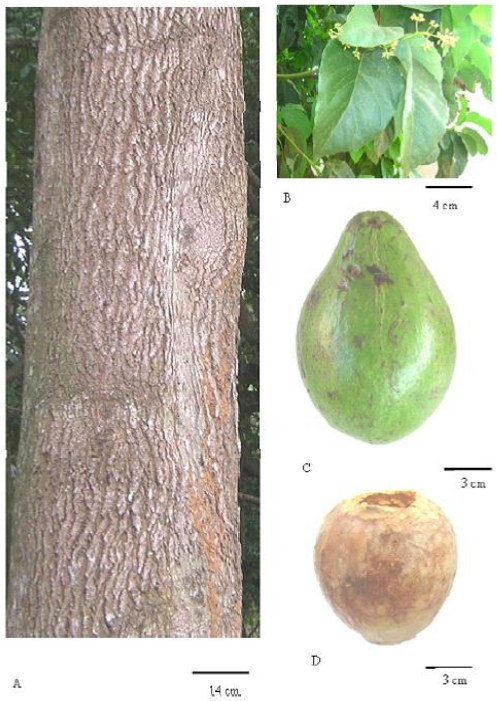

Plate 1: Showing the various part of the *Peasia americana*

Sample Collection and Preparation

Fresh leaf of *Persea Americana* was collected from an uncultivated farm land in Aroje, Ogbomosho in Oyo State (Latitude 8^{o} 07’ 60.00”N, Longitude 4^{o} 14’ 60.00” E). The leaves were taken to a botanist in the Dept. of Pure and Applied biology, Ladoke Akintola University of Technology, Ogbomoso, for proper identification and classification, (Voucher No. LHO 656). The leaves was then taken to chemistry laboratory and air-dried at room temperature for three weeks. The dried leaves were pulverized using electric grinder to increase its surface area and allowing a better contact of the extracting solvents with the sample for effective extraction.

Extraction

This was achieved using the cold extraction method. 1.7 kg of the powdered sample was soaked with n-hexane (non-polar solvent) in a beaker. The solvent was decanted and filtered off after 24 hours. This was carried out 3 times to ensure complete extraction of the major constituents. Distillation of the crude extract was carried out to concentrate the crude extract and also to remove the n-hexane. The process was repeated using methanol (polar solvent) in the same manner.

Anti-microbial Analysis

Collection of micro-organisms

The micro-organisms used for the study are clinical isolates obtained from patients with wound infections at the Micro-biology laboratory of Bowen University Teaching Hospital, Ogbomoso, Oyo State. The organisms include; *E colli spp, Klebsellia spp, Pseudomona spp.* The slants of these bacteria were cultured in Microbial Laboratory of Pure and Applied Biology Department, LAUTECH Ogbomosho.

Preparation of nutrient agar

12 g of nutrient agar powder was dissolved in 200 ml of distilled water and sterilized in an autoclave at 120^0 C for 120 minutes.

Sterilization of petri-dishes

Cleaned disposable petri-dishes and test tubes were sterilized in an oven for about 15 minutes at 120^0 C.

Culturing of micro-organisms

Sterilized inoculating wire loop was used to collect micro-organism from slant culture of all micro-organism collected and were inoculated separately into fresh sterilized nutrient agar. The resulting bacteria sub-culture were incubated for 24 hours at 37^0C.

Agar dilution method

A serial dilution of the crude extract was done to get minimum inhibition concentration. 4 different concentrations of the extract (250 g/mls, 150 g/mls and 50 g/mls) and a standard antibiotics as control. The nutrient agar was poured in the dishes and each organism was inoculated into it by streaking method. The sterilized filter discs were dipped into solution of the sample prepared in the test tube and laid on the agar. The petri-dishes were placed in the oven at 37^0C for 18 hours

RESULTS AND DISCUSSION

Table 1: Percentage of the crude extract obtained from the leaves of *persea Americana*

Weight of the powdered sample (g)	Weight of the dried beaker (g)	Weight of the beaker + extract(g)	Weight of methanolic extract(g)	Weight of n-hexane extract(g)
2000	100.6	519.28	235.61	308.9

$$\% \text{ yield of extract} = \frac{\text{weight of extract}}{\text{weight of powdered sample}} \times 100$$

$$\% \text{ yield of methanolic extract} = \frac{235.6\ g}{2000\ g} \times 100 = 11.78\%$$

% yield of methanolic extract =11.78%

$$\% \text{ yield of n-hexane extract} = \frac{308.9\ g}{2000\ g} \times 100 = 15.45\ \%$$

% yield of n-hexane extract = 15.45 %

The result here indicate that the methanolic extract of *Persia americana* leave have the highest yield with 15.45% than the n-hexane extract with 11.78%. with these result, the methanolic extract of the *Persia americana* leave have the potential of containing the more bioactive chemical components which can subjected to more analysis.

Table 2: Anti- bacterial activity of methanolic leaf extract

Concentration	***Escherichia coli*** **Zones of inhibition (mm)**	***Pseudomonas spp*** **Zones of inhibition (mm)**	***Klebsiellia spp*** **Zones of inhibition (mm)**
0.05	NZI	NZI	4.0
0.10	NZI	NZI	2.0
0.15	NZI	NZI	4.0
0.20	NZI	NZI	N.Z.I

NZI= No zone of inhibition; ZI= Zone of inhibition; Above 14= Active Below 14= Non-active; Control Plate= No growth at all.

In these present study, three different bacteria which was isolated from an infectious wound patients namely; *Escherichia coli spp, Psuedomonas spp and Klebsiellia spp.* Was tested against the methanolic extract of *Persia americana* leaf, *Klebsiellia spp* only produce inhibitory activity against the extract with minimum inhibitory concentration (MIC) ranging from 0.05-0.15mg/ml. The methanolic extract showed no zone of inhibition with *E. coli* and *Psuedomonas spp* indicating that it does not have any inhibition activity against such bacteria with minimum inhibitory concentration (MIC) ranging from 0.05-0.15mg/ml.

CONCLUSION

This study has the potential value to develop antibacterial agent against resistant and susceptible bacteria supporting the significant use of plant extracts in treating wounds related infection to bacteria and this extract will provide useful information for discovering new compounds with better activity and susceptible bacteria responsible for wound infections than current available antibiotics. From the above result, it indicates that *Persia americana* leaf will be a remedy for the treatment of infectious wounds mainly caused by *klebsellia spp.*

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