

Phytochemical Screening and In Vitro Antibacterial Activity of Doum Palm (*Hyphanaene Thebaica*) Nut: A Comparative Analysis between Fresh and A Season Year Old

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Abstract

The result of phytochemical analysis shows the presence of major constituents (alkaloids, flavonoids, steroids and polyphenols) in both the fresh and year old powdery samples. The antimicrobial analysis which is done with the crude extract of both fresh and seasonal sample shows a significant inhibition zone on the ethanolic extract of both class of samples with increase in concentration. 16.00mm, 4.00mm and 2.30mm zone of inhibition was observed for 200mg/ml, 100mg/ml and 50mg/ml of fresh sample ethanolic extracts on gram positive bacteria (*Staphylococcus Aureus*) respectively and for gram negative bacteria (*Esterichia Coli*) it shows 12.60mm, 5.00mm and 2.40mm Zone of inhibition for 200mg/ml, 100mg/ml and 50mg/ml of sample concentrate respectively. The result for the year old sample also shows 17.10mm, 5.70mm and 2.10mm zone of inhibition for 200mg/ml, 100mg/ml and 25mg/ml ethanolic extracts on gram positive bacteria respectively while for gram negative bacteria it shows 13.60mm, 4.50mm and 1.20mm for 200mg/ml, 100mg/ml and 50mg/ml of the sample respectively. The negative control of distilled water tends to show 0.00 mm zone of inhibition on every type of the microbes.

Keywords: Phytochemicals, Bacteria, Doum Palm, Medicinal, Ethanol.

Introduction

Medicinal plants play vital roles in the control and prevention of various diseases such as cancers, diabetes, and cardiovascular diseases (Mohanta et al. 2003). Some medicinal plants have been used for production of various drugs and as principal raw materials for production of other conventional medicines due to their antimicrobial activities (Srivastava et al. 2005). Antimicrobial activity involves complex mechanisms such as prevention of cell membrane, cell wall formation and inhibition of nucleic acid and protein metabolism. *Hyphaene thebaica* is an oval fruit that belongs to the mint family (Arecaceae). It is a desert palm fruit native to Nigeria, Senegal, Egypt, Tanzania, Kenya, Arabian Peninsula and West India (Orwa et al. 2009)

The root is used in the treatment of bilharzia, while the fruit pulp is used in controlling hyper-lipidaemia and hypertension (Coopoosamy and Magwa 2007). Aboshora et al. (2014)

reported on the antioxidant, anticancer, antimicrobial, and anti-inflammatory activities of *Hyphaene thebaica* fruit due to the high amounts of phenol, and flavonoids.

Among the locals in Hausa land, the nut inside the seed of a Season year old is used to treat a very popular infection called HANGUN in Hausa, which is basically the inflammation of the ear side, which lead to a swollen face side and is associated with pain. This traditional practice was the basis behind the comparative analysis of this research as it aligns with many scientific reports showing how the chemical components of a plant might change over time, this change could be in a positive enhancement way or vice versa.

Statement of Research Problem

Conventional drugs are highly reliable and very applicable in our day to day activities, but they are often accompanied with some adverse effects and are comparatively expensive, that is why the need to explore more medicinal options on natural products is vital, this made the traditional herbal medicine to gain more attractive attention due to easy accessibility, affordability, safety, promising efficacy, and being environmentally friendly (Shahrajabian et al. 2019). At the same time a lot of researches shows Many herbs and plants included in several traditional systems have promising bioactive compounds for modern drug therapy (Shahrajabian et al. 2020),

Access to proper medical care is getting challenged every day, due to poverty, lack of awareness, traditional beliefs and many more. The use of Natural products as medicinal remedies has been an ancient practice but has gain more audience due to the latter reasons, even at the moment, billions of people around the world are taking traditional herbal medicine daily in form of food, drugs or supplements (Aydin et al. 2016). In other hand the lack of modern chemical quantitative/qualitative analyzing strategy, Neglect of government regulations, poor extraction methods by unqualified medical practitioners, could lead to numerous unwanted results like outbreak or even death.

The need to bridge the gap between the two form of medical practices is highly needed, by merging the traditional practitioners knowledge and modern scientific methodologies and discoveries, That way a very reliable, cheaper, less toxic, environmental/societal friendly drug could be developed. This could be achieved by sustainable management towards traditional herbal medicine, the reactions, and challenges in the monitoring and safety of plant resources are essential sources of new drugs development which are used in treating several diseases ranging from general body pain to complicated diseases in humans (Kutalek and Prinz 2005; Maroyi and Cheikhoussef 2015)

Herbal medicine is playing in National Economy and educational system; Due to the importance of traditional chinese herbal medicine in Chinese culture, Beijing University of Chinese Medicine and the Chinese Academy of Chinese Medical Sciences were the highest ranked in Organizations-enhanced traditional herbal medicine researches, as the top ten affiliations based on traditional herbal medicines were mainly Chinese domiciled, while the other developing countries are still lagging in conventional herbal medicine research

productivity, although most developing countries depend on conventional herbal medicine to treat many diseases (Sen and Chakraborty 2017).

Aims and Objectives

The aim of this research is to analyze the presence of some phytochemicals and determine the antibacterial effect of different extracts of the sample (Doum palm nut) in comparison between a fresh and season year old.

The objectives of this research are;

- To analyze some phytochemicals present in both category of grounded sample.
- To determine the anti-bacterial effect of the extracts.
- To understand the role of solvent (solubility of the potential active ingredient) in relation to the anti-microbial effect.

Materials and Method

Sample Collection: The fresh and year old Doum palm nut was collected at Rimi market, Kano, Nigeria (11°59'28.9"N 8°31'46.2"E), coordinates; 11.991352, 8.5295053 and identified by an Herbarium in the department of Biology, Sa'adatu Rimi University of Education, Kumbotso.

Sample Preparation: The Doum palm nuts was dried in shade for 3 weeks before broken and grounded mechanically into powdery form using a mechanical grinder.

Qualitative Phytochemical Screening: The phytochemical screening was done on the grounded powdery sample for both fresh and Season year old via standard method. Analysis was done to detect the present of the following phytochemicals base on their effect in different therapeutic activity. Terpenoids, Alkaloids, Flavonoids, Steroids, Saponins and Polyphenols.

Extraction: extraction was done using three solvents; Distilled water, ethanol and n-hexane via maceration. 10g of the powdery sample was soaked in 100ml of 90% ethanol, 100 ml Distilled water, and 100ml n-hexane at room temperature 20°C. The mixture was shaken every day for at least one hour for one week before filtration using Whatman filter paper (6 mm) and concentrated using water bath at 100°C. The concentrate was stored for further analysis.

Antibacterial Assay: Disk diffusion method was used in testing the inhibitory effect of the crude extract on one gram positive bacteria (*Staphylococcus aureus*) and gram negative bacteria (*Escherichia. Coli*) according to Paper disk diffusion method described by Choudhury *et al.* Suspension or dilution of bacteria were prepared by dipping a loop of bacterium in 10 ml distilled water taken in sterilized labeled test tube. One milliliter dilution was transferred from test tube to the respective sterilized Petri plates by using sterilized

pipette. The Petri plates were gently rotated to mix the dilution and medium and allowed to solidify at room temperature.

Antibacterial test: Different concentration of the extract was prepared (200, 100 and 50 mg/ml) and Chloremphenical (Antibiotic) 50mg/ml was used as the positive control while distilled water was used as negative control. The concentrate was loaded onto sterilized paper disks (Whatman filter paper, 6 mm). The media were incubated at 37° (for bacteria) for 24h for 48h and the test was carried out. Evaluation of antimicrobial activity was done by observing the clear zone around disks and measuring the diameter of the zone of inhibition around disks in mm by sliding digital vernier caliper.

Results

Result of Phytochemical analysis for fresh grounded sample

Phytochemicals	Result
Flavanoids	+
Alkaloids	+
Steroids	+
Terpenoids	-
Saponins	-
Polyphenols	+

Table 1.1

Result of phytochemical analysis for season-year old grounded sample

Phytochemicals	Result
Flavanoids	+
Alkaloids	+
Steroids	+
Terpenoids	-
Saponins	-
Polyphenols	+

Table 1.2

Result of Anti-Bacterial Activity (Zone of inhibition)

Result of Fresh sample zone of inhibition in (mm)

Concentration /Microbial sample	200 mg/ml Ethanolic extract	200 mg/ml Water extract	200mg /ml n-hexane extract	100mg /ml Ethanolic extract	100mg /ml Water extract	100mg/ml n-hexane extract	50mg /ml Ethanolic extract	50mg/ml Water extract	50mg/ml n-hexane extract	Chloram phenicol 50mg/ml	Distilled water
Gram positive bacteria (S. Eurus)	16.00	0.00	0.00	04.00	0.00	0.00	2.30	0.00	0.00	30.00	0.00
Gram negative bacteria (E. Coli)	12.60	0.00	0.00	05.00	0.00	0.00	2.40	0.00	0.00	34.00	0.00

Table 1.3

Result of Fresh sample zone of inhibition in (mm)

Concentration/ Microbial sample	200 mg/ml Ethanolic extract	200 mg/ml Water extract	200mg /ml n-hexane extract	100 mg/ml Ethanolic extract	100 mg/ml Water extract	100mg /ml n-hexane extract	50mg /ml Ethanolic extract	50mg /ml Water extract	50mg /ml n-hexane extract	Chloram phenicol 50mg/ml	Distilled water
Gram positive bacteria (S. Eurus)	17.10	0.00	0.00	5.70	0.00	0.00	2.10	0.00	0.00	30.00	0.00
Gram negative bacteria (E. Coli)	13.60	0.00	0.00	4.50	0.00	0.00	1.20	0.00	0.00	34.00	0.00

Table 1.4

Discussion

The result of phytochemical analysis shows the presence of major constituents (alkaloids, flavonoids, steroids and polyphenols) in both the fresh and year old powdery samples, this signals a very promising activity as the phytochemicals in recognition has a reputable range of therapeutic function in the human body which is in-line with numerous findings by many scholars. The antimicrobial analysis which is done with the crude extract of both fresh and seasonal sample shows a significant inhibition zone on the ethanolic extract of both class of samples with increase in concentration. 16.00mm, 4.00mm and 2.30mm zone of inhibition was observed for 200mg/ml, 100mg/ml and 50mg/ml of fresh sample ethanolic extracts on gram positive bacteria (*Staphylococcus Eurus*) respectively and for gram negative bacteria (*Esterichia Coli*) it shows 12.60mm, 5.00mm and 2.40mm Zone of inhibition for 200mg/ml, 100mg/ml and 50mg/ml of sample concentrate respectively. The result for the year old sample also shows 17.10mm, 5.70mm and 2.10mm zone of inhibition for 200mg/ml, 100mg/ml and 25mg/ml ethanolic extracts on gram positive bacteria respectively while for gram negative bacteria it shows 13.60mm, 4.50mm and 1.20mm for 200mg/ml, 100mg/ml and 50mg/ml of the sample respectively.

Conclusion

The result of this study indicates the presence of some key phytochemicals in the grounded sample which are responsible for the anti-microbial effect on the bacteria. It also signifies a very promising nature on the effectiveness of the extracts with zone of inhibition of ethanol extracts. While the water and n-hexane extracts show a blank zero inhibition, indicating that the active constituents do not dissolve in the water and n-hexane.

Recommendation

Further researches can be made in light of the current study, which include;

- wider phytochemical screening
- Exploration of more solvents
- Broader range of microbes, for antimicrobial activity
- Use of separation techniques (advance chromatography) in isolation of numerous compounds.
- Identification of pure compounds using spectrometric analysis (NMR, FTIR, etc.)
- Drug design and structure activation.
- In-vivo analysis
- And possible clinical trial of resultant drug

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