PHYTOCHEMICAL ANALYSIS OF JACKAL BERRY (Diospurus Mesilliformis)

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A

PROJECT

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DECLARATION

I hereby declare that this project is written by me and it has not been presented before in any application for Bachelor Degree except for quotations and summaries which have been duly acknowledged.

Farida 10-12-2019

Farida yusuf moriki Date

DEDICATION

This work is dedicated to Almighty Allah for His endless love, mercy, provision, protection, safety and direction throughout my academic pursuit. To my dad, Alh. Yusuf moriki and my mum(s) Haj. Hassana, my Husband and also my beloved son Usman Ayman for their unalloyed support and unquantifiable investment in my life may, Almighty Allah reward you abundantly.

CERTIFICATION

This project Entitled "Phytochamical analysis of Jackal berry (*Diospyrus mesilliformis*) Stem bark and Root" meets the regulation governing the award of Bachelor of Science at the federal university Gusau and is approved for its contribution to knowledge and literary presentation.

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ABSTRACT

Ethno medical practice mostly revolves round Phytochemistry. Phytochemical screening of medical plants would therefore help discover a wide range of more efficacious drugs safer and cheaper drugs. The various plant parts of *Diospyros mespiliformis* were collected from Federal University Gusau. They were oven dried, pulvurised and serially extracted by cold extraction procedure starting with methanol. The methanol extract was then serially extracted with aqeous. The result revealed the presence of cardiac glycosides, saponin glycosides alkaloid, glycosides etc. in various extracts of the various plant parts. The work 'phytochemical analysis' of the root extract of the root, steam and bark extracts of *Diospyros mespiliformis* has opened up a fresh line of research into discoveries of new drugs in many classes. It will be more fortunate if the new drugs thereafter discovered have better efficacy and less toxicity than the already existing ones in the classes.

CHAPTER ONE

1.0 INTRODUCTION

Phytochemicals are chemical compounds that occur naturally in plants e.g. alkaloids, tannins, flavonoids, cardiac glycosides, Anthraquinones etc). In other words, they are natural chemicals that are produced by plants. These compounds are required by plants for defense against predator's and disease pathogen. These compounds are mostly secondary metabolites such as alkaloids, steroids, tannin and phenol compounds which are synthesized and deposited in specific part or in all part of the plant (Joseph *et al.*, 2010). Generally leaves are the favourable storage site for desired compounds. (Yussof et *al.*, 2012).

Plant secondary metabolites are used as the basic for the production of valuable synthetic compounds such as pharmaceutical cosmetic, or more recently nutraceutical (Bourguad *et al.*, 2001). These secondary metabolites are largely viewed as potential sources of new drugs, antibiotic, insecticides, and herbicides. Tannins and flavonoids are thought to be responsible for antidiarrhoeal activity by increasing colonic water and electrolyte re absorption (Palombo, 2006). Alkaloids are the best known nitrogen containing metabolic of plant and are sparsely distributed in the plant kingdom, but much more specific to defined plant genera and species. This is probably due to limited supply of nitrogen in plant (Herbone, 1999).

Jackal berry *(Diospyrus mesiiliformis)*, (also known as African ebony is a large dioecious evergreen tree belong to family Enabaceae found mostly in the savannas of Africa. Mature trees have dark gray fissured bark. An adult tree reaches an average of 4 to 6 metres in height, though occasionally trees reach 25 metres. The foliage is dense and dark green with elliptical leaves, which are often eaten by grazing animals such as elephants and buffalo. The tree flowers in the

ainy season; the flowers are imperfect, with genders on separate trees, and are cream-colored. he female tree bears fruit in the dry season and these are eaten by many wild animals; they are val-shaped, yellow or purple when ripe and about 20–30 mm in diameter. The fruit remain mbedded in the persistent calyxlobes., the fruit has potential to improve nutrition, boost food ecurity, foster rural development and support sustainable landcare. The fruit is edible for umans; its flavor has been described as lemon-like, with a chalky consistency. They are ometimes preserved , can be dried and ground into a flour, and are often used for brewing beer nd brandy . (Shaanika, 2012).

he leaves, bark and roots of the tree contain tannin , which can be used as a styptic to staunch leeding. The roots are consumed to purge parasites and are thought to be a remedy for leprosy. annin is contained in the leaves, bark and roots, and acts as an astringent that helps stop leeding. The tree is also supposed to have antibiotic substances that help heal wounds. A ixture made from the roots is used to get rid of parasites like ring worm, and dysentery and ever. It is also considered a remedy for leprosy. Seeds are eaten as nuts. Fruits are often used to ew beer or fermented for wine. Fruit and plant parts contain tannin. The Jackalberry tree is und throughout Africa, from Senegal and the Sudan to Namibia and the northern Transvaal. It most commonly found on savannas or savanna woodlands where it can be found growing on rmite mounds. In heavy soils the termite mounds provide the tree with aerated soil, and a ource of moisture, and the roots provide protection for the termites. As they live on dead rganic matter the termites don't eat the living wood. Jackalberry wood is almost termite-esistant after it has been cut down (Ombike, 2012).

1.1 Statement of the problem

Before the advent of drugs, barks of trees, leaves, roots, of plant have been used to cure ailments. Through several studies that have been carried out on this plant but there is need for determination of phytochemical compound present in Jackal berry roots and stem bark. Because a large population of people are using this plant to cure various ailments.

1.2 Justification

Naturally, Jackal berry plant is rich in nutrients and phytochemical which improved good health and preventing diseases. This plant is abundantly and little research was conducted on its phytochemical. These necessitate the others of the topic, to enlight the community the potential of plant as food and medicine.

1.3 Aim and objective

To determine the phytochemical contents of Roots, Stem bark, Bark of Jackal berry

1.3.1 The objective

1. To compare the phytochemical contents present in roots and stem bark of Jackal berry

CHAPTER TWO

2.0 LITRETURE REVIEW

African ebony (Diospyros mespiliformis) has a good mutualism and symbiotic network with many living organisms, from human beings to small insects. There is a complex ecological system revolving around this tree. It is one of the savanna giants that can live for more than 200 years. It is a tall, upright tree that can reach a height of 25 m, with a trunk circumference of more than 5 m. It has a dense evergreen canopy (Belemtougri *et al.*, 2006). The bark is black to grey, with a rough texture. The fresh inner skin of the bark is reddish. Leaves are simple, alternate, leathery and dark green. The margin is smooth and new leaves in spring are red, especially in young plants. Flowers are cream-coloured and bell shaped (Belemtougri *et al.*, 2006). Among Hausa Fulani people of Jigawa in Nigeria, *Diospyros mespiliformis* happens to be one of such plant used in the treatment of tumor-related disease. In local Nigerian language (Hausa) the plant is known as Kanya. This plant is not used only to treat tumor related disease but also to treat diseases such as fever, malaria, pneumonia, syphilis, leprosy, dermatomycoses (Mohamed et al., 2009). It is also used in the treatment of diarrhea, whooping cough, wounds.(Adzu et al., 2002). These studies were designed to test the inhibitory effects of methanol crude extract against tumor related cells. However a model was designed to use by (Ayinde and Agbakwuru 2010), where cancer cell lines are not available in order to mimic the situation.Phytochemical screening which revealed the presence of carbohydrate, glycoside, anthraquinone, steroid, triterpenes, saponin, tannins, flavonoids and alkaloid. Thin layer chromatographic profiles of crude extract and ant proliferative studies were carried out in this research.

The ethnopharmacological uses of different organs of *D. mespiliformis* have been reported in the literature e.g leaf decoctions are used against fever, whooping cough and wounds (Adzu *et al*

2002). Barks and roots are used to treat malaria, pneumonia, syphilis, leprosy, dermatomycoses, diarrhea, facilitation of delivery and as psycho-pharmacological drug (Mohamed *et al.*, 2009). Studies have shown that the leaf extracts of *D. mespiliformis* are effective against *Plasmodium falciparum* invitro (Etkin, 1997), *Plasmodium berghei* in vivo (Adzu and Salawu, 2009) and relieved pains and fever in rodent models (Adzu *et al.*, 2002). Tannins, alkaloids, diosquinone and plumbagin were isolated from *D. mespiliformis* and demonstrated activity against *Staphylococcus aureus, Escherichia coli* and *Pseudomonas aeruginosa* thus providing a basis for the plant usage in herbal medicine (Adeniyi *et al.*, 2006; Lajubutu *et al.*, 2006). The global acceptability, application and efficacy of herbal medicaments have proportionately exposed millions of people to the underlying but seldom reported problems of medicinal plant toxicity and misadventuring. Most herbal treatments are in the form of crude extracts administered over a few days or weeks with the likelihood of cumulative intoxication of recipients (Jigam *et al.*, 2011, Klink, 1997). The therapeutic window of digoxin is so narrow that maintenance doses could be as small as 50ug/day whereas digitalis leaf often preferred to the pure compounds (digoxin and digitoxin) is often given in doses of 1.5g/day (Gamaniel, 2000). The present study was therefore undertaken to evaluate some biochemical indices in mice sub-chronically administered *D. mespiliformis* root extracts in an attempt to further rationalize its use in herbal medicine.

2.3 [illegible]

[illegible] Coast, Upper Volta to be haemostatic and cicatrisant [illegible] wounds and to prevent infection; the sap is [illegible] is taken as a febrifuge and stimulant. A decoction of leafy twigs is [illegible] in draught as a poison antidote. In Senegal the leaves are prescribed for serious illnesses such as pneumonia, infectious fevers, syphilis, leprosy and yaws. Such reliance is placed on this drug-plant that it is usually prescribed alone. Leaves and fruit are used internally for [illegible] and dysenteriform diarrhoea, and externally for headaches, arthritis and dermal troubles. The use of the leaves, with the bark, husks of tamarind and guinea-corn gruel, as a leprosy remedy is recorded in N Nigeria. Leaves in cold infusions are commonly

taken in the soudanian region for dysentery, and a leaf- decoction by draught or as a wash for fever (NRC, 2008).

2.4 BOTANICAL DESCRIPTION JACKAL BERRY

Diospyros mespiliformis also called "African Ebony" or Kanya by the Hausa of Nigeria is a large deciduous tree confined to tropical and subtropical regions including Africa and Asia (Dalziel, 1955; Mohamed *et al.*, 2009). Mature trees average 4-6 metres occasionally reaching 5 metres in height. Foliage is dense and dark green with elliptical leaves and fruits often eaten by grazing animals (Belemtougri *et al.*, 2006;). *Diospyros* is reportedly one of the most important genera of *Ebenaceae* which species have been used over the millennia in traditional medicinal systems including Ayurveda, Chinese and African folklores (Mallavadhani *et al.*, 1998; Kantamreddi and Wright, 2007).

2.5 Extraction of phytochemical in plant

Medicinal plants are raw materials for extraction of active constituents, several formulations like herbal teas, extracts; decoctions, infusions, tinctures, are prepared from medicinal plants (Kraisintu, 1997). For developing phyto medicines as a major area of concern, it would be essential to adopt a holistic interdisciplinary approach, have a scientific basis of the understanding of the plant systems, new innovations and their conservation for utilisation in future on a sustainable basis (Sharma, 1997).

CHAPTER THREE

3.0 Materials and Methods

3.1 Study Area

This research was carried out in Biochemistry laboratory Federal University Gusau. Between August to November 2019.

3.2 Sample Collection and Preparation

Fresh roots and stem-bark of jackalberry was collected in Federal University Gusau. In the month August 2019. The fresh roots and stem-bark of jackalberry collected was washed under running tap water and drained to remove dust particles. The sample was then air dried for 6 days and then crushed into powder with the aid of pestle and mortar. The powder was used for the analysis.

3.3 Phytochemical analysis

3.3.1 Alkaloid

The presence of alkaloid was determined as described by Harborne 1973; Trease and Evans 1996. A portion of the plant powder (5g) was reacted with a few drops of hagers reagent (1.0 cm3) and another 5g portion was treated with wegners reagent (1.0 cm3) turbidity or precipitate with either of these reagents was taken as an evidence for the presence of alkaloids.

3.3.2 Tannins

Ferric chloride solution 5% ferric chloride solution was added drop by drop 2-3ml of the extract and the colored produced is noted. Condensed tannins usually give a dark green color; hydrolysable tannins give blue- black color.

3.3.3 Saponnins

Five mills (5ml) of the extract was placed in a test tube + 5ml of water and sharked strongly.the whole tube was filled fronth that lasts for several minutes.

3.3.4 Flavonoids

Three mill (3ml) aliquot of the filtrated and 1ml of 10% NaOH sodium hydroxide, if a yellow color is developed this indicates the possible presence of flavonoid compounds.

3.3.5 Glycosides

Five mill (5ml) of 50% H_2SO_4 was added to 5cm³ of the extracts in a test tube. The mixture was heated in boiling water for 15minutes. Cool and neutralize with 10% NaOH, add 5ml of fehling's solution will be added and the mixture is boiled. A brick- red precipitate is observed which indicate the presence of glycosides.

3.3.6 Cardiac Glycoside

To one of herb extract two ml of 3.5% ferric chloride solution was added and allowed to stand for one minutes. Or 1 of conc H2SO4 was carefully poured down the wall of the tube so as form a lower layer. A reddish brown ring the interface indicates the two layer presence cardiac glycoside.

3.3.7 Steroids

This was carried out according to the method of harbone 1973. 5ml of the extract will be dissolved in 2ml of chloroform 2ml of sulphuric acid was carefully added to form lower layer. A reddish brown color at the inter face indicate the presence of a steroidal ring.

3.3.8 Saponin Glycosides

To 2.5ml of the extract was added 2.5ml of fehing's solution A and B. a bluish green precipitate showed the presence of saponnin glycosides.

3.3.9 Balsams

The extract was mixed with equal volume of 90% ethanol. 2 drops of alcoholic ferric chloride solution was added to the mixture. A dark green colour indicates the presence of balsams.

3.3.10 Anthraquines

0.50gm of each plant extract was shaken with 10ml benzene, and 5ml of 10% ammonia solution is added. The mixture was shaken and the presence of a pink, red, or violet color in the ammoniacal (lower) phase indicates the presence of anthraquinones.

3.3.11 Volatile Oils

1ml of the fraction was mixed with dil. HCL. A white precipitate was formed which indicated the presence of volatile oil.

3.4 Statistical Analysis

Data obtained were statistically analyzed using descriptive statistics on SPSS statistical software plat form, and values was expressed as mean ± standard deviation of the triplicate values.

CHAPTER FOUR

4.0 Results

The phytochemical analysis of jackal berry root, steam, and steam bark shows the absent of flavonoids in the root, steam, and steam bark using aqeous and absent using methanol. It shows the presence of saponin using aqeous and methanol in both root, steam and steam bark, it shows the presence of alkaloid in root using aqeous and presence in steam and steam bark using methanol. Glycoside, saponin glycoside and cardiac glycoside are presence in both aqeous and methanol. while flavonoids, tannins saponin glycoside are absent in root using both aqeous and methanol.(Table 1)

Table 1: phytochemical identified in Bark, steam bark and Root using Aqeous

Parameters	Bark	Steam bark	Root
Flavonoids	–	–	–
Tannins	–	–	–
Saponins	+	++	+
Glycoside	+++	+++	+
Cardiac glycoside	+	+	–
Saponin glycoside	+	+	–
Alkaloids	+	+	+

Key : + trace amount, ++ moderately, +++ higher concentration and - absent

Table 2: phytochemical identified in Bark, steam bark and Root using Methanol

Parameters	Bark	Steam bark	Root
Flavonoids	–	–	–
Tannins	–	–	–
Saponins	+	++	+
Glycoside	++	+++	++
Cardjac glycoside	++	+++	++
Saponin glycoside	++	++	–
Alkaloids	++	++	+++

Key : + trace amount, ++ moderately, +++ higher concentration and – absent

Table 3: phytochemical identified in Bark, steam bark and Root using Aqueous and Methanol

Phytochemicals	Aqueous			Methanol		
	Bark	Steambark	Root	Bark	Steam bark	Root
Flavonoids	–	–	–	–	–	–
Tannins	–	–	–	–	–	–
Saponins	+	++	+	–	–	–
Glycoside	+++	+++	+	++	+++	++
Cardiac glycoside	+	+	–	++	+++	++
Saponin glycoside	+	+	–	++	+	–
Alkaloids	+	+	+	++	+	+++

Key: + trace amount, ++ moderately, +++ higher concentration and – absent

CHAPTER FIVE

5.0 Discussion, Conclusion and Recommendation

5.1 Discussion

The medicinal value of plants used in traditional medicine derives due to presence of phytochemical principles, which are found in certain parts of the plants. Therapeutic plants contain naturally dynamic synthetic substances, for example, Saponins, tannins, fundamental oils, Flavonoids, alkaloids and other chemical compounds that poses remedial properties. In this study, the phytochemical analysis was carried out by using water extraction method. Table 1 shows the presence of some secondary metabolites that cardiac glycoside and glycoside were found in high quantity in sample which is in line with similar findings of (Erukaine., 2011) reports. Alkaloids, and Saponin glycosides was found to be moderately present which correlate with (Ogungbenle., 2011) and finally flavonoids and tannins were absent.

5.2 Conclusion

It is concluded that this phytochemical analysis of the root extract of the root, steam and bark extracts of *D. mespiliformis* has opened up a fresh line of research into discoveries of new drugs in many classes. It will be more fortunate if the new drugs thereafter discovered have better efficacy and less toxicity than the already existing ones in the classes.

5.3 Recommendation

Further research should be carried on flowers to know more about its medicinal values and hence how it could be used in promoting modern pharmaceutical.

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