



**DEPARTMENT OF SCIENCE LABORATORY TECHNOLOGY**

**QUALITATIVE DETERMINATION OF THE BIOACTIVE  
CONSTITUENTS IN XYLOPIA AETHIOPICA**

***BY***

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HND/23/SLT/FT/0270**

**BEING PROJECT SUBMITTED TO  
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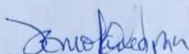
**SUPERVISED BY: MR. A. P. OKEDIRAN**

**2024/2025 SESSION**



### CERTIFICATION

This is certify that this project work presented by **RASAQ GBEMISOLA IMOLEAYOMIDE** with Matriculation Number **HND/23/SLT/FT/0270** has been read, approved and submitted to the Department of Science Laboratory Technology (Biochemistry Unit), Institute of Applied Science, Kwara State Polytechnic, Ilorin.

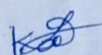


**MR.A.P. OKEDIRAN**

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23<sup>rd</sup> July, 2025

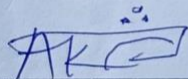
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**DATE**



**DATE**

**EXTERNAL EXAMINER**

**DATE**

## **DEDICATION**

This seminar is dedicated to God Almighty, the giver of wisdom, knowledge and understanding. The beginning and the end, who has made this work successful. This work is also dedicated to my beloved parents MR. and MRS. RASAQ.

## **ACKNOWLEDGEMENT**

My gratitude goes to Almighty God, the doer of all good things, most gracious, most merciful, the Lord of the universe, the giver of life. I thank God for giving me the strength and ideas for writing this seminar work.

I express my profound gratitude to my experienced, knowledgeable seminar supervisor MR. OKEDIRAN. A.P for his direction and dedicating his time to go through my seminar work. May Almighty God continue to strengthen you in more wisdom and knowledge (Amen) and I say a big thank to the H.O.U MRS. SALAUDEEN K.A and to the H.O.D DR. USMAN ABDULKAREEM. My amiable lecturer Mr. Adeyemo O.E. Thank you for your support, may your days be filled with goodness(Amen). My appreciation goes to all the lecturers in the Department of Science Laboratory Technology, may God shower his blessings upon you all(Amen).

My appreciation and special thanks goes to my beloved, irreplaceable and caring parents MR. and MRS. RASAQ for financial support, endless prayers, words of encouragement, I pray that you will live long, in good health and wealth to eat the fruit of your labour(Amen).

My profound gratitude goes to my loving and supportive friends, ODENIYI DEBORAH, MATTHEW RAPHAEL and to my most loving friend FARINLOYE AKINYEMI, thank you all for your support and encouragement, I pray that the God Almighty grant your heart desires(Amen). I LOVE YOU ALL.

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##### 3.1 RESULTS

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## **ABSTRACT**

11.8g of powdered sample of *xylopia aethiopica* was extracted in 300ml of methanol using sohxlet method to yield 3.63g of crude extract (30%). The crude extract was subsequently subjected to phytochemical analysis using standard methods. The presence of alkaloids, sterols, tannins, triterpenes, and glycosides were confirmed while lactones, diterpenes, saponins and flavonoids were absent.

## **CHAPTER ONE: INTRODUCTION AND LITERATURE**

### **REVIEW**

#### **1.1 INTRODUCTION**

*Xylopia aethiopica* or Ethiopian pepper as it is usually called, is an angiosperm belonging to the family Annonaceae and is among the species that thrive in the evergreen rain forests of tropical and subtropical Africa. *Xylopia aethiopica* matures as a slim, tall tree of approximately 60 cm in diameter and up to 30 m high with a straight stem having a slightly stripped or smooth bark. It bears odoriferous fruits, which are slender pods slightly curved with about 15 carpels and are arranged in capitula to form bouquets of 12-20 bacciferous-like capsules. *Xylopia* is a compression from the Greek words “xylon pikron” which mean "bitter wood". The second part of the plant's binomial name, *aethiopica*, refers to its origin. This plant has played a key role in African traditional medicine for several centuries owing to its

wide array therapeutic indications. *Xylopia aethiopica* is used in the treatment of cough, biliousness, bronchitis, rheumatism, dysentery, malaria, uterine fibroid, amenorrhea, boils, sores, wounds and cuts among others (Busia K, 2007).

Typically, studies on *Xylopia aethiopica*, like most medicinal plants have focused on the biological activities of its chemical constituents, ethnobotany, pharmacology, and taxonomy. However, a comprehensive or systematic review on the plant is generally lacking. That notwithstanding, it is worth mentioning that some attempts have been made in this regard. Clearly, this study focuses mainly on *Xylopia aethiopica* fruits from Ghana and as such is barely comprehensive. Again, the study was conducted almost a decade ago and throughout this period, several studies have been carried out on the plant in various fields of science. Consequently, this paper presents information on but not limited to the morphology, ecology,

ethnopharmacology, phytochemistry, biological activities and toxicological properties of *Xylopia aethiopica* and aims at providing an up-to-date detail that should constitute baseline information for future research and commercial exploitation of the plant (Ruddfo Juliani H, 2008).

## **1.2 TAXONOMY**

The genus *Xylopia* is consists of about 150 species which occur in tropical and subtropical Africa. *Xylopia aethiopica*, also known as Negro pepper, is an angiosperm belonging to the custard apple family, Annonaceae (Choumessi AT, 2012).

Kingdom: Plantae

Subkingdom: Viridiplantae

Infrakingdom: Streptophyta (land plants)

Superdivision: Embryophyta

Division: Tracheophyta (vascular plants)

Subdivision: Spermatophytina (seed plants)

Class: Magnoliopsida

Superorder: Magnolianaes

Order: Magnoliales

Family: Annonaceae (custard apples)

Genus: *Xylopia*

Species: *Xylopia aethiopica*

### **1.3 CULTIVATION**

*Xylopia aethiopica* grows into a giant tree in most of the evergreen rain forests of tropical and subtropical Africa. The plant thrives in humid forest zones of West Africa. It can grow to as high as 20 m or even up to 45 m. The stem is generally



straight with a smooth or slightly striped bark and has a diameter of about 60cm. The leaves are elliptically shaped and can be longer than 15 cm at the offshoots with thick fringes and feel fatty to touch. They appear blue-green on the upper side, with a big clear green median nerve, while the secondary veins are much less pronounced. The trees also bear odoriferous greenish-white flowers with external petals up to 5 cm long. *Xylopia aethiopica* is cultivated mainly for the fruits, made up of clusters of about 30 mericarps, 5 to 6 cm long and 0.5 to 0.8 cm wide. For each mericarp there are 1 to 9 seeds, which are 5 to 6 mm long and 2 to 3 mm wide, covered with a bright brown tegument (Lamaty G, 1987). In West Africa, the tree flowers twice every year (i.e. March-July and October-December) while fruiting occurs in December-March and June-September with harvesting time running from February to May and from August to October (Orwa C, 2010).

## 1.4 ETHNOBOTANICAL

*Xylopia aethiopica* possesses great nutritional and medicinal values in traditional medicine. Almost all parts of *Xylopia aethiopica* are very useful medicinally, but the fruits are most commonly used for therapeutic purposes. Extracts of the fruits are used in the treatment of cough, biliousness, bronchitis, rheumatism, dysentery, malaria, uterine fibroid and amenorrhea (Busai K, 2007). The fruits can also be crushed and mixed with Shea butter and used as body creams, cosmetic products or perfumes. It has also been showed that the essential oil from the seeds of *Xylopia aethiopica* can be used in the formulation of shampoos due to its high saponification value ( $207.2 \pm 8.0$ ). Conversely, in a preliminary evaluation of the physical and chemical properties of essential oils from the seeds *Xylopia aethiopica*, Ogbonna and others also determined the saponification value of the oil to be 130.18.

This value is lower than that obtain in the instance described earlier. The geographical location of the plant and the time of harvest could all affect the nature and composition of essential oils in the plant thus resulting in the differences observed. In Benin, the dried fruits are commonly used as a constituent of extracts for bathing, and as a potion administered to new-borns. The seeds are crushed and applied topically on the forehead in the treatment of headache and neuralgia. It can also be taken as a decoction, concoction or even chewed and swallowed for the management of various aches and pains. It has also been shown experimentally, that the seeds possess good anthelmintic activity against *Nippostrongylus brasiliensis* and as such its use in man as an anthelmintic may be investigated. Various extracts of *Xylopia aethiopica* have also demonstrated some promise in its employment as an adjunct therapy in the management of sickle cell disease. An oily extract of the seeds is used as a

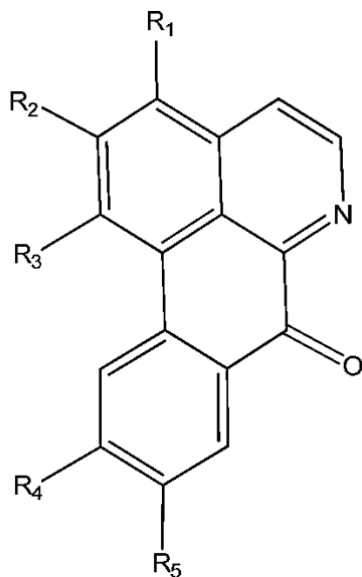
lotion for boils and eruptions, and as a liniment for lumbago. Traditional medical practitioners and birth attendants use a decoction of the seeds to induce placental discharge postpartum due to its abortifacient effect (Woode E, 2012).

## **1.5 CHEMICAL COMPOSITION OF THE LEAF, BARK, FRUIT, SEED AND ESSENTIAL OIL**

*Xylopia aethiopica* is known to have myriad chemical constituents with diverse therapeutic and pharmacological properties. These compounds, most of which have been isolated and characterized include saponins, sterols, carbohydrates, glycosides, mucilage, acidic compounds, tannins, balsams, cardiac glycosides, volatile aromatic oils, phenols, alkaloids, rutin and fixed oils. The plant also contains vitamins A, B, C, D, and E, and proteins together with high amounts of minerals like copper, manganese and zinc (Orwa C, 2010).

### 1.5.1 ALKALOIDS

Alkaloids isolated from the methanolic extracts of the aerial parts of *Xylopi aethiopica* include the oxoaporphine alkaloids, oxophoebine and liriodenine. Also, the alkaloids oxoglaucine, O-methylmoschatoline and lysicamine have been isolated from ethyl acetate extracts of the plant. Most of these alkaloids have demonstrated some cytotoxic effects in various studies. The chemical structures of these alkaloids are as shown below.

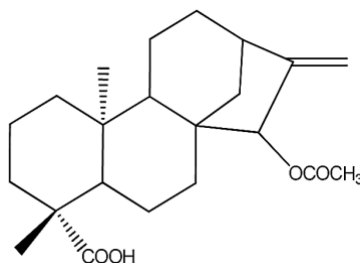


**Fig. 1: General structure of *Xylopia aethiopica* alkaloids**

### **1.5.2 DITERPENES**

Most of the acidic compounds isolated from *Xylopia aethiopica* are the various kaurane, kolavane and trachylobane diterpenes which are reportedly present in the stem bark and fruit of the plant. Quite an extensive research has been conducted on most of these diterpenes leading to the elucidation of their structures. Typically, xylopic acid (Fig. 2), a kaurene

diterpene has been taken through extensive research to the extent that several derivatives of it have been synthesized.



**Fig. 2. Chemical structure of xylopic acid**

Ekong and Ogan (Ekog D, 1968) isolated xylopic acid from the dried powdered fruits of *Xylopia aethiopica* by extracting the latter with light petroleum (b.p.60-80°C). The extract was subsequently concentrated and crystallised from ethyl acetate to obtain xylopic acid, its melting point was determined to be 259-260°C. Again, Soh et al. (Soh D, 2013) also extracted xylopic acid using hexane where the extract was chromatographed over silica gel using hexane-ethyl acetate (95:5) mixtures to obtain xylopic acid as a white powder with

melting point of 230-232°C. Although the two groups of researchers reported to have isolated xylopic acid, there was a vast difference in the melting point of the crystals obtained. This could possibly be attributed to the fact that a particular group isolated xylopic acid of low purity and as such the impurities might have altered the actual melting point of the compound. Better still, it could be that one group isolated a compound closely related to xylopic acid but not xylopic acid itself. Apart from the difference in melting point, Ekong and Ogan elucidated the structure of the compound isolated as 15 $\beta$ -acetoxy-(-)-kaur-16-en-19-oic acid while Soh et al determined theirs to be 15 $\alpha$ -acetoxy-ent-kaur-16-en-19-oic acid. Fiagbe et al. also solved the structure of xylopic acid as 15 $\beta$ -acetoxy-(-)-kaur-16-en-19-oic acid by use of crystallography. Elsewhere, Adosraku and Oppong Kyekyeku isolated xylopic acid from the dried fruits of *Xylopia aethiopica* using petroleum ether (40-60°C) and recrystallized the former with



distilled alcohol. The melting point obtained for these crystals was 261-262°C. In another research, Fahim et al. determined the melting point of isolated xylopic acid as 265 – 266°C. The melting points obtained in these two separate studies for xylopic acid are in much agreement with that obtained by Ekong and Ogan (259-261°C) as compared to the melting point obtained by Soh et al. (230-232°C). A major issue that requires further research is whether only one of these stereochemical forms of xylopic acid actually exists, or whether both do exist concurrently in the same plant or separately depending on the geographical location of the plant. And finally, the effects of the stereochemical difference if any on the melting point of the compound should be investigated (Lamaty G, 1987).

### 1.5.3 ESSENTIAL OILS

Among the commonest groups of chemical compounds conspicuously present in the various parts of *Xylopia aethiopica* are the essential oils. Different studies conducted on these essential oils have shown the presence of a wide diversity of chemical compounds. In one of the early studies conducted, Ogan (Ogan A, 1971) identified for the first time, an aromatic aldehyde specifically, cuminal (*p*-isopropyl-benzaldehyde) as a component of the essential oils obtained from the fruits of *Xylopia aethiopica*. The essential oil from the dried fruits of *Xylopia aethiopica* and the only aldehyde identified was cuminic aldehyde at a concentration of 6.5%, corroborating Ogan's work. Other compounds were also identified; namely  $\beta$ -pinene a monoterpene hydrocarbon, bisabolene a sesquiterpene hydrocarbon, terpinene-4-ol which is an alcohol and the oxide 1, 8-cineole among others.

In a study to characterize the key aroma compounds in dried fruits of *Xylopia aethiopica* using aroma extract dilution analysis, Tairu et al. observed that  $\beta$ -pinene, myrtenol, and  $\beta$ -phellandrene were important contributors to the overall odour of the *Xylopia aethiopica* fruit. It was also revealed that linalol,  $\alpha$ -farnesene, (E)- $\beta$ -ocimene, and  $\alpha$ -pinene are responsible for the flowery and terpeny odour notes detected in the corresponding odour-active regions. Most of these chemical constituents discussed potentially possess one biological activity or the other and thus it is imperative that researchers critically investigate these compounds for any relevant pharmacological activity.

Furthermore, it is observed from this study that, there are many more compounds present in various parts of *Xylopia aethiopica* that are yet to be isolated and characterized. Therefore further research should be carried out in this regard

to identify more compounds and also investigated their biological activity. In cases where some biological activity is observed, the possibility of chemically modifying the lead molecule to obtain a more active compound should also be considered (Busai K, 2007).

## 1.6 PHARMACOLOGICAL PROPERTIES

An ethanolic extract of the fruits of *Xylopia aethiopica* showed significant analgesic activity against acetic acid-induced visceral nociception, formalin-induced paw pain (both neurogenic and inflammatory), thermal pain as well as carrageenan-induced mechanical and thermal hyperalgesia in rats and mice when the extract was administered orally. Xylopic acid isolated from the dried fruits of *Xylopia aethiopica* also showed comparable results (Soh D, 2013). The aqueous ethanolic fruit extract of *Xylopia aethiopica* at concentrations of 100, 300 and 600 mg/kg all exhibited good anti-arthritic effect when Adjuvant arthritis was induced in Sprague-Dawley rats by intraplantar injection of Complete Freund's Adjuvant into their right hind paw. The anti-arthritic effect was achieved as a result of the suppression of both

inflammation and the destruction of the joint in adjuvant arthritic rats.

In an investigation of the neuropharmacological effects of an ethanolic fruit extract of *Xylopi aethiopica* and xylopic acid, in vivo experiments involving mice were conducted. From the results of this study, ethanolic fruit extract of *Xylopi aethiopica* and xylopic acid both showed significant central nervous system depressant effects in pentobarbitone-induced hypnosis and spontaneous activity test. Xylopic acid significantly decreased spontaneous locomotion at 30-1000 mg/kg doses ( $F_{7,48}=6.320$   $p<0.0001$ ) while the ethanolic extract of *Xylopi aethiopica* caused a significant reduction in activity ( $F_{6,42}=6.078$   $p<0.0001$ ) at doses of 300 and 1000 mg/kg. Xylopic acid showed a significant and dose dependent reduction of the onset of sleep from doses of 100-1000 mg/kg and also sleep duration was prolonged significantly,

only at 300 and 1000 mg/kg. Again, it was reported that the ethanolic extract (which was obtained by extracting the plant material with ethanol and subsequent evaporation of the latter to obtain a dry mass) did not have a significant effect on the onset of sleep. However, it significantly ( $F_{6, 42}=133.0$   $p<0.0001$ ) prolonged sleep duration in a dose-dependent manner. Xylopic acid and the ethanolic fruit extract of *Xylopic aethiopica* both showed neuromuscular coordination impairment potential at doses above 300 mg/kg. The ethanolic fruit extract of *Xylopic aethiopica* significantly increased seizure threshold at all doses while xylopic acid had no effect on PTZ (Pentylentetrazole)-induced convulsion. The ethanolic fruit extract of *Xylopic aethiopica* exhibited an induction of hepatic enzymes at lower doses whereas xylopic acid showed a bidirectional effect by inhibiting hepatic enzymes at lower doses and inducing these enzymes at higher doses. It was also suggested that, both the ethanolic fruit extract of

Xylopia aethiopica and xylopic acid may be metabolized by hepatic enzymes.

## 1.7 ANTIOXIDANT EFFECT

The free radical scavenging effects, the antioxidant and ion toxicity preventive effect of ethanolic extracts of Xylopia aethiopica stem bark was investigated. From the results of this research, it was observed that vitamin C used as standard had a significantly ( $p < 0.05$ ) lower value of IC<sub>50</sub> on nitric oxide (NO), hydroxyl (OH), 2,2-diphenyl-1-picrylhydrazyl (DPPH) and 2,2'-azinobis(3-ethylbenzthiazoline)-6-sulfonic acid (ABTS) radicals as compared to the extracts. The extracts also showed a protective effect against lipid peroxidation. It may therefore be inferred from this study that Xylopia aethiopica has a good antioxidant and protective potential against ion-mediated oxidative damage and may be considered as a potential drug against metal mediated toxicity. Phenol-



rich fruit extracts of *Xylopia aethiopica* exhibited very good free radical scavenging ability in a concentration dependent fashion (0.08-0.53 mg/mL). Again in assessing the ferric reducing antioxidant property of the extract, the study showed that the extract possesses good reducing potentials (Adefegha S, 2012).

## **1.8. ANTIMICROBIAL ACTIVITY**

*Xylopia aethiopica* has gained wide application in traditional medicine partly because of its usefulness as an effective antimicrobial agent. Extracts and isolates from various parts of the plant through in vitro studies have confirmed its anti-microbial activity. For instance, Konning et al. (Konning GH, 2004), reported that a 3%(w/v) methanol extract of *Xylopia aethiopica* (dried fruits) showed some antimicrobial activity against gram negative (*Escherichia coli* and *Pseudomonas aeruginosa*) and gram positive

(*Staphylococcus aureus* and *Bacillus subtilis*) bacteria. It also exhibited good antifungal activity against *Candida albicans* and *Aspergillus niger*. The ethanol extract of the dried fruits of *Xylopia aethiopica* was also investigated for antimicrobial activity. In this study, the agar diffusion method was used to determine both the zones of inhibition and the minimum inhibitory concentration (MIC). The extract exhibited activity against *E. coli*, *S. typhi*, *Candida albicans*, *B. aurum* with 15mg/mL MIC. The extract however, did not show any activity against *S. aureus* and *B. subtilis*. This study has therefore shown to some extent that *Xylopia aethiopica* ethanol extract contain compounds that could be further investigated as potential source of broad spectrum antibacterial agents. Hot water extracts of *Xylopia aethiopica* dried fruits in a research conducted by Awuah exhibited antifungal activity with *Rhizopus* sp. and *Ustilago maydis* being the most susceptible microorganisms while *Ustilaginoidea*

virens was the least susceptible among the organisms tested. The essential oil of *Xylopia aethiopica* has been shown to possess antibacterial as well as antifungal activity. In a study, the disk diffusion method was employed for antimicrobial assay with the minimum and maximum zones of growth inhibition as 18mm (against *Bacillus subtilis*) and 32 mm (against *E. coli*) respectively (Tatsadjieu LN, 2003).

## **1.9 AIM AND OBJECTIVES**

### **1.9.1 AIM**

To qualitatively determine the bioactive constituents present in *xylopia aethiopica* (negro pepper) using conventional phytochemical screening methods.

### **1.9.2 OBJECTIVES**

To collect and prepare samples of *xylopia aethiopica* fruits for analysis

To extract the sample with ethanol using infusion method at room temperature

To carry out qualitative phytochemical screening of the ethanolic crude extract of *Xylopiya aethiopica*

To identify the presence of the bioactive compounds such as alkaloids, flavonoids, tannin, saponin, diterpene, lactones, sterols, triterpenes and glycosides.

## **CHAPTER TWO**

### **2.1 MATERIALS AND METHOD**

The xylopia Aethopia (Negro pepper) used in this project was purchased from Oja Oba market in Ilorin, Kwara State Nigeria.

The plant material was authenticated at the Department of Science Laboratory Technology (Biochemistry unit) Kwara State Polytechnic. The Negro pepper were washed with clean water to remove dirt, air dried at room temperature for two weeks, and then grounded into fine powder using a mechanical grinder. The powdered sample was stored in an air tight container until use.

### **2.2 APPARATUS AND EQUIPMENT**

Measuring cylinder, weighing balance, Beaker, round bottom flask, Burette, water bath, Conical Flask, Soxhlet extractor, heating mantle magnetic stirrer, foils, multifunctional

kitchen blender, test tube, test tube racks, test tube holder, thimble, separator funnel, Electrical weigh balance, glass rod, filter paper and distillation apparatus.

## **2.3 REAGENTS**

The reagents used were Absolute ethanol,  $H_2SO_4$ , Ferric oxides Fehling's solution, acetic acid, hydrochloric acid, distilled water, methanol, sodium carbonate, silver nitrate, sodium nitroprusside (SNP), DPPH solution drangendoff, sulphuric acid, lead acetate, acetic anhydride, Benedict's reagent, Hager's reagent, Mayer's reagent, Wagner's reagent.

## **2.4 EXTRACTION OF PLANT MATERIAL**

The negro pepper sample was pulverized using a high-powered multifunctional kitchen blender "SAMSUNG (model No:2022L) with 5000w and 32,000RP, made in Japan. 11.98g of the sample was packed into a cellulose thimble and placed in a

1L beaker 300ml of methanol solvent was measured and transferred to the beaker to cover the sample in the thimble.

A magnetic bar was placed at the bottom of the beaker. The beaker and its content were placed on a magnetic-stirrer temperature regulated hot plate. The cold maceration extraction was done at room temperature (i.e the heat knob of the hot plate was off) but the stirring knob was switched on so that the magnetic bar stirs at about 100rpm. The extraction was done for about 2hrs. The colored extract solution was removed and another 200ml of fresh methanol was added, and the extraction process repeated until the sample was exhaustively extracted. All the extractions were pulled together and transferred into a 1L round bottom flask. The extract solution was distilled to remove the methanol solvent. The concentrated extract was subsequently transferred into a beaker and placed in a water-bath. Heating was done until all solvent almost completely evaporated. The beaker

and its content were left to cool at ambient temperature and until it dried. The weight of the crude extract obtained was determined, from which the extract yield was calculated. The crude methanol extract of Negro pepper obtained was kept in the laboratory at ambient temperature for further analysis. The yield was calculated as follows.

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

## **2.5 PHYTOCHEMICAL SCREENING OF EXTRACT**

The extract was tested for the presence of bioactive compound by using the following standard methods.

### **2.5.1 Test for Steroids**

**2.5.1.1 Salkowski Test:** Chloroform solution of the extract when shaken with concentrated Sulphuric acid and on standing yield red colour.



**2.5.1.2 Lieberman Burchardt Test:** Chloroform solution of the extract with few drops of acetic anhydride and one ml of concentrated sulphuric acid from the sides gives reddish ring at the junction of 2 layers (Raaman N, 2006).

### **2.5.2 Test for Triterpenes**

**2.5.2.1 Solkowski Test:** Chloroform Solution of the extract when shaken with concentrated sulphuric acid, lower layer turns to yellow on standing.

**2.5.2.2 Lieberman Burchardt Test:** Chloroform solution of the extract with few drops of acetic acid and one ml concentrated sulphuric acid gives deep red at the junction of 2 layers (Silva GO, 2017).

### **2.5.3 Test for Alkaloids**

The extract was dilute with ammonia and then extracted with chloroform solution of this dilute hydrochloric acid was added. The acid was used for chemical test for alkaloids.

**2.5.3.1 Hager's Test (Saturated Solution of Pieric acid):** the acid layer with Hager's reagent gives yellow precipitate

**2.5.3.2 Mayer's Test (Potassium Mercuric Iodide):** The acid layer with few drops of Mayer's reagent gives a creamy white precipitate (Auwal MS, et al., 2014).

**2.5.3.3 Wagner's Test (Solution of Iodide in Potassium Iodide):** The acid layer with few drop of Wagner's reagent gives reddish brown colored precipitate.

**2.5.3.4 Dragendroff's Test (Solution of Potassium Bismuth Iodide):** Acid layer with few drops of dragendroff's reagent gives reddish brown precipitate (Singh V, 2017).

## **2.5.4 Test for Tannins**

**2.5.4.1 Ferric Chloride Test:** Extracts mixed with 1% ferric chloride solution gives blue, green or brownish green colour (Audu SA, 2007).

### **2.5.5 Test for Lactones**

**2.5.5.1 Legal's Test:** The extract mixed with mixture of sodium nitroprusside and Pyridine and treatment with methanol alkali gives deep red colour.

### **2.5.6 Test for Flavonoids**

**2.5.6.1 Lead acetate Test:** Alcoholic solution of the extract mixed with few drops of 10% lead acetate gives yellow precipitate (Tiwari P, 2011).

### **2.5.7 Test for Diterpenes**

**2.5.7.1 Copper acetate Test:** The extracts, mixed with solution of copper acetate gives green colour.

### **2.5.8 Test for Glycosides**

**2.5.8.1 Sodium hydroxide reagent:** Dissolve a small amount of alcoholic extract in 1ml water and add sodium hydroxide solution. A yellow colour indicates the presence of glycosides (Kumar R, 2018).

### **2.5.9 Test for Saponin**

Negro pepper was mixed with 2ml of distilled water in a test tube, the mixture was shaking vigorously and observed for the formation of persistent confirms and observed, foam that is presence of saponin (Nanna RS, 2013).

## CHAPTER THREE

### 3.0 RESULTS AND DISCUSSION

#### 3.1 RESULTS

The results of the qualitative screening of bioactive constituents in Negro pepper methanolic extract is present in the table below.

**Table 3.1.1: Result of Bioactive Constituents Test**

Bioactive Constituents	Result
Sterols	+
Alkaloids	+
Diterpenes	-
Tannins	+
Lactones	-

Triterpenes	+
Flavonoids	-
Glycosides	+
Saponins	-

**NOTE:** (+) Indicates present    (-) Indicates Absent

### **Percentage Calculation of Extract Yield**

The percentage crude extract yield of aethiopica (Negro Pepper) is calculated as follows:

Formula: % extract yield =  $\frac{\text{Weight of crude extract}}{\text{Weight of Sample}} \times 100$

Given values:

Weight of crude extract = 3.63g

Weight of Negro Pepper Sample = 11.98g

% extract yield =  $\frac{3.63}{11.98} \times 100$

$$\% \text{ extract yield} = \text{ } \times 100 = 0.303 \times 100$$

$$\% \text{ extract yield} = 30\%$$

Therefore the % extract yield = 30%.

### 3.2 DISCUSSION

The phytochemical screening of *xylopia aethiopica* carried out in this study revealed the presence of important secondary metabolites including alkaloids, flavonoids, tannins, steroids, lactones, triterpenes, glycosides, diterpenes (Ekong D 1968). These compounds are known to contribute to the medicinal value of many plants.

Alkaloids are known to possess antimicrobial and analgesic properties, which may support the use of *xylopia aethiopica* in the treatment of infections and pain. Flavonoids are well known for their antioxidant and anti-inflammatory properties. They play a key role in protecting the body against oxidative stress and the degenerative disease such as cancer and cardiovascular disorders. Tannins, which were detected in the extract are polyphenolic compounds with astringent properties.



They have been reported to exhibit antimicrobial, antiparasitic and wound-healing activities.

The presence of steroids and triterpenes suggests potential anti-inflammatory and anticancer properties. Both classes of compounds have been widely studied for their role in stabilizing cell membranes and interfering with tumor development. Diterpenes, although less commonly screened, have been associated with antimicrobial anti-viral and cytotoxic properties. Their presence in *xylopia aethiopica* indicates possible pharmacological uses in managing infections and certain chronic diseases. Glycosides, which were also detected, are important because they can act as prodrugs-biologically inactive compounds that are metabolized in the body to release active ingredients. Lactones detected through the legal test are compounds with known antibacterial and antifungal activities.

Their presence contributes to the broad-spectrum medicinal potential of the plant.

The detection of these bioactive compounds supports the use of *xylopia aethiopica* in herbal medicine for treating different health problems.

However, this project was limited to qualitative analysis. Future research involving quantitative phytochemical analysis and biological activity testing is essential to better understand the medicinal potential and safety of the plant (Silva GO, 2017).

## CONCLUSION

This study confirms that *xylopia aethiopica* (Negro pepper) contains several vital bioactive constituents including alkaloids, flavonoids, tannins, steroids, saponins, triterpenes, glycosides/diterpenes and lactones. These compounds contribute to the plant's well-documented therapeutic properties, supporting its traditional use in treating various ailments. The presence of these phytochemicals highlights the potential of *xylopia aethiopica* as a valuable source of natural drugs or health-promoting agents.

*Xylopia aethiopica* is not only a culinary spice but also a promising medicinal plant. Further studies including quantitative analysis and biological activity testing are recommended to fully explore and validate its pharmacological potentials.

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