

Bioinsecticidal Efficacy and Antioxidant Potency of *Jatropha curcas* and *Ocimum gratissimum* against *Anopheles gambiae* complex in Gombe State, Northeast Nigeria

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Abstract

Background: Safer control measures against malaria vector can address the problems of pollution and resistance from synthetic insecticides. This study aimed to determine the susceptibility of malaria vector to methanolic leaf extracts of *Jatropha curcas* and *Ocimum gratissimum* and their antioxidant profiles.

Methods: Fresh leaves of *Jatropha curcas* and *Ocimum gratissimum* were collected from Bajoga in northern Nigeria. The leaves were taxonomically identified and crude extracts obtained. Larvae of Anopheles mosquito were reared to adult stage and the adult females used for the adulticidal bioassay. CDC bottles were coated with 0.2g/L to 0.8g/L methanol leaf extracts of the plants and 10 – 25 female Anopheles introduced into each test bottle to observe mortality. Mortality was assessed at 0 hour, after 2 hours and after 24 hours. Lethal Concentrations and mortality were determined using LC^{50} and LC^{90} . Standard procedures were followed for qualitative and quantitative screening and antioxidant activity. Excel spreadsheet was used for data analysis.

Results: Findings revealed adulticidal efficacy of *Jatropha curcas* and *Ocimum gratissimum* of 100% mortality with LC^{50} value of 0.01g/L and LC^{90} value of 0.4g/L after 24 hours against *Anopheles gambiae*. Qualitative phytochemical screening of the leaf extract of *Jatropha curcas* contained alkaloids, flavonoids, tannins, saponins anthraquinones, phenolics and glycosides while that of *Ocimum gratissimum* had steroids in addition. The percentage inhibition of 2, 2-diphenyl-1-picryl-hydrazyl-hydrate radical scavenging potential was dose dependent. *Ocimum gratissimum* showed higher inhibition potency than *Jatropha Curcas*.

Conclusion: The study reported adulticidal efficacy of the two plants and can be purified for controlling malaria vector.

Keywords: Susceptibility, Malaria vector, *Ocimum gratissimum*, Phytochemical, Antioxidant profile

Introduction

Diverse methods have been used to lessen the prevalence of malaria globally. The most common measure include the use of Indoor Residual Spraying (IRS). The achievement of IRS of houses to control adult female anopheline vectors of malaria depends on mosquitoes resting indoors before or after feeding, access to the interior of all houses, the presence of walls and surfaces to be sprayed in human shelters and willingness of people to use their vicinity.¹

Larval control measures also play principal roles and are among the major components of malaria control programs in areas where mosquito breeding sites are accessible. This is because unlike the adult mosquitoes that can easily detect and avoid synthetic indoor residual spray chemicals, immature stage mosquitoes are confined within relatively small aquatic habitats and cannot readily escape control measures.² Both the larval and the adult mosquitoes are controlled using synthetic chemicals such as temephos, fenthion, malathion, chloropyrifos; and methoprene.³ However, there are reports that the use of those synthetic compounds have triggered the development of physiological resistance by the adult mosquitoes.⁴

Furthermore, widespread use of synthetic insecticides are reported to cause environmental degradation. Hence, the necessity to identify safer and environmentally friendly insecticidal substances from plant materials. Many studies have reported the existence of diverse plants belonging to different families in different parts of the world with various levels of insecticidal potency against malaria vector.³ Plants are the richest resources for drugs in the traditional and modern systems of medicine, pharmaceutical intermediates and chemical entities for synthetic drugs. Medicinal plants are of great importance to the health of individuals and the communities. The medicinal values of some plants lie in some chemical substances that produce definite physiological actions in the human body. The most important of these bioactive constituents are alkaloids, tannis, flavonoids and phenolic compounds and are used to cure diseases such as parasitic infections and cancer.⁵

About 33 medicinal plants belonging to 27 families have been screened for their mosquitocidal activities. However, considering the abundance of plants with their medicinal values, the level of nuisance caused by the mosquitoes and the burden of malaria in Nigeria, current level of research in Nigeria on the adulticidal efficacy of plants against the mosquito vector is low.⁶ Thus, this research was planned to provide such needful information in order to fill the gap.

Methodology

Study Area

Gombe State is located between latitude 10°16'44.91"N and longitude 11°10'23.02"E and bordered on the east by Adamawa and Borno States, Yobe State to the north, Taraba and Bauchi States to the south and west respectively. The southern part of Gombe State has many hills while flat landscape is the most visible feature of the northern part of the State.⁷ It is characterized by Sudan savanna vegetation with trees of up to 3 m in height, with an annual rainfall of 850 mm. The State has a population of 2,353, 879 people.⁸ Wet and dry seasons is the dominant climate of Gombe State with the extreme temperatures of up to 38°C during the hot dry seasons, usually in March to May. The inhabitants of Gombe metropolis comprise mostly of civil servants, some fulltime farmers and traders. The tribes are mixed but the predominant ones are Hausa, Fulani, Tangale, Waja, Tera, Bolawa. Malaria is an endemic parasitic disease in Gombe State and many researchers have reported the burden of the infection in Gombe State. Such prevalence include 68.4%, 74.9% and 8.48%.⁹⁻¹¹ The presence of malaria vector and the species responsible for transmitting the malaria parasite such as *Anopheles gambiae* s.s. has been reported in Gombe State and a variety of medicinal plants capable of mosquitocidal activity against *Anopheles gambiae* s.s. has been likewise been documented in Gombe state.¹²⁻¹⁴

Collection of plant materials

The mosquitoes and the plants samples were all collected from Gombe State, the mosquitoes from within Gombe metropolis while the plant samples from Bajoga, Funakaye Local Government Area

(LGA). Fresh leaves of *J. curcas* and *O. gratissimum* were collected from Shuwari (latitude 10.879840° N and longitude 11.445795 E) and Bulagaidam (latitude: 10.867570° N and longitude 11.491756° E) villages respectively of Bajoga, Funakaye Local Government Area (LGA) of Gombe State, Nigeria, between March and April 2021. Shuwari and Bulagaidam are located 2 km and 5 km north and east respectively of the capital city of Bajoga. The leaves were taxonomically identified by an experienced botanist using the plant identification keys, in the Department of Biological Sciences, Gombe State University, where voucher specimen number were prepared and deposited at the herbarium.

Processing and extraction of the plant material

The extraction of the plant materials was done using the established standard method.¹⁵ Leaves of *J. curcas* and *O. gratissimum* were air dried under shade and ground using a grinding mill (Straub Model4-E, Philadelphia USA). One hundred grams of the sample was macerated with 900 ml of methanol and shaken for 1 hour and was left standing for 24 hours and then shaken for another 1 hour using orbital shaker, and then filtered with No. 1 whatman filter paper. The filtrate was subjected to Rotary Evaporator to obtain a residue called crude extract. The crude extract was stored in a refrigerator at 4 °C until needed for bioassay.

Mosquito culture and processing

The larvae of the mosquitoes were collected from various breeding sites in Gombe LGA of Gombe State. Sampling of larvae was done using dipper. Collected larvae were transported inside a container to the laboratory of the Department of Biological Sciences, Gombe State University. They were reared to adult mosquitoes in the insectary, maintained on a 10% sugar solution and were sorted into males and females based on the morphological characteristics of their antennae, palps and proboscis. The females were kept in a separate container for the bioassay.

Adulticidal bioassay procedure using CDC bottles

The adulticidal bioassay was conducted at the

Biological Science laboratory of Gombe State University. The preparation, coating of the CDC bottles and adulticidal bioassay potency of the extracts were determined using established procedures.¹⁵⁻¹⁷ The extracts were dissolved in 98% methanol. The choice of this chemical was because of the ability of this solvent to evaporate very fast. To obtain the concentrations of 0.2g/L, 0.4g/L, 0.6g/L and 0.8g/L/bottles of the methanol extracts, products were dissolved in adequate quantities of acetone to make 10 ml of total solution. Each 1 ml of this solution contained 0.2g/L, 0.4g/L, 0.6g/L and 0.8g/L of methanol extracts. After cleaning and drying the bottles, 1 ml of the solution of each concentration of the prepared extract was added to the bottles. One ml of 98% methanol was added to the control bottle. The content of each bottle was swirled and inverted by gentle rotation so that the sides, all the way around were coated. After that, the caps were removed and the bottles continued rolling on their sides until all visible signs of the liquid were gone from inside and the bottles were completely dried. The bottles were left for 24 hours on their sides and covered with aluminium foil to keep them protected from light.

Five 250ml Wheaton bottles with screw lids were properly cleaned and dried. After drying, they were coated with 1ml of 0.2g/L, 0.4g/L, 0.6g/L, 0.8g/L of the extracts by swirling assuring complete coating of the bottle and its cap. One ml of absolute alcohol was added to the control bottle. Ten to twenty five mosquitoes were introduced into each test bottle including the control bottle using an aspirator. At start time (Time 0), the bottles were examined to count the number of dead and live mosquitoes. Mortality was assessed after 2 hours and subsequently 24 hours after the mosquitoes' introduction and mosquitoes were considered dead if they could no longer stand.¹⁸ The experiment was done under normal room conditions. Abbott's formula was used to correct results if the mortality in the control bottle was between 3% and 10%. The bioassay results were discarded if mortality in the control bottle at the end of the test was >10%.

Corrected Mortality =

$$= \frac{\% \text{ mortality in test bottle} - \% \text{ mortality in control bottle}}{100 - \% \text{ mortality in control bottle}} \times 100$$

Standard methodology according to World Health Organization (WHO) and Centers for Disease Control and Prevention (CDC) was followed in the determination of the lethal concentration (LC) doses of adult's mortality (LC⁵⁰ and LC⁹⁰.¹⁹)

Procedure for qualitative phytochemical screening

The phytochemical and antioxidant activity screening was done at the Biochemistry Laboratory, Faculty of Science, Gombe State University. Qualitative phytochemical screening of the plant samples were done based on standard methods.²⁰

Alkaloid

A weighed amount (2g) of each powdered sample of the plants was transferred into a 250ml beaker. Two hundred ml (200 ml) of 10 % acetic acid was added and then covered to stand for 4 hours. Filtration was done, and water bath used to apply the concentration of the extracted content to one quarter of original volume. Drop-wise addition of concentrated ammonium hydroxide to the extract followed until the precipitate was complete. The entire solution was allowed to settle and collection of the precipitate was done by filtration and then weighed.²¹

Anthraquinone

Using Borntrager's test, about 50 mg of the extract was heated with 1ml 10% ferric chloride solution and 1ml of concentrated hydrochloric acid.²² The extract was cooled and filtered and the filtrate was shaken with equal amount of diethyl ether. The ether extract with strong ammonia Pink or deep red coloration of aqueous layer was observed. The appearance of the pink to red colour indicated the presence of anthraquinone.

Flavonoid

To determine the flavonoid content in the leaves of the *Jatropha* species studied, the aluminium chloride colorimetric method was employed. One millilitre of each plant extract was mixed with 2 ml of methanol, 0.2 ml of 10 % aluminium trichloride (AlCl₃), 0.2 ml of 1M potassium acetate, and 5.6 ml of distilled water. The entire mixture was allowed to stand at room temperature for 30 minutes after

which the absorbance was measured at 420 nm. The total flavonoid content in each plant part was expressed in terms of standardized quercetin equivalent (QE) per 100mg of each extracted compound.

Terpenoid

The method of ferguson (1956) was used to evaluate the total terpenoid contents of the leaves of the studied species.²³ Ten grams of plant powder were taken separately and soaked for 24 hours. After filtration, the filtrate was extracted with petroleum ether and the ether extracted was treated as total terpenoids. The assay was carried out in triplicate.

Saponin

Standard method was used for the determination of saponin.²¹ About 100 cm³ of 20% aqueous ethanol was added into a conical flask containing 20 g of the powdered samples. The mixture was properly shaken together, and then heated over a hot water bath for 4 hours with continuous stirring at 55° C. It was filtered and re extraction of residue was performed with 200 ml of 20 % aqueous ethanol. The combined extracts were then reduced to 40 ml over water bath at 90° C after which the concentrate was transferred into a 250 ml separating funnel with the addition of 20 ml diethyl ether, and shaken vigorously. The aqueous layer of the solution was recovered while the ether layer was discarded. The purification process was then repeated and thereafter, 60 ml of n-butanol was added. The combined n-butanol extracts were washed twice with 10 ml of 5% aqueous sodium chloride and the remaining solution was heated in a water bath. After evaporation the samples were dried in the oven to a constant weight, the saponin content was calculated as percentage at a wavelength of 380 nm.

Tannin

The tannin content was determined using standard procedure.²¹ Half a gram of the dried powdered leave sample was weighed into a 50 ml plastic bottle and 50 ml of distilled water was added and then shaken thoroughly for about 1 hour. The solution was filtered into a 50ml volumetric flask and made up to the mark. Five millilitre of the filtrate was pipetted out into a test tube and mixed

with 2 ml of 0.1M FeCl₃ in 0.1N HCl and 0.008M potassium ferrocyanide. The absorbance was measured at 120 nm within 10 min.

Procedure for quantitative phytochemical screening

Quantitative phytochemical screening of the two plants was conducted according to the method described by Obadani & Ochuku.²⁴

Preparation of fat free sample

Two grams of the sample were defatted with 100 ml of diethyl ether using a Soxhlet apparatus for 2 hours.

Determination of total phenols by spectrophotometric method

The fat free sample was boiled with 50ml of ether for the extraction of the phenolic component for 15 minutes. Five millilitre of the extract was pipetted into a 50ml flask, then 10ml of distilled water was added. Two millilitre of ammonium hydroxide solution and 5 ml of concentrated amyl alcohol were also added. The samples were made up to mark and left to react for 30 minutes for colour development. This was measured at 505nm.

Alkaloid determination

A 5g sample of the plant was weighed into a 250 ml beaker and 200 ml of 10% acetic acid in ethanol was added, covered and allowed to stand for 4 hours. This was filtered and the extract concentrated on a water bath to one quarter of the original volume. Concentrated ammonium hydroxide was added drop wise to the extract until the precipitation was complete. The whole solution was allowed to settle and the precipitate was collected and washed with dilute ammonium hydroxide and then filtered. The residue was the alkaloid, which was dried and weighed.

Flavanoid determination

A 10g plant sample was extracted repeatedly with 100 ml of 80% aqueous methanol at room temperature. The whole solution was filtered through whatman filter paper No 42 (125 mm). The filtrate was later transferred into a crucible and evaporated to dryness over a water bath and

weighed to a constant weight.

Saponin determination

A 20g plant sample was dispersed in 200 ml of 20% ethanol. The suspension was heated over a hot water bath for 4 hours with continuous stirring at about 55°C. The mixture was filtered and the residue re-extracted with another 200 ml of 20% ethanol. The combined extracts were reduced to 40 ml over water bath at about 90°C. The concentrate was transferred into a 250 ml separating funnel and 20 ml of diethyl ether was added and shaken vigorously. The aqueous layer was recovered while the ether layer was discarded. The purification process was repeated. Then 60 ml of normal butanol extracts were washed twice with 10 ml of 5% aqueous sodium chloride. The remaining solution was heated in a water bath. After evaporation the sample was dried in the oven into a constant weight. The saponin content was calculated in percentage.

Determination of antioxidant activity of 2, 2-diphenyl-1-picrylhydrazyl (DPPH) radical

Slight modifications were done to the standard method used for the determination of scavenging activity of DPPH free radical in the extract solution.²⁵ Briefly, a 2.0 ml solution of the extract at different concentrations diluted two-fold in methanol was mixed with 1.0 ml of 0.3 mM DPPH in methanol. The mixture was shaken vigorously and allowed to stand at room temperature in the dark for 25 minutes. Blank solutions were prepared with each test sample solution (2.0 ml) and 1.0 ml of methanol while the negative control was 1.0 ml of 0.3 mM DPPH solution plus 2.0 ml of methanol. L-ascorbic acid was used as the positive control. Thereafter, the absorbance of the assay mixture was measured at 518 nm against each blank with a UV-visible spectrophotometer.²⁶ DPPH radical inhibition was calculated using the equation:

% Inhibition =

$$x = \frac{A_{\text{Control}} - A_{\text{Sample}}}{A_{\text{Control}}} \times 100\%$$

Where, A_0 is the absorbance of the control, and A_s is the absorbance of the tested sample. The IC_{50} represented the concentration of the extract that inhibited 50% of radical.

Data Analysis

Data was entered and processed in Excel spreadsheet (Microsoft® Excel® for Microsoft 365 MSO (Version 2110). Mean percentage mortality of adult *Anopheles* mosquitoes and the concentration in g/L lethal to 50% (LD_{50}) and 90% (LD_{90}) with its 95% confidence interval (CI) and chi-square were computed using IBM® SPSS® Statistics (Version 25). The IC_{50} value of test plants, which is the concentration of sample required to inhibit 50% of the DPPH free radical was calculated using log transformed dose inhibition curve in GraphPad Prism (Version 9.3.1).

Results

Susceptibility of female *Anopheles* mosquitoes to *J. curcas* and *O. gratissimum* methanolic leaf extract

It was revealed that the susceptibility of adult female *Anopheles* mosquitoes to graded concentrations of *J. Curcas* leaf extract was dose dependent. A mean mortality rate of 73.2% was noted at a concentration of 0.2 g/L. However, at 0.8 g/L concentration of *J. curcas* leaf extract treatment, mean mortality was 100% (Table 1). Fifty percent mortality (LD_{50}) was observed at 0.099 g/L (95% CI: 0.003 – 0.183 g/L) while the concentration of *J. curcas* leaf extract lethal to 90% of adult *Anopheles* mosquitoes (LD_{90}) was 0.419 (95% CI: 0.273 to 0.984 g/L) (Table 2).

Extracts of *Ocimum gratissimum* gave 100% mortality against adult *Anopheles* mosquitoes after 24 hours at 0.2 g/L, 0.4 g/L, 0.6 g/L and 0.8 g/L (Table 1). Mortality observed in the control group was between 0 and 2%. The LD_{50} and LD_{90} for *O. gratissimum* leave extract against adult *Anopheles* mosquitoes were not computed due to 100% mortality obtained at various concentration.

Phytochemical components of *J. curcas* and *O. gratissimum* methanolic leaf extract

Qualitative assessment of the phytochemicals conducted on *Jatropha curcas* and *Ocimum*

gratissimum revealed the presence of alkaloids, flavonoids, tannins, saponins anthraquinones, phenolics and glycosides in both plants (Table 3).

Comparative analysis between *J. curcas* and *O. gratissimum* revealed that in both plants, tannins, glycosides and anthraquinones were detected in excess, moderate and trace amounts. Flavonoids was present in excess in *J. curcas* but in trace amounts in *O. gratissimum*. *J. curcas* recorded moderate levels of alkaloids while excess amounts were detected in *O. gratissimum*. The saponin were moderately detected in *J. curcas* while only trace levels were present in *O. gratissimum*. The opposite was true for phenols – trace amounts were presents in *J. curcas* but moderately present in *O. gratissimum*. Steroids were absent in *J. curcas* but was moderately detected in *O. gratissimum* (Table 3).

Quantitative analysis result (Table 4) shows the concentration of the different phytochemicals in *J. curcas* to be in the following decreasing order: tannins 552 mg/100g eqv., saponins 439 mg/100g eqv., flavonoids 319 mg/100g eqv., phytates 294 mg/100g eqv., t. phenols 286 mg/100g eqv., alkaloids 235 mg/100g eqv. and oxalate 152 mg/100g eqv.

The phytochemicals with the highest concentration detected in *O. gratissimum* were tannins 823.29 mg/100g eqv. followed by t. phenols 562.25 mg/100g eqv. and alkaloids 385.10 mg/100g eqv. The concentrations of other phytochemicals detected include oxalate 228.60 mg/100g eqv. steroids 162 mg/100g eqv. and saponins 141 mg/100g eqv. Flavonoids and phytates had the least concentrations in *O. gratissimum* – 107.3 and 73.80 mg/100g eqv. respectively.

Between the two plants, *O. gratissimum* had higher concentrations of tannins, phenols, alkaloids and oxalate, while the concentrations of saponin, flavonoids and phytates were higher in *J. curcas* (Table 4).

Antioxidant activity of the leaf extract of *Jatropha curcas* and *Ocimum gratissimum*

DPPH radical scavenging activities of the leaf extract of *Jatropha curcas* and *Ocimum gratissimum* are shown in Figure 1 and Figure 2 respectively. The result showed radical scavenging

ability in a dose dependent manner. At the highest concentration of 500 µg/ml, leaf extract of *O. gratissimum* showed a higher inhibition activity at 93.02% (Figure 1) compared to *J. curcas* at 69.62% (Figure 1). The IC₅₀ value of *J. curcas* and *O. gratissimum* in the DPPH assay was 247.9 µg/ml and 77.98 µg/ml respectively. Ascorbic acid as the

reference compound alongside for *J. curcas* and *O. gratissimum* showed the highest inhibition activity at 84.81% and 95.25% respectively.

Table 1. Susceptibility of adult female *anopheles* mosquitoes to *Jatropha curcas* and *Ocimum gratissimum* methanolic leaf extract

| Plant extract | % mortality, mean ± SD | | | |
|---------------------------|------------------------|-------------|-------------|-------------|
| | 0.2 g/L | 0.4 g/L | 0.6 g/L | 0.8 g/L |
| <i>Jatropha curcas</i> | 73.2 ± 11.4 | 92.4 ± 14.8 | 88.3 ± 16.9 | 100 ± 0.0 |
| <i>Ocimum gratissimum</i> | 100.0 ± 0.0 | 99.0 ± 2.0 | 100.0 ± 0.0 | 100.0 ± 0.0 |

Table 2. Probit analysis of adulticidal toxicity of *Jatropha curcas* and *Ocimum gratissimum* leaf extract against adult *anopheles* mosquitoes

| Plant extract | LD ₅₀ g/L (95% CI) | LD ₉₀ g/L (95% CI) | χ ² (df = 2) |
|--------------------------|-------------------------------|-------------------------------|-------------------------|
| <i>Jatropha curcas</i> | 0.099 (0.003 – 0.183) | 0.419 (0.273 – 0.984) | 2.889 |
| <i>Ocimumgratissimum</i> | NA | NA | NA |

LD: Lethal dose; CI: Confidence interval; df: Degree of freedom; NA: Not applicable

Table 3. Qualitative screening of phytochemicals in *Jatropha curcas* and *Ocimum gratissimum*

| Phytochemicals | <i>Jatropha curcas</i> | <i>Ocimum gratissimum</i> |
|----------------|------------------------|---------------------------|
| Tannins | +++ | +++ |
| Flavonoids | +++ | + |
| Alkaloids | ++ | +++ |
| Saponins | ++ | + |
| Glycosides | ++ | ++ |
| Phenolics | + | ++ |
| Anthraquinones | + | + |
| Steroids | -- | ++ |

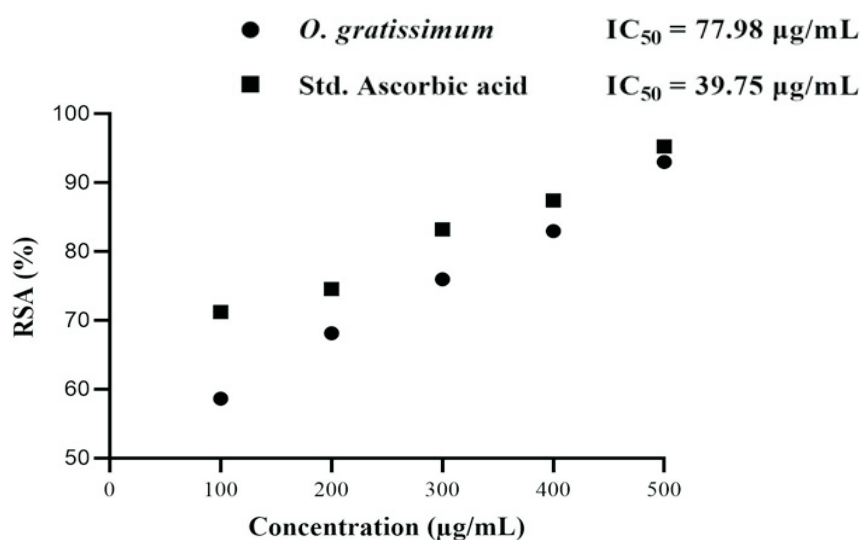
+ (trace); ++ (moderate); +++ (excess); -- (absent).

Table 4. Concentration of phytochemicals present in *Jatropha curcas* and *Ocimum gratissimum*

| Phytochemicals | Concentration; mg/100g eqv. | |
|----------------|-----------------------------|---------------------------|
| | <i>Jatropha curcas</i> | <i>Ocimum gratissimum</i> |
| Tannins | 552.00 | 823.29 |
| Saponins | 439.00 | 141.00 |
| Flavonoids | 319.00 | 107.13 |
| Phytates | 294.00 | 73.80 |
| T. Phenols | 286.00 | 562.25 |
| Alkaloids | 235.00 | 385.10 |
| Oxalate | 152.00 | 228.60 |
| Steroids | --- | 162.00 |

eqv: Equivalent

Figure 1. DPPH radical scavenging activity (RSA) of methanolic extract of *Jatropha curcas*



at different concentrations.

Figure 2. DPPH radical scavenging activity (RSA) of methanolic extract of *Ocimum gratissimum* at different concentrations.

Discussion

The findings of this study revealed that the extract of the *Jatropha curcas* indicated adulticidal efficacy of 100% of mortality and LD⁵⁰ value of 0.01g/L and LD⁹⁰ value of 0.4g/L after 24 hours against *An. gambiae*. This adulticidal efficacy of the *Jatropha curcas* may be attributed to the presence of bioactive constituents, such as tannins, anthraquinones and steroids that are toxic to mosquitoes as documented in a research conducted at Enugu to evaluate the toxicity of *Jatropha curcas* against mosquitoes.²⁷ The effectiveness of this plant could also be attributed to the presence of saponin, a phytochemical component known to be insecticidal as documented in a study in Japan.²⁸ The reason behind the high potential of *J. curcas* leaf extracts on *An. gambiae* was due to its high composition of saponins present in the extract compared to *O. gratissimum*. Saponin works by interacting with the cuticle membrane of the larvae and possibly the adult mosquito, changing the microstructure of the cell membranes and finally disrupting the membrane, which is one of the likely reasons for adult mosquitoes death.²⁸ Insecticidal efficacy of *J. curcas*, which tallies with the findings of this research was also reported elsewhere in Nigeria.²⁹ The results obtained in this study agree with the findings from a study which reported an efficacy of *Jatropha curcas* against *Anopheles gambiae* with 87% mortality in a research conducted in Potiskum, Yobe State, Nigeria.³⁰ The result however, disagrees with the findings from another study³¹ with a methanol extract of *Jatropha curcas* against *Culex quinquefasciatus* showing highest mortality at the LD⁵⁰ value of 1.2%. The reason for the disparity may be due to the genera of the mosquitoes subjected to the adulticidal test.

On the other hand, the leaf extract of *Ocimum gratissimum* reveals the adulticidal potency of 100% of mortality of the *Anopheles gambiae* after 24 hours. All the mosquitoes became susceptible after 24 hours and this signified the potential efficacy of the leaf extract of the plants. The reason for this highly potent adulticidal activity may be attributed to phytochemical constituents of the plants. The results agree with the finding of a research conducted to investigate larvicidal, pupicidal and adulticidal activities of acetone,

hexane and chloroform extracts of *Ocimum gratissimum* against filariasis mosquito vector, *Culex quinquefasciatus*.³² The result differed from others where the activity of *Ocimum gratissimum* against the larval stage of *Culex quinquefasciatus* was drastically lower than the one reported in this research.^{33,34} The reason for the discrepancies may be due to the type of chemical used for the extraction of the active elements from the plant. It can also be attributed to the genera/species of mosquitoes to which the extract was subjected, and it was reported that *Culex quinquefasciatus* was more resistant to chemical than *Anopheles gambiae*.³⁵

The qualitative phytochemicals screening of the leaf extract of *Jatropha curcas* was found to have presence of alkaloids, flavonoids, tannins, saponins, anthraquinones, phenolics and glycosides while that of *Ocimum gratissimum* have steroids in addition to the alkaloids, flavonoids, tannins, saponins, anthraquinones, phenolics and glycosides, which are in form of secondary metabolites majorly for the protection of the plants. Previous studies have reported these phytochemicals in *Jatropha curcas* and the studies include those conducted in Ibadan and Ogun state, Nigeria.^{36,37} The reasons for the discrepancies may be owing to the extraction method used.

The phytochemicals screening of *Ocimum gratissimum* revealed the presence of alkaloids, flavonoids, tannins, saponins, anthraquinones, phenolics and glycosides and steroids. Findings from previous studies are also in line with the findings of this study and also reported similar bioactive constituents.³⁷⁻³⁹ The current finding disagrees with the report in a study in Imo State, Nigeria to evaluate phytochemicals content and antioxidant potential of *Ocimum gratissimum* and *Telfairia occidentalis* leaves.³⁷ They reported the bioactive constituents without anthraquinone, steroid and glycosides, which are found in high contents in this study.³⁷

These compounds are known to be biologically active and therefore aid the antioxidant activities of *O. gratissimum*. Alkaloids was reported to act against foreign organisms due to their toxicity while flavonoid exhibited a wide range of biological activities like antimicrobial, anti-inflammatory, anti-angionic, analgesic, anti-

allergic, cytostatic, and antioxidant properties.^{39,40} It was observed that the quantitative concentration of saponin was just slightly higher in *J. curcas* compared with that of *O. gratissimum*. It has been reported that saponin has a range of biological activities and potential health benefits such as antioxidant activity among others.³⁷ Saponin has also been observed to kill protozoans and molluscs and act as an antifungal and antiviral agent.⁴¹ Tannins were higher in *O. gratissimum* compared to *J. curcas*. Tannin-containing plant extracts are used as antioxidants and also as astringents against diarrhoea, as diuretics, against stomach and duodenal tumors, and as anti-inflammatory, antiseptic and haemostatic pharmaceuticals.⁴² Tannins have been used traditionally for the treatment of diarrhoea, haemorrhage and detoxification⁴³ and were found in both plants qualitatively but were more concentrated in *O. gratissimum*. The composition of tannins as observed in this study may justify traditional usage of the plant extract in the management of diarrhoea.³⁷ The concentration of phenols was quantitatively higher in *O. gratissimum* than *J. curcas*. Phenolic acid possesses diverse biological activities, such as antiulcer, anti-inflammatory, antioxidant, cytotoxic and anti-tumor, anti-spasmodic, and antidepressant activities. This implies that *J. curcas* and *O. gratissimum* might have a high potential in reducing oxidative stress.³⁷

Another secondary metabolite compound recorded in the leaf extracts of *O. gratissimum* was alkaloid though its concentration compared with *J. curcas* had no significant difference. Alkaloids have many pharmacological activities and this implies that both *J. curcas* and *O. gratissimum* might have great potentials in preventing malaria, hypertension, cancer and cough (codeine). It was observed in this study, that the quantitative concentration of flavonoid was significantly higher in *J. curcas* when compared with that of *O. gratissimum*. Flavonoids have been documented to play roles as antimicrobials, cytotoxics, anti-inflammatory as well as antitumor activities but the best-described property of almost every group of flavonoids is their capacity to act as antioxidants, which can protect the human body from free radicals.⁴⁴

Flavonoids constitute a wide range of substances that play important roles in protecting biological systems against the harmful effects of oxidative processes on macromolecules, such as carbohydrates, proteins, lipids and DNA.⁴⁵ This implies that both *J. curcas* and *O. gratissimum* might have higher potential in fighting against free radicals and in preventing harmful damage of DNA (which could lead to aging), lipids (which causes lipid peroxidation) and protein (which could lead to mutation).³⁷

A high concentration of phytates was revealed in this study in *J. curcas* when compared with *O. gratissimum*. Phytates have been reported to play principal roles as antioxidants and help in the prevention of heart diseases. This investigation disagrees with the findings where low concentration of phytates was reported in *O. gratissimum*.³⁷ The concentration of this bioactive compound as shown in this study could contribute to the significant antioxidant potency of these plants and consequently support its local usage for the treatment of free radical related diseases for many years. The DPPH scavenging activity assay has been utilised to gain an understanding of antioxidant potentials.⁴⁶ The percentage inhibition of 2,2-diphenyl-1-picryl-hydrazyl-hydrate (DPPH) radical scavenging potential was observed to be dose dependent. *O. gratissimum* showed higher inhibition potency when compared with *J. curcas*. This shows that *O. gratissimum* has more scavenging activity and in effect more helpful in protecting the body system from oxidative stress through removing free radicals. This finding disagrees with the results in a study that reported the scavenging activities in decreasing order with decreasing concentration in *O. gratissimum* in a research to evaluate the phytochemical contents and antioxidant potential of *O. gratissimum* and *Telfaria occidentalis* leaves in Imo State, Nigeria.³⁷ The findings agree with the study conducted in Benin City, Nigeria.⁴⁰ The results obtained from this study are in line with the previous report where the antioxidant potential of *J. curcas* was due to high concentration of phenolic compounds.³⁹ Consequently, the strong antioxidant activity of *J. curcas* and *O. gratissimum* as shown in the present might be related to high contents of flavonoids and

phenolic compounds respectively, as revealed by the qualitative and quantitative analysis. This simply implies that both plants under study have antioxidant capacity and as such, can effectively scavenge free radicals and prevent stress.

Conclusion

The findings of this study confirmed the susceptibility of adult female *Anopheles* mosquitoes to *J. curcas* and *O. gratissimum* methanolic leaf extracts. The study likewise confirmed the presence of active metabolites in both plants which qualify them as potential adulticidal alternative in the control of malaria vector. The bioactive constituents also indicate that the plants are potential antioxidant agents capable of scavenging free radicals. These plants can be used for the development of easily biodegradable insecticides which will serve as best alternatives to the expensive and environmentally hazardous existing insecticides to which the mosquitoes are already developing resistance. Evidently, *O. gratissimum* methanolic leaf extracts showed the best efficacy in the control of *A. gambiae*, the vector of malaria and the more potent antioxidant in this study. We recommend that the two plants can be optimised and further purified to be used as insecticide against malaria vector.

Conflict of Interest

Authors declare that they have no conflicts of interest.

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