

## EVALUATION OF ANTIPLASMODIAL EFFECTS OF BLACK SEEDS, FENUGREEK AND CORIANDER SEEDS IN ALBINO MICE INFECTED WITH *Plasmodium berghei*.

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### ABSTRACT

*Malaria remains a disease of public concern according to reports by the World Health Organization which recommends diverse therapeutic approach including the use of plant derived products to combat drug resistance in malaria treated individuals. This study therefore, evaluates the antiplasmodial effects of black seeds (Nigella sativa), fenugreek (Trigonella foenum-graecum) and coriander seeds (Coriandrum sativum) and their effects on packed cell volume and weight of experimental mice. Phytochemical analysis of the extract was conducted using Gas Chromatography-Mass Spectrometry analysis to establish the presence of antimalarial constituents. The plant extracts were screened for curative activities at dosages of 50mg/kg, 100mg/kg and 200mg/kg, in mice inoculated with Plasmodium berghei from donor mice. Chloroquine (6mg/kg body weight) was used as standard control drug. A 4-day curative assay was used to evaluate the antimalarial potentials of the plant extracts. Comparison between treated samples were analyzed using single factor analysis of variance at 95% confidence interval and 5% significance level (P<0.05). The results obtained from this study revealed significant suppression rate at 50mg/kg for Nigella sativa (67.0%), 200mg/kg for Trigonella foenum-graecum (68.3%), and 200mg/kg for Coriandrum sativum (66.7%). Chloroquine (6mg/kg) showed a higher inhibitory performance at 86.6% on day 7 post treatment compared to the plant extracts. The packed cell volume increased in groups treated with chloroquine compared to the extract treated group while the weight of mice increased in all extract groups significantly compared to the chloroquine treated group. The results of the study were statistically significant at P<0.05.*

**Key words:** Antimalarials; *Plasmodium*; Phytochemicals; Ethnopharmacology.

### INTRODUCTION

Malaria remains a disease of public concern according to data infographic reports by the World Health Organization which estimates about 241 million malaria cases and puts mortality cases as a result of malaria at 627,000 worldwide as at 2020, a high trend compared to reports accumulated in 2019 with

229 million cases of malaria and 409,000 deaths worldwide, largely attributed to bridges in the provision of malaria therapy as prevention, diagnosis and treatment measures during the pandemic era (WHO, 2020). Demographic statistics acknowledges pregnant women, children (<5years) and those infected with Acquired Immune Deficiency

Syndrome (AIDS) as the most vulnerable to malaria disease.

Plants contain active or complementary compounds referred to as secondary metabolites which are useful for treatments (Pirintsos *et al.*, 2017; Hartmann, 2007). Natural products offer immense contribution in the control of malaria acting as substantial components in antimalarial drug production. For instance, quinine, a product of the Cinchona tree bark, is one of the earliest natural compounds manufactured for the treatment of malaria and a template for the production of derivatives such as chloroquine, mepacrine, primaquine, and mefloquine (De Oliveira *et al.*, 2009). Worldwide analysis puts over 1200 plant species as active sources of treatment for malaria and its symptoms as reported by Wilcox *et al.* (2011).

*Nigella sativa* is a flowering plant, native to Asia, the Mediterranean and Africa. This plant is also known as black seed or cumin. These seeds have been found to contain potential phytochemicals, predominantly quinine compounds (thymoquinone) potent for antimalarial activities. The presence of thymoquinone in black cumin seeds is responsible for its therapeutic, anti-inflammatory and anti-cancerous potentials (Tembhurne *et al.*, 2014; Burits and Bucar, 2000; Mansour *et al.*, 2001). The antimalarial activity of these ethnopharmacological plant can be confirmed in various researchers (Promise *et al.*, 2014; Udu *et al.*, 2021; Aschroft *et al.*, 2018; Oreagba *et al.*, 2013; Rhama *et al.*, 2021) report on black cumin seed. These researchers attest to the prophylactic, suppressive and curative potentials of these seeds and also identify the presence of metabolites referred to as phytocompounds (phenol, tannins, terpenoids, alkaloids, saponin, carbohydrate, flavonoids, anthraquinones, cardiac glycosides and proteins components etc.) in the seed extract.

Fenugreek is an annual plant, native to the Mediterranean, Europe and Asia region but is now cultivated in central and southern Europe, India and Northern Africa. The seeds of

fenugreek contain polyphenol compounds such as rhapoticin and isovitexin, major bioactive compounds as reported by He *et al.*, 2015. A study by Palaniswamy *et al.* (2010) was conducted to analyze the anti-plasmodial activity of *Trigonella-graecum* L. in vitro with isolates of *Plasmodium falciparum* that have been modified for use in laboratories and are both sensitive and resistant to chloroquine. Multiple extracts (methanol, ethanol, butanol, chloroform, ethyl acetate) used for the study displayed potent antimalarial capabilities and revealed the existence of alkaloids, steroids, saponins, tannin-like phenolic compounds and flavonoids via phytochemical analysis.

Coriander is an annual herbal plant of the parsley family. The name coriander denotes the dry fruits and seeds of this plant while the term “cilantro” denotes its delicate young leaves. These plants are native to the Mediterranean region. Gabriella and Dewi (2023) unveiled and identified the potency of *Coriandrum sativum* as repellent for malaria vector control by investigating its bioactive components in order to resolve the toxicity levels posed by N, N-diethyl-3-methylbenzamide (DEET) application. Results from the research identified two bioactive compounds; cyclododecanol and 2-Decenoic acid, capable of performing an inhibition at the specific sites of DEET-AgamOBP-1 binding. The inhibition of AgamOBP-1 prevents mosquitoes from taking blood from the host. The pharmacological relevance of these seed is illustrated in the work of Silva and Domingues, 2017; Sundar *et al.*, 2016; Hosseinzadeh *et al.*, 2016; Duke *et al.* 2002; and Gazwiet *et al.*, 2022. Linalool compound has been identified as the main phytochemical component in Coriander seeds according to the GC-MS analysis carried out by de Figueiredo *et al.* (2004) and High-performance thin-layer chromatography conducted by Jafari and Mori (2021), who also identified a key component, myrcene in the oil of the extract.

Owing to recent drug resistant trend of allopathic drugs and the restricted supply of

new vaccine modules for malaria treatment, this study aimed to determine the phytochemical constituents of methanolic extracts of selected experimental herbal seeds (black seed, fenugreek and coriander seeds), evaluate their curative activity and investigate the extracts' impact on the packed cell volume and weight of study mice.

### Research Objectives:

The objectives of the research are to determine the phytochemical constituents of methanolic extracts of selected experimental herbal seeds (black seed, fenugreek and coriander seeds), evaluate their curative activity and investigate the extracts' impact on the packed cell volume and weight of study mice.

## MATERIALS AND METHODS

### Experimental animals

Sixty (60) albino mice weighing 20-30g bred in the animal house owned by the Department of Pharmacology, Faculty of Basic Medical Science, College of Medicine, University of Port Harcourt, were acclimatized for two (2) days prior to experimental procedures. The *Plasmodium berghei* purchased was sub-passage into three laboratory mice which

served as the stock animals from which the infected inoculum was produced for infecting the experimental mice in each group. The experimental mice were housed in plastic cages containing wood shavings as beddings and covered using a wire gauze. The mice were equally fed using standard grower's mash.

### Collection of plant materials (seeds)

Plant materials (seeds) used for this research was purchased from herbal stores (Jules and Deluxe organics herbal stores) located in Lagos, Nigeria. The seeds purchased were properly packaged, labelled and authenticated by a technologist in the department of pharmacy with total weight at 500g for each sample. The seeds were grounded and soaked in maceration jar containing 2000ml of Methanol (MeOH) solvent and allowed to stand for 72hrs. Continuous stirring was carried out in order to enhance the extraction of active constituents. The soaked seed samples were filtered and concentrated to dryness by mounting the filtrates on water bath regulated at a temperature set at 55°C so as not to denature the filtrate and to enable the residue come out properly for use.



Figure I: Thermostat water bath used for the evaporation of liquid from extract.

### Phytochemical analysis of Plant extracts

Phytochemical analysis and profiling of methanol fractions of *Nigella sativa*, *Trigonella foenum-graecum* and *Coriandrum sativum* seeds were carried out by

implementing the procedures described by Harborne (1973, 1998), and Trease and Evans (2002). Phytochemical screening of the plant seeds was carried out using an analytical technique referred to as Gas Chromatography-Mass Spectrometry (GC-MS), a tool for

detecting volatile and semi-volatile organic molecules, fatty acids, steroids, hormones in liquid, gaseous or solid states, by picking up components of the compound as they separate into varying columns at different times referred to as the compound's retention time. The GC-MS analytical tool was used to determine the bioactive and phytochemical constituent of the extracts by compiling the relative percent amount of each constituent and by comparing the average peak area and also the retention time of each constituent. The identification of the constituents was achieved by determining the retention time/indices and interpretation of data using standard mass spectra presented in the NIST library. This analysis was carried out at Ebic Integrated Services, a laboratory located at Igbo-Etche Road, New Rumuokwurushi, Rivers State, Nigeria.

### Research Design

In line with the objectives of the research, ten animal cages containing six mice each were established. The animals were inoculated with *Plasmodium berghei* (rodent malaria parasite) via intraperitoneal administration in order to establish parasitemia levels for a period of 72hrs (3 days), which was confirmed via microscopy of infected blood samples compared to non-infected samples, before the study mice received doses of plant extract treatment and standard chloroquine drug to assess the curative activity of the treatments. The mice used for the experimental study was categorized as follows: Group 1-3 represents

*Nigella sativa* extract at different doses of 50mg/kg, 100mg/kg and 200mg/kg respectively; Group 4-6 represents *Trigonella foenum-graecum* extract at different doses of 50mg/kg, 100mg/kg and 200mg/kg respectively; Group 7-9 represents *Coriandrum sativum* extract at different doses of 50mg/kg, 100mg/kg and 200mg/kg respectively, while Group 10 represents the standard reference drug Chloroquine considered the positive control group. To determine how many millilitres (ml) of extracts should be given to the mice, 1g of plant extract was dissolved in 10ml of distilled water while considering the average weight of the study mice. Also, 0.25g of chloroquine tablet was dissolved in 10ml of distilled water while considering the weight of the study mice. The liquid formulation was determined at 0.01ml, 0.02ml and 0.05ml for 50mg/kg, 100mg/kg and 200mg/kg of extracts respectively and 0.3ml for standard chloroquine drug used as the positive control. The treatments were fed to the mice through oral administration using a syringe with measured calibration. Samples of blood were drawn from each group, 72hours post inoculation and before treatments in order to determine the initial mean parasitemia level. The treatment doses were administered for 3 days post inoculation of *Plasmodium berghei* parasite and blood samples withdrawn on day 4 and day 7 for analysis of parasitemia level to determine the extent of curative action of extract and standard chloroquine drug according to Rane's curative test procedure.



Figure II: 0.3ml inoculum injected into each mouse.

## Sample and Sampling techniques

The chloroquine sensitive rodent malaria parasite *Plasmodium berghei* used for this study was obtained from the Nigerian Institute of Medical Research, Yaba, Lagos and was maintained by sub-passage into three laboratory mice which served as the donor mice. Sixty (60) albino mice were used to test the curative antimalarial activity of the plant extracts and standard Chloroquine drug. 2 ml of blood was withdrawn from the donor mice and dissolved in 20ml of physiological saline to produce the inoculum. Each study mouse was inoculated with 0.3ml of infected blood (inoculum) containing *Plasmodium berghei* parasitized erythrocytes intraperitoneally. Rane's curative test was used to analyze the schizonticidal activity of the extracts in inoculated mice with established parasitemia as described by Ryley and Peters, 1970. The experimental mice were left for 72 hours to develop malaria infection after inoculation. The plant extracts used for treatments in this study was prepared by dissolving 1.0g of each extract in 10ml of distilled water to required concentrations at 0.01ml, 0.02ml and 0.05ml to achieve the low, middle and high dose (Bahekar and Kale, 2016).

Standard Chloroquine tablets (0.25g) was dissolved in 10ml of distilled water to achieve a dosage of 0.3ml (6mg/kg) administered to the control group for the period of the experiment. The extracts and standard chloroquine drug solution were administered orally to the mice. At the end of the 72 hours post-inoculation, blood samples were collected from each mouse by making a tail snip/incision and using a spreader to make a thin film on the microslide, which was allowed to air-dry before transporting to the laboratory for microscopic viewing (analysis). This sample was considered the basal/initial parasitemia (day 0). The study mice then received treatments for 3 days according to the measurement established for all groups. Blood samples for curative analysis were also collected on day 4 and day 7 post-treatment. The air-dried samples were stained with Giemsa (20%) at the laboratory before viewing under a microscope using the x100 oil immersion objective lens to reveal parasitized erythrocytes out of 500 random clear erythrocytes in fields of the microscope. (CDC.gov; 2016).

$$\text{Percentage parasitemia} = \frac{\text{Number of parasitized cells}}{\text{Total number of red blood cells}} \times 100$$

The mean percentage parasitemia inhibition was calculated using the formula:  $100 - (a/b \times 100)$ .

[Where a represents mean percentage parasitemia on treatment day, while b represents the mean percentage parasitemia on day 0] (Onyegbule *et al.*, 2019).

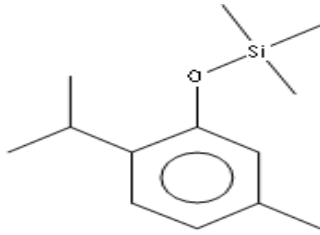
## Packed cell volume and weight analysis

Samples of blood were collected from the experimental animals using a heparinized capillary tube on day 0, 3 and 7 in order to ascertain the packed cell volume of the mice. Three-quarter of the capillary tubes were filled with the blood of the mice and sealed at one end with a sealant. The filled capillary tubes were then placed in numbered slots of the haematocrit centrifuge and allowed to spin at 12,000 revolution per minute (RPM) for 10 minutes, after which the packed cell volume was determined. The effects of the extracts and standard drug on the weight of the experimental mice was also monitored on day 0, 3 and 7 using the Golden-Mettler- U.S.A sensitive weighing balance scale.

## Methods of Data analysis

Descriptive statistical analysis was adopted to analyze the data gotten from the experimental design. Mean and standard deviation were determined using Microsoft excel. Comparison between treated samples were analyzed using single factor analysis of variance (ANOVA) at 95% confidence interval and 5% significance level ( $P < 0.05$ ), followed by Tukey's post-hoc test. Also, error bar charts were used for data presentation and analysis.

## RESULTS



Name: Thymol, TMS derivative

Formula: C<sub>13</sub>H<sub>22</sub>O<sub>Si</sub>

MW: 222 Exact Mass: 222.143991 CAS#: 55012-80-1 NIST#: 108990 ID#: 188652 DB: mainlib

Other DBs: None

Contributor: Philip Morris R&amp;D

InChIKey: UTGMDFONUNJYQP-UHFFFAOYSA-N Non-stereo

10 largest peaks:

207 999 | 73 724 | 222 330 | 208 186 | 96 96 | 45 81 | 74 73 | 223 64 | 95 60  
 | 75 53

Figure III: Thymol constituent identified in the GC-MS analysis of *Nigella sativa*, *Trigonella foenum-graecum* and *Coriandrum sativum*.

Thymol is considered an alkaloid. It is a known antimalarial constituent and is used as a synergetic drug in combination with certain antimalarial drugs. Eg. Chloroquine.

[<https://doi.org/10.57760/sciencedb.14134> full details of the GC-MS analysis of the plant seeds studied.]

**TABLE 1: Curative activity of *Nigella sativa*, *Trigonella foenum-graecum* and *Coriandrum sativum* at all doses and standard Chloroquine drug on Parasitemia level in percentage (%)**

Group	Dose (mg.kg <sup>-1</sup> )	Percentage parasitemia level				
		Day 0 Basal parasitemia	Day 4 post- treatment	Day 7 post- treatment	% Inhibition Day 4	Day 7
<b><i>Nigella sativa</i></b>						
1	50mg/kg	40.6±0.87 <sup>a</sup>	29.2±2.83 <sup>ab</sup>	13.4±0.74 <sup>a</sup>	28.1	67.0
2	100mg/kg	44.9±3.17 <sup>a</sup>	27.4±3.16 <sup>ac</sup>	20.4±1.96 <sup>a</sup>	39.1	54.7
3	200mg/kg	36.6±3.05 <sup>a</sup>	19.2±1.91 <sup>a</sup>	17.9±2.02 <sup>ab</sup>	47.5	51.1
<b><i>Trigonella foenum-graecum</i></b>						
4	50mg/kg	39.2±2.17 <sup>a</sup>	26.8±3.49 <sup>a</sup>	16.96±2.44 <sup>a</sup>	31.6	56.7
5	100mg/kg	43.4±2.76 <sup>a</sup>	35.8±4.52 <sup>a</sup>	18.2±2.88 <sup>a</sup>	17.5	58.1
6	200mg/kg	32.8±4.49 <sup>a</sup>	29.6±6.63 <sup>a</sup>	10.4±2.11 <sup>a</sup>	9.8	68.3
<b><i>Coriandrum sativum</i></b>						
7	50mg/kg	37.8±3.78 <sup>a</sup>	31.2±4.09 <sup>b</sup>	17.7±1.31 <sup>b</sup>	17.5	53.2
8	100mg/kg	42.2±2.93 <sup>a</sup>	25.6±3.24 <sup>b</sup>	15.1±1.18 <sup>b</sup>	39.3	64.2
9	200mg/kg	39.6±4.66 <sup>a</sup>	24.2±1.67 <sup>b</sup>	13.2±1.25 <sup>b</sup>	38.9	66.7
<b>+ve control</b>						
Chloroquine	6mg/kg	43.8±4.60 <sup>a</sup>	12.4±1.22 <sup>ab</sup>	5.85±1.09 <sup>a</sup>	71.7	86.6

Each value is a mean of six replicates expressed as mean ± S.D. Values in the same column with common superscript letters (a, b...) are significantly different at p < 0.05 when compared with one another. Statistical level of significance was determined by one-way Analysis of Variance (ANOVA).



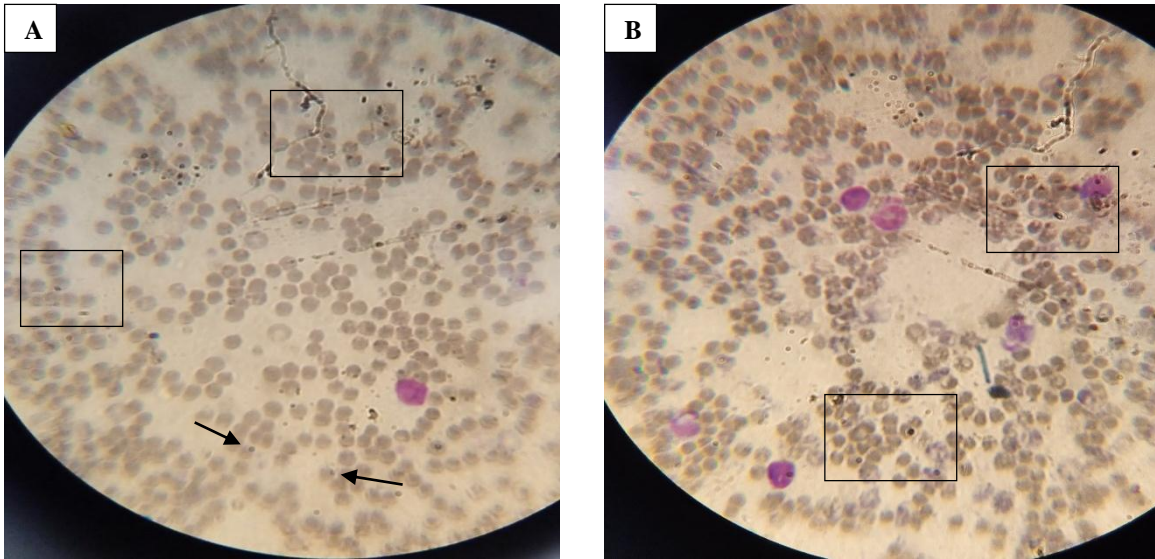


Figure IV: Plate A and B showing peak of parasitemia across groups at day 3 post-inoculation. The areas marked with the square and arrows indicate areas of concentrated parasitemia.

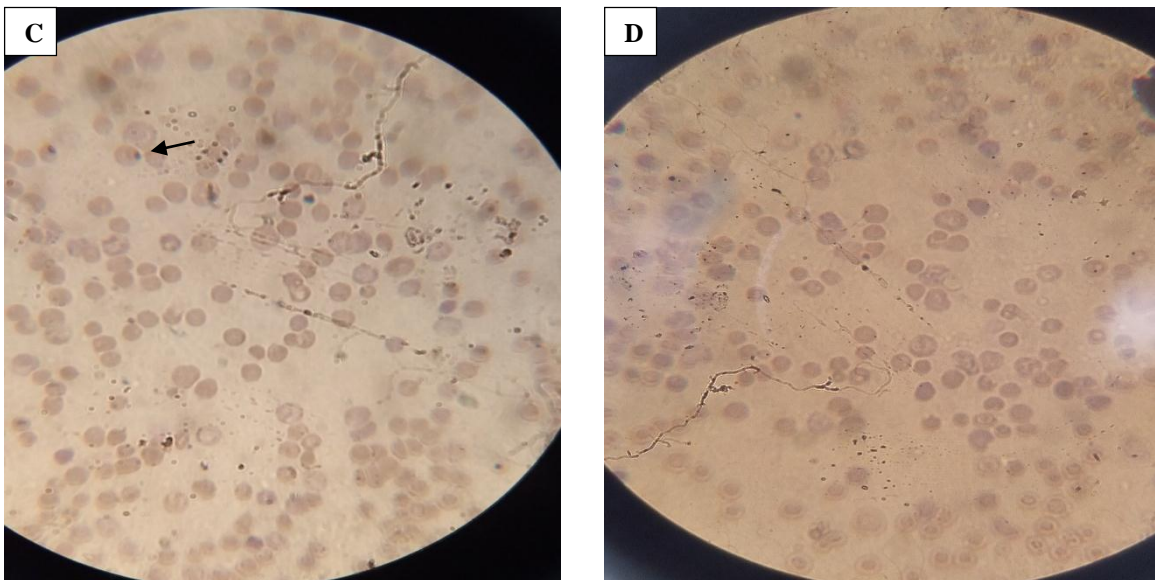


Figure V: Indications of parasitemia but at a lower level as a result of treatment. Plate C is an indication of positive response to treatment from the extracts and D: is a sample from groups treated with chloroquine.

**TABLE 2: Percentage Packed Cell Volume of mice treated with *Nigella sativa*, *Trigonella foenum-graecum* and *Coriandrum sativum* at all doses and Standard Chloroquine drug**

Group	Dose (mg.kg <sup>-1</sup> )	Percentage packed cell volume		
		Day 0	Day 3	Day 7
<i>Nigella sativa</i>				
1	50mg/kg	51.67±1.31 <sup>a</sup>	50.5±0.84 <sup>ab</sup>	38.6±1.59 <sup>a</sup>
2	100mg/kg	48.67±2.33 <sup>a</sup>	49.5±2.13 <sup>a</sup>	43.8±2.85 <sup>ab</sup>
3	200mg/kg	51.0±1.96 <sup>a</sup>	36.0±3.25 <sup>a</sup>	34.6±2.98 <sup>a</sup>

***Trigonella foenum-graecum***

4	50mg/kg	52.33±0.47 <sup>a</sup>	36.0±2.12 <sup>b</sup>	32.3±2.84 <sup>b</sup>
5	100mg/kg	52.0±0.98 <sup>a</sup>	37.0±2.87 <sup>b</sup>	36.5±2.59 <sup>b</sup>
6	200mg/kg	48.67±1.57 <sup>a</sup>	34.6±1.72 <sup>b</sup>	34.2±2.19 <sup>b</sup>

***Coriandrum sativum***

7	50mg/kg	50.5±2.62 <sup>b</sup>	35.5±2.08 <sup>a</sup>	33.0±1.94 <sup>a</sup>
8	100mg/kg	41.33±4.21 <sup>b</sup>	37.25± 3.16 <sup>a</sup>	35.4±3.01 <sup>a</sup>
9	200mg/kg	50.0±1.36 <sup>b</sup>	36.42±2.98 <sup>a</sup>	36.23±3.41 <sup>a</sup>

**+ve control**

Chloroquine	6mg/kg	43.67±1.27 <sup>a</sup>	48.67±2.25 <sup>a</sup>	46.56±2.02 <sup>a</sup>
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Each value is a mean of six replicates expressed as mean ± S.D. Values in the same column with common superscript letters (a, b...) are significantly different at  $p < 0.05$  when compared with one another. Statistical level of significance was determined by one-way Analysis of Variance (ANOVA).

**TABLE 3: Weight of mice treated with *Nigella sativa*, *Trigonella foenum-graecum* and *Coriandrum sativum* at all doses and Standard Chloroquine drug**

Group	Dose(mg.kg-1)	Weight (g)		
		Mice weight (g) on day 0	Mice weight (g) on day 3	Mice weight (g) on day 7
<b><i>Nigella sativa</i></b>				
1	50mg/kg	16.15±0.62 <sup>a</sup>	20.32±0.67 <sup>a</sup>	22.08±2.69 <sup>a</sup>
2	100mg/kg	23.47±2.23 <sup>a</sup>	21.58±2.52 <sup>a</sup>	23.58±2.42 <sup>a</sup>
3	200mg/kg	21.12±0.33 <sup>a</sup>	22.47±3.76 <sup>a</sup>	22.84±2.26 <sup>a</sup>
<b><i>Trigonella foenum-graecum</i></b>				
4	50mg/kg	18.10±0.89 <sup>b</sup>	22.25±1.01 <sup>a</sup>	23.02±1.40 <sup>a</sup>
5	100mg/kg	16.45±0.39 <sup>b</sup>	18.45±2.56 <sup>a</sup>	21.94±1.48 <sup>a</sup>
6	200mg/kg	18.90±0.73 <sup>b</sup>	19.48±0.38 <sup>a</sup>	21.90±0.99 <sup>a</sup>
<b><i>Coriandrum sativum</i></b>				
7	50mg/kg	18.90±1.93 <sup>a</sup>	19.48±2.41	21.90±2.15
8	100mg/kg	24.43±0.58 <sup>a</sup>	23.88±1.35	24.68±1.26
9	200mg/kg	25.82±132 <sup>a</sup>	25.10±0.9	28.18±1.40
<b>+ve control</b>				
Chloroquine	6mg/kg	32.70±3.12 <sup>a</sup>	29.57±2.71 <sup>a</sup>	29.82±2.25 <sup>a</sup>

Each value is a mean of six replicates expressed as mean ± S.D. Values in the same column with common superscript letters (a, b...) are significantly different at  $p < 0.05$  when compared with one another. Statistical level of significance was determined by one-way Analysis of Variance (ANOVA).

## DISCUSSION OF FINDINGS

An identification of active principles in natural drug sources has been established as key to the development of drugs, synthetic and natural forms. This is essentially aided via plant screening to discover therapeutically active components of such plants (Gopalakrishnan and Udayakumar, 2014; Starlin *et al.*, 2019). In this study, rodent model of *Plasmodium* (the causative agent for malaria), *Plasmodium berghei* was used In-vivo for the evaluation of

antimalarial agents of natural and synthetic formulations. The application of these drug formulations In-vivo revealed the effect of the extracts and standard drug.

The results obtained from the study showed significant decrease in mean parasitemia of *Plasmodium berghei* infected mice treated with methanolic extracts of *Nigella sativa*, *Trigonella foenum-graecum* and *Coriandrum sativum*. The results also revealed that high dosages of extracts do not necessarily imply



higher suppressive effects as seen in the inhibitory action of *Nigella sativa* extracts at 50mg/kg compared to the 100mg/kg and 200mg/kg dosages. This result is in agreement with findings from Abdulelah and Zainal-Aidin, 2007; Ashcroft *et al.*, 2018 and Oreagba *et al.*, 2013 who found out that higher doses of extract did not necessarily impose higher suppressive activity compared to lower doses of *Nigella sativa* extract in curative activity. However, Higher dosages of *Trigonella foenum-graecum* at 200mg/kg exhibited greater chemosuppressive inhibitory performance (68.3%) on day 7 compared to dosages at 100mg/kg (58.1%) and 50mg/kg (56.7%) respectively. The same was observed in treatments using methanolic extracts of *Coriandrum sativum*, which indicated higher inhibitory performances at 200mg/kg at 66.7% compared to the 64.2% and 53.2% from those treated with 100mg/kg and 50mg/kg respectively. The curative activities of these extracts have been attributed to the presence of secondary metabolites/compounds such as phenols, alkaloids, saponins, flavonoids and tannins which are considered free radical blockers due to their antioxidant nature and which enables the restriction of oxidative damages caused by *Plasmodium* parasites (Ezenyi *et al.*, 2014). The phytochemical component thymol was identified in all samples of plant extract analyzed using GC-MS technique. Thymol is known for its active antimalarial potentials as seen in the work of Dell'Agli *et al.* (2012) who claimed that thymol-enriched oil fractions of plants had very active antiplasmodial effects on *Plasmodium falciparum* as the main components found in certain plants in Sardinia, a Mediterranean region. Thymol has been synthesized separately and also used as a synergetic drug in combination with antimalarial drugs (chloroquine) for the effective control of malaria parasitemia. Quinoline compounds identified in the analysis are known to block the digestion of haemoglobin in the blood feeding stages of the *Plasmodium* cycle. The synergistic effects of all the identified phytochemicals in each extract summed up to exert thorough

antiplasmodial activity in the treated mice. A reduced level of parasitemia can also be attributed to the consistent administration of the extracts and control drug up until the fourth day of the Rane's curative analytic technique. Although the extracts showed moderate to good measure of curative activity in terms of mean parasitemia reduction, the standard drug chloroquine was by far better indicating a higher chemosuppressive inhibitory performance at 86.6% on day 7 compared to the extracts at all doses. There was no recorded toxicity in the application of extracts at all doses administered. The effectiveness of any substance in the treatment of malaria is based on the likelihood that such extract could exercise some level of specificity and effective clearance of the parasite, without exhibiting any toxicity traces.

Haematological indices are key factors in the analysis of parasitemia effect in malaria pathology (Maina *et al.*, 2010). Investigations carried out indicated an increase in packed cell volume of mice for groups treated with chloroquine as at day 7 compared to those treated with plant extracts, as doses administered were considered insufficient in the reversal of PCV levels. In the work of Kaur *et al.*, 2009, these effects were attributed to the presence of saponin, a bioactive component found in seed extracts which has been associated with the reduction of packed cell volume due to its haemolytic effect. In the work of Bissinger *et al.*, 2014, saponins, a naturally occurring glycoside in plants was associated with haemolysis and as well as triggering erythrocytic death. Asgary *et al.*, 2012 reported that *Nigella sativa* did not have any effects on haematological factors except for the increase of platelets. Contrary to reports from Asgary *et al.*, 2012 and Kaur *et al.*, 2009, the extracts of the plants under study significantly increased packed cell volume in the reports of Ali and Blunden, 2003; Ashcroft *et al.*, 2018; Abubakar *et al.*, 2018 and Metung *et al.*, 2022. These variations may be attributed to the parasitic load inoculated in mice (hyperparasitemia), the parasite species, time of recovery, concentration of extracts and the

presence of saponin as a bioactive compound in all tested extracts, which has been associated with decrease in PCV levels. The reduction in PCV levels is not only peculiar to the plant extracts identified in this research but has also been associated with other plants.

Body weight increase in experimental mice treated with methanolic extracts of *Nigella sativa*, *Trigonella foenum-graecum* and *Coriandrum sativum* were observed at all levels of treatment. Several researchers are in agreement with the research result on the body weight gain potential of *Nigella sativa* when consumed. Mohammed (2007), Abd El-Rahman *et al.*, 2011 and Khattab *et al.*, 2011 reported an increase in body weight performance when *Nigella sativa* is fed to Ruminants (camels, goats and calves). Abdel-Magid *et al.*, 2007 claimed that supplementation of black seeds in animal feed ration helps in promoting the growth performance and feed conversion rate of animals. The increase in body weight of animals fed with supplementation of *Nigella sativa* has been associated with the presence of essential fatty acids that cannot be synthesized such as oleic and linoleic acids. Mice treated with *Trigonella foenum-graecum* extracts also showed increase in body weight gain. This is in agreement with the work of Atta *et al.*, 2012 who reported a significant increase in body weight gain when healthy female Wister albino rats were supplemented with fenugreek seed. The increment of body weight through extract feeding of Coriander aligns with the result established by Naeemasa *et al.*, 2015 and Rahimi *et al.*, 2011. Contrary to the results from the three different extracts at all doses/level, mice given standard chloroquine drug at 6mg/kg, showed a significant decrease in body weight from 32.70g on day 0 to 29.82g on day 7. This can be attributed to weight control measures associated with the use of chloroquine drug and also conditions of low feed intake.

## CONCLUSION

This study provides insight on the relevance of ethnobotanicals/ethnopharmacological plants

as proposed remedies for malaria diseases. Following the trends of resistance in malaria parasites, these plants have had a far-reaching effect as remedial cure for malaria as a result of biochemical components which are considered primary or secondary metabolites commonly referred to as phytochemicals, with potential antimalarial, anti-oxidant and anti-inflammatory properties. Etc. This research was able to identify the antimalarial potentials of black seeds, fenugreek and coriander seed, establish the effect of these plant extracts on the packed cell volume and weight of test animals and also confirm that these plants possess certain compounds "phytochemicals" relevant for malaria treatment.

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