Original Research Article

EFFECT OF *GARCINIA KOLA* ON IVERMECTIN INDUCED TOXICITY IN THE HEMATOLOGY OF TREATED RATS (RATTUS NOVERGICUS)

ABSTRACT

Garcinia kola is prized in African herbal medicine for its diverse uses, including social, economic, and medicinal purposes. A recent study examined the dose-dependent toxicity of ivermectin and the protective effects of *Garcinia kola* extract on the hematological parameters of Wistar rats. 32 Wistar Rats (120 – 150g) were randomly chosen from PAMO University of Medical Sciences' Animal House. They acclimatized for two weeks. Divided into 8 groups (A, B, C, D), each with 4 rats in subgroups 1 and 2. Group A: negative control (0 mg/kg BW of ivermectin, no *Garcinia kola*), subgroup A1. Subgroup A2: positive control (0 mg/kg BW of ivermectin and 20 mg/kg/ml *Garcinia kola*). The other groups (B1, B2, C1, C2, D1, and D2) were treated similarly to the control. Test Subgroups 1 received doses of 10 mg/kg/ml, 20 mg/kg/ml, and 40 mg/kg/ml of Ivermectin intraperitoneally, Subgroups 2 received the same doses followed by oral administration of *Garcinia kola* at 20 mg/kg BW of rat per ml daily, starting 24 hours after toxicity induction. On the eighth day, each rat was anesthetized with chloroform. 5mls of blood was collected via cardiac puncture into anticoagulant bottle for the analysis of the CBC parameters. The results indicated ameliorative effect in some hematological parameters of ivermectin induced group, while showing no significant difference in other parameters. Therefore, it's believed that GK could have ameliorative effect on liver functionality under toxic insults

Keywords : *Garcinia kola, Hematology, Ivermectin-induced toxicity, Wistar rats*

INTRODUCTION

With the shifting of attention from synthetic drugs to natural plant products, plant that were once considered of no value are now investigated, evaluated and developed into drugs [1]. One of such plant is *Garcinia kola*. *Garcinia kola* (G. kola) is commonly known as bitter kola, male kola or false kola; 'Adu' in Esan, 'Miji-goro' in Hausa, 'Akilu or Ugolo' by the Igbos, 'Orogbo' among the Yoruba tribes of Nigeria. It is a medium-sized tree growing up to 12 m tall and 1.5 m wide and usually found in the rain forest of Nigeria [2].

Toxicants whether household, environmental, industrial or biologica have impacted on both the environment and the living components existing in it [3-11].

Ivermectin (IVM) is a macrolide antibiotic produced from a fungus first isolated from a soil sample in Japan, Streptomyces avermitilis. The avermectins are a class of chemicals that have a novel mode of action against nematode and arthropod parasites [12]. All major gastrointestinal and lung nematodes and certain ectoparasites of cattle, sheep, horses and swine, intestinal nematodes, ear mites and sarcoptic mange of dogs, infective- stage heartworm and microfilariae of dogs, and certain intestinal nematodes of chickens are effectively eliminated

by IVM [13], Eight fractions of avermectins contains a-major and b-minor components for the prevention of cellular hyperpolarization [14]. Ivermectin (IVM), doramectin (DME) are the two main types of avermectins [15]. Both drugs are fermentation products of avermectins. IVM is formed from a B1-a, and B1-b (80: 20) mixture of natural avermectin, used in animals and human antiparasitic medication [16]. It is oxidised in the liver primarily and excreted in the faeces and urine causing significant alterations in the liver and kidney tissues [17]. The persistence of avermectins' residues in the tissues and fluids is dose-dependent and producing variable side effects from mild to extremely severe [18,19], up to CNS (central nervous system) depression as a sign of poisoning [20]

Historically, plants have provided a source of inspiration for novel drug compound as plant derived medicines have made a large contributions to human health and well-being. *Garcinia kola* (bitter kola a name sometimes also used for G. afzelii) is a specie of flowering plant belonging to the Mangosteen genus Garcinia, of the family Clusiaceae (also known as Guttiferae). It is found in Benin, Cameroon, The Gambia, Democratic Republic of Congo, Ivory Coast, Mali, Gabon, Ghana, Liberia, Nigeria, Senegal and Sierra Leone. Its natural habitat is subtropical or tropical moist lowland forests [21]. The fruit, seeds ("bitter kola nuts") and bark of the plant have been used for centuries in folk medicine to treat ailments from coughs to fever [22]. According to a report from the Center for International Forestry Research, *Garcinia kola* trade is still importhe indigenous communities and villages in Nigeria [23]. *Garcinia kola* is traditionally used by African folk healers who believe that it has purgative, antiparasitic, and antimicrobial properties [24]. The seeds are used for liver disorders, bronchitis, throat infections, colic, head or chest colds, and cough [24]

More than 900 drugs have been implicated in causing liver injury [25], and it is the most common reason for a drug to be withdrawn from the market, Drug induced liver injury is responsible for 5% of all hospital admissions and 50% of all acute liver failures. There is limited knowledge of the mechanism and effects of *Garcinia kola* on the liver due to sparsity of research around that area and the fact that it is an important traditional nut. Not many works have been reported by scholars on the effect of *Garcinia kola* in health and its ability to hepato-repair. The chemical properties of *Garcinia kola* has been claimed to be effective in the treatment of liver disorders, bronchitis, cough in traditional medicine .The liver metabolic activities was significantly reduced in rats by inducing toxicity with the use of Ivermectin and then compared to the control groups and the treatment with *Garcinia kola* [26]. This will prove beneficial in hematology improvement therapy as well as increasing its metabolic activities.

Less priority has been given to some of the native edibles we take in, as it relates to the effect it has on the livers metabolic activity. Therefore *Garcinia kola* which is an accepted well known masticatory in Nigeria especially in the southern part of the country was chosen because studies have been reported by [27] that *Garcinia kola* has protective effect against variety of experimental hepatotoxins. It is on this note that this research work is designed to establish if ethanoic extract of *Garcinia kola* has any effect on hepatotoxicity of ivermectin. Despite the acclaimed myriad of uses of G. kola seeds in the folklore medicine of Nigeria, the effects of the seed extract on the haematological parameters (Hb, PCV, RBC, MCV, MCH, MCHC, WBC, platelets, neutrophils and lymphocytes) of male rats has not been reported in the open scientific literature. It is a well-known knowledge that *Garcinia kola* has antioxidants properties as a result of its large phytochemical ranges of flavinoids, alkaloids and others. These properties if harnessed properly and utilized as medicaments in cases of hematotoxicity, may go a long way in ameliorating the toxicity of most toxicant (e.g Ivermectin). By evaluating the hematological parameters, as indices of toxicity, the oral administration of *Garcinia kola* will provide a direction to follow during desired repairs of hematotoxicity incidents[19]. The study is aimed at investigating the ameliorative effects of *Garcinia kola* on Ivermectin induced hematologicl toxicity in albino rats (*Rattus Norvegicus*) and the objectives of this study are The hematological parameters such as Complete

Blood Count, was studied, after placing the rats under induced toxicity by singular (I.P) administration of Ivermectin, dose dependently, and subsequently treating them, orally, with daily ethanolic extract of *Garcinia kola* for a period of eight (8) days, to compare the dose-dependency values of such toxic insult on blood function parameters, to compare the effects of *Garcinia kola* on the hematology of controlled group rats, to compare the ameliorative effect *Garcinia kola* (ethanoic extract) may have on the induced toxicity.

MATERIALS AND METHODS

STUDY DESIGN

The design for The Singular, Acute intraperitoneal (I.P) dose-dependent administration of Ivermectin and the subsequent co-administration of *Garcinia kola* (GK), per oral, to the Rats (*Rattus Norvegicus*) is as tabulated in the Table 1 below:

TABLE 1: TREATMENT DESIGN

Treatment Groups:	A		B		C		D	
Subgroups	A1	A2	B1	B2	C1	C2	D1	D2
IVM mg/kg (IP)	0	0	10	10	20	20	40	40
GK mg/kg Per oral(daily)	0	20	0	20	0	20	0	20
Duration(days)	8	8	8	8	8	8	8	8

The animals were randomly assigned to different Control and Test groups (8 animals in each group and divided as 4 animals per subgroup). The experimental groups consist of: (A) Control (no Ivermectin); (B) Ivermectin, 10 mg/kg body weight; (C) Ivermectin, 20 mg/kg body weight and (D) Ivermectin, 40 mg/kg body weight. After two weeks of acclimatization, the treatment started by the animals being singularly administered with 1 ml of different equivalent doses of the toxicant, Ivermectin, intraperitoneally (IP). After 24 hours of intoxication, oral administration of 1 ml of equivalent dose of Garcinia (20mg/kg BW) were given to the rats in subgroups (A2, B2, C2 and D2) daily till the end of the experiment. On the eight day, all the animals were sacrificed using anaesthetic chloroform. 5 mls of blood samples were collected by cardiac puncture [28,29] into anticoagulation bottle with ethylenediaminetetraacetic acid disodium (EDTA). The samples were analysed using the haematological methods of analysis as stated below.

STUDY AREA

The study was conducted in the Animal house of PAMO University of Medical Sciences, Port Harcourt, Rivers State. Nigeria with feeding environment: temperature is 20–26°C, relative humidity is 40 – 70% and animals were under a 12 h light-dark circle

STUDY POPULATION

A total of 32 Male Albino Rats (*Rattus Norvegicus*) weighing between 120 – 150g, was purchased from the Experimental Animal House of PAMO University of Medical Sciences, Port Harcourt, Rivers State. Nigeria.

INCLUSION CRITERIA

Male Albino rats were used, Rats weighing only within 120 - 150g were used, Only healthy rats obtained after two weeks of acclimatization was also used.

EXCLUSION CRITERIA

Female Albino rats were excluded, Rats weighing below 120g or above 150g were excluded, Unhealthy rats were also excluded.

ETHICAL COMMITTEE CONSIDERATION

This research was approved by the Ethics Committee of the University, PAMO University of Medical Sciences with Approval No: PUMS/REC/07.21/VOL.1/0110, after careful scrutiny of the Research design, mode of sampling and disposal of animal carcases

SAMPLE COLLECTION

The doses of Ivermectin (IVM) selected were 0, 10, 20, and 40 mg/kg body weight (BW). The highest dose was close to 3/4 the LD50 (50 – 55 mg/kg B.W.) intraperitoneal dose for rats [28,29]. Suspension of the equivalent dose per weights of rats of Ivermectin in saline was administered by intraperitoneal (I.P) route as a single dose. Biochemical Procedures: Complete Blood Count (CBC); was determined using the Automation method Mindray BC 5300 by [30].

QUALITY ASSURANCE

For accuracy of laboratory results obtained, known controls were used to assay every functional parameter

STATISTICAL ANALYSIS

The data were expressed as mean ± standard deviation, and the significance of the data between groups was tested by SPSS 26.0 software. The different studies undertaken were statistically analyzed by One way Analysis of Variance and Post hoc Tukey- Kramer multiple comparison test [31]. The level significance was set at alpha 0.05.

RESULT

Table 2: Comparisonof HaematologicalParametersamongIvermectinOnly Treated Subgroups

Groups	**HB(g/dl)**	**HCT(%)**	**TWBC**	**RBC**	**MCV**	**MCH**	**MCHC**	**PLT**
			($X10^9$/L)	**($X10^6$/L)**	**(fL)**	**(pg)**	**(g/dL)**	**($X10^3$/l)**
A1	12.9±0.4	44.4±2.7	7.5±2.1	7.0±0.2	63.3±6.3	18.4±1.2	29.1±1.1	873.5±54.6
B1	13.1±0.4	43.8±1.3	7.6±1.3	7.0±0.2	62.3±1.6	18.7±0.5	30.1±0.3	895±187.6
C1	13.7±1.1	47.3±3.6	7.7±4.4	7.4±0.8	63.7±3.8	18.5±0.5	29.0±1.1	829.8±148.8
D1	15.2±0.9	54.2±3.3	10.9±4.8	8.5±0.4	64.0±2.9	18.0±0.6	28.1±1.0	881.8±146.6
F-value	7.48	7.62	0.90	8.66	0.14	0.70	3.05	0.16
P-value	0.004	0.003	0.46	0.002	0.94	0.57	0.07	0.92
Remark	SS	SS	NS	SS	NS	NS	NS	NS
Post-hoc								
A1vsB1	NS	NS	NS	NS	NS	NS	NS	NS
A1vsC1	NS	NS	NS	NS	NS	NS	NS	NS
A1vsD1	SS	SS	NS	SS	NS	NS	NS	NS
B1vsC1	NS	NS	NS	NS	NS	NS	NS	NS
B1vsD1	SS	SS	NS	SS	NS	NS	NS	NS

C1vsD1	NS	NS	NS	NS	NS	NS	NS	NS

NS:Non-significant
SS:Statistically significant

Table 2 compares hematological parameters among subgroups treated with ivermectin only. Significant differences were found in Hb, HCT, RBC, and MCHC levels between some subgroups. However, TWBC, MCV, MCH, and PLT levels showed no significant differences.

Table3:ComparisonofHaematologicalMarkersamong*Garcinia kola*TreatmentSubgroups

Groups	**HB(g/dl)**	**HCT(%)**	**TWBC**	**RBC**	**MCV**	**MCH**	**MCHC**	**PLT**
			($X10^9$/L)	**($X10^6$/L)**	**(fL)**	**(pg)**	**(g/dL)**	**($X10^3$/l)**
A2	12.9±1.1	43±4.2	8.5±3.7	6.7±0.2	64.6±6.4	19.4±1.8	30.1±0.8	926.8±176.7
B2	14.1±1.3	49.3±6.2	4.9±1.8	8.3±0.9	58.0±14.9	17.9±1.0	28.7±1.2	1004±65.9
C2	13.5±0.5	45.3±3.1	7.0±2.6	7.3±0.4	62.0±3.1	18.5±0.4	29.8±1.1	896.3±12.3
D2	12.8±2.1	44±7.3	5±1.6	7.1±1.5	62.7±3.7	18.1±1.0	28.9±0.4	856±80.6
F-value	0.78	1.00	1.82	2.21	0.44	1.41	2.15	1.49
P-value	0.53	0.42	0.20	0.14	0.73	0.29	0.15	0.27
Remark	NS	NS	NS	NS	NS	NS	NS	NS
Post-hoc								
A2vsB2	NS	NS	NS	NS	NS	NS	NS	NS
A2vsC2	NS	NS	NS	NS	NS	NS	NS	NS
A2vsD2	NS	NS	NS	NS	NS	NS	NS	NS
B2vsC2	NS	NS	NS	NS	NS	NS	NS	NS
B2vsD2	NS	NS	NS	NS	NS	NS	NS	NS
C2vsD2	NS	NS	NS	NS	NS	NS	NS	NS

NS:Non-significant
SS:Statistically significant

Table 3 compares hematological parameters among *Garcinia kola* treatment subgroups (A2, B2, C2, and D2). No significant differences were found in Hb, HCT, TWBC, RBC, MCV, MCH, MCHC, and PLT levels between the subgroups. Post hoc analysis confirmed the absence of significant differences in all comparisons ($p > 0.05$).

Table4:ComparisonofHaematologicalParametersbetweeneachSubgroups

Groups	**HB(g/dl)**	**HCT(%)**	**TWBC**	**RBC**	**MCV**	**MCH**	**MCHC**	**PLT**
			($X10^9$/L)	**($X10^6$/L)**	**(fL)**	**(pg)**	**(g/dL)**	**($X10^3$/l)**
A1	12.9±0.4	44.4±2.7	7.5±2.1	7.0±0.2	63.3±6.3	18.4±1.2	29.1±1.1	873.5±54.6
A2	12.9±1.1	43±4.2	8.5±3.7	6.7±0.2	64.6±6.4	19.4±1.8	30.1±0.8	926.8±176.7

B1	13.1±0.4	43.8±1.3	7.6±1.3	7.0±0.2	62.3±1.6	18.7±0.5	30.1±0.3	895±187.6
B2	14.1±1.3	49.3±6.2	4.9±1.8	8.3±0.9	58.0±14.9	17.9±1.0	28.7±1.2	1004±65.9
C1	13.7±1.1	47.3±3.6	7.7±4.4	7.4±0.8	63.7±3.8	18.5±0.5	29.0±1.1	829.8±148.8
C2	13.5±0.5	45.3±3.1	7.0±2.6	7.3±0.4	62.0±3.1	18.5±0.4	29.8±1.1	896.3±12.3
D1	15.2±0.9	54.2±3.3	10.9±4.8	8.5±0.4	64.0±2.9	18.0±0.6	28.1±1.0	881.8±146.6
D2	12.8±2.1	44±7.3	5±1.6	7.1±1.5	62.7±3.7	18.1±1.0	28.9±0.4	856±80.6

a:Statisticallysignificantat$P<0.05$

Table 4 compares hematological parameters within subgroups (A1 vs A2, B1 vs B2, C1 vs C2, and D1 vs D2). No significant differences were observed in Hb, HCT, TWBC, RBC, MCV, MCH, MCHC, and PLT levels between corresponding subgroups. All comparisons yielded p-values greater than 0.05, indicating no significant variations within each subgroup pair.

DISCUSSION

The result of this study in Table 2 explains the comparison of the control and the test subgroupsthatwereadministeredIvermectinindifferentdoses.Acrossthegroups,hemoglobin, hematocrit and red blood cell count showed statistical significance ($P < 0.05$) whereas total white blood cell count, mean cell volume, mean cell hemoglobin, mean corpuscularhemoglobin concentration and platelet showed no statistical significance, the parameters that were of statistical significance also showed increasing values, this could imply, that the dose dependent increase in Ivermectin administration across the group could be responsible for the significantincreaseinthevaluesofhemoglobin,hematocritandredbloodcellcountat$P<0.05$. To understand the particular group that had significant differences, Post-hoc analysis was done, when group A1 was compared to D1, a significant increase in D1(HB, HCT and RBC) was seen in their mean levels, significant increase was also seen in D1(HB, HCT and RBC) when compared to B1. This result points to the fact that there was actually a dose dependent toxicity of ivermectin on the rats across the different groups, following significant increase in the blood erythropoietic functions which can cause hematological fluctuations. Some studies that observed alterations in hematopoietic system parameters may indicate interference with the hematopoietic function of the bone marrow system by exogenous substances [18,32]. Contrarily another study on ivermectin haematotoxicity, reported decrease in hemoglobinlevelsinivermectininducedrats,although therewasinsufficientevidencetoprove that the decline in hematological parameters was due to ivermectin suppression of the hematopoietic system [33].

The present study was designed to scientifically scrutinize the ethno-medicinal benefit of *G. kola*seedinevaluatingitstherapeuticeffectonthewistarrats.Accordingtotheresult,Table3 showed no significant difference in the levels of the haematological parameters studied (HB, HCT, RBC, TWBC, MCH, MCHC, MCV). This could be due to the fact that the concentration of *Garcinia kola*administered was not sufficient enough to effect significant difference as the levelof insultwas increased acrossthe group.A plausible explanationfor this occurrence may be that, the presence of higher amount of *G. kola* would have been used if a proper effect was to be gotten from hematopoietic activities for a short term treatment. In agreement with

this study, another a notable change in erythrocyte values when different concentration of the *Garcinia kola*was administered to fish [34]. The finding on the medicative effect of *Garcinia kola*extract on hematological parameters such as hemoglobin, park cell volume and red blood cells is in disagreement with a different study that reported a reduction in Hb (g/100ml), Hematocrit (%), and erythrocytes (10^6ml) [35].In addition,another study reported that PCV, Hb, and RBC value increased in response to the administration of *Garcinia kola* [36] butthis study reported that *Garcinia kola*hadnohaematological effect ontheratswithinthesevendays ofadministrationacrossthe groups.

As shown by the levels of hematological parameters in Table 4, the comparison between A1 and A2; B1 and B2; C1 and C2; D1 and D2, indicated that there was no ameliorative effect of *Garcinia kola*in the haematological parameters of the rats, as it relates to the effect of induced toxicity by ivermectin. This finding could be due to the fact that after the toxic insult, the red bloodcellsthatwereinjuredwerenotproperlyrepairedbythe*Garcinia kola*asaresultofthe short duration of the treatment thus, the red blood cells were eliminated from the circulation. The result of this experiment also showed that *Garcinia kola*caused no significant increase or decrease in total white blood cell, this result is similar to the findings of other authors [37] where they demonstrated that white blood cell count did not show any significantchangeinvalues.Someliteraturesthatwereindisagreementwiththefindings of this study were basically long term treatment of *Garcinia kola*, where theyreported steady increase in the red blood cell count (RBC), Packed cell volume (PCV), Hemoglobin concentration (Hb), Meancell Hemoglobin concentration (MCHC) [37,38].

Conclusion

The findings from the present investigation showed that induction of ivermectin has a toxic effect on hematological parameters, dose dependently, In addition the result obtained from this work is seen to be in agreement with the implication of *Garcinia kola*seed in traditional medicine and most herbal preparations for treatment of blood disorders, which is also contributing to the screening of its natural antioxidant properties. Therefore, providing persuasive scientific support for the traditional use of *Garcinia kola* in the treatment of blood disorders.

Recommendation

The study performed was an acute study which can be provided as a guide for future chronic studies, therefore further investigations is required especially chronic studies to encompass all the effects of *Garcinia kola*. This studywas an acute study and the duration limitation could not allow the elucidation of both the toxicity effect of ivermectin on the blood and the ameliorative effect of *Garcinia kola*on the hematological toxicityas shown by the parametric findings, therefore it is suggested that further chronic studiesshould be carried out to completely understand the directions the parameters will follow and also histopathological and histochemical studies should also be carried out to understand how blood cellsitselfwillrespondtothetoxicinsultandalsotheremediatingeffectof *Garcinia kola*.

REFERENCES

1. Dada AA, Ikuerowo M. Effects of ethanolic extracts of *Garcinia kola* seeds on growth and haematology of catfish (Clarias gariepinus) broodstock. African Journal of Agricultural Research. 2009 Apr 1;4(4):344-7.
2. Iwu MM. Handbook of African Medicinal plants CRC press. Boca Raton, FL. 1993:183-4.
3. Onwuli D, Ajuru G, Holy B, Fyneface CA. The concentration of lead in periwinkle (*Tympanotonos fuscatus*) and river sediment in Eagle Island River, Port Harcourt, Rivers state, Nigeria. American Journal of Environmental Protection. 2014;2(2):37-40
4. Onengiyeofori I, Fyneface CA. Assessment of Serum Levels of some Heavy Metals in Carpenters Residing in Port Harcourt in Relation to their Lifestyle. Asian Journal of Research in Medical and Pharmaceutical Sciences.2018;4(4), 1-7
5. Fyneface CA, Emeji R, Osere H, Nwisah L. Concentrations of Nickel in Sediment and Periwinkle of Eagle Island River, Port Harcourt. Asian Journal of Fisheries and Aquatic Research, 2018;1(4):1-5
6. Biambo KG., Nyebuchi J, Roseline E, Fyneface AC, Laurretta N. Zinc Composition in Breast Milk of Lactating Mothers in Urban and Sub-urban Areas in Rivers State. Asian Journal of Research in Nursing and Health. 2021;4(4):165-169
7. Catherine I, Biambo KG, Nyebuchi J, Fyneface AC, Goodnews N.Evaluation of Nutrient Composition in Breast Milk of Breast Feeding Mothers in Urban and Sub-urban Subjects in Rivers State. Asian Journal of Pediatric Research. 2021;7(2):39-44
8. Faith D, Biambo GK, Nyebuchi J, Amadi FC, KonneFE. Comparative Study of Heavy Metals in Breast Milk of Breast Feeding Mothers in Urban and Sub-Urban Subjects in Rivers State. Journal of Applied Life Sciences International. 2021;24(8):31-36
9. Okolonkwo BN, Amadi CF, Chimekagbe OE. Organophosphates Toxicity: Pathophysiology, Diagnosis and Treatment. Asian Journal of Biochemistry, Genetics and Molecular Biology. 2022;12(2):27-37
10. Amadi CF, Aleruchi-Didia TN, Konne FE, Nyenke CU. Environmental Effect of Kpo-fire in Niger Delta and Future Health Implications. Biotechnology Journal International. 2022;26(2):18-24
11. Nwisah L, Emeji R, Idoko RA, Odiabara KK, Nyebuchi J, Amadi, CF,….. Samuel BE. Unraveling the Preventive and Therapeutic Roles of Costus Afer in Oxidative Stress Management in Acetaminophen-poisoned Rats. Journal of Biomedicine and Biosensors. 2023;3(4):7-15.
12. Saqib M, Abbas G, Mughal MN. Successful management of ivermectin-induced blindness in an African lion (Panthera leo) by intravenous administration of a lipid emulsion. BMC Veterinary Research. 2015 Dec;11:1-7.
13. Yanuartono Y, Indarjulianto S, Nururrozi A, Raharjo S, Purnamaningsih H. Penggunaan Antiparasit Ivermectin pada Ternak: Antara Manfaat dan Risiko. Jurnal Sain Peternakan Indonesia. 2020 Mar 28;15(1):110-23.
14. Merola VM, Eubig PA. Toxicology of avermectins and milbemycins (macrocylic lactones) and the role of P-glycoprotein in dogs and cats. Veterinary Clinics: Small Animal Practice. 2012 Mar 1;42(2):313-33.
15. El-Saber Batiha G, Alqahtani A, Ilesanmi OB, Saati AA, El-Mleeh A, Hetta HF, Magdy Beshbishy A. Avermectin derivatives, pharmacokinetics, therapeutic and toxic dosages, mechanism of action, and their biological effects. Pharmaceuticals. 2020 Aug 17;13(8):196.
16. Adedara IA, Awogbindin IO, Anamelechi JP, Farombi EO. Garcinia kola seed ameliorates renal, hepatic, and testicular oxidative damage in streptozotocin-induced diabetic rats. Pharmaceutical biology. 2015 May 4;53(5):695-704.
17. Afolabi BT, Agu GC, Onajobi IB. Phytochemical screening and antibacterial activity of Garcinia kola (hackel) and Cola nitida (vent) extracts. Nigerian Journal of Technology. 2020 Jul 16;39(2):379-85.
18. Danaher M, Radeck W, Kolar L, Keegan J, Cerkvenik-Flajs V. Recent developments in

the analysis of avermectin and milbemycin residues in food safety and the environment. Current pharmaceutical biotechnology. 2012 May 1;13(6):936-51.
19. Epstein SE, Hollingsworth SR. Ivermectin□induced blindness treated with intravenous lipid therapy in a dog. Journal of Veterinary Emergency and Critical Care. 2013 Jan;23(1):58-62.
20. Mishra S, Aeri V, Katare DP. Hepatoprotective medication for liver injury. Journal of Pharmacy and Pharmaceutical Sciences. 2014 Mar 10;3:891-932.
21. Omeh YN, Onoja SO, Ezeja MI, Uchendu WC, Okorie E, Raymond M. Quantitative phytochemical, proximate analysis and hypolipidemic effect of Garcinia kola. British Journal of Medicine and Medical Research. 2014 Dec 21;4(36):5770.
22. Buba CI, Okhale SE, Muazzam I. Garcinia kola: The phytochemistry, pharmacology and therapeutic applications. Int J Pharmacognosy. 2016;3(2):67-81.
23. Etim II, Etukudoh NS, Olumide OB, Uchejeso OM, Lucy NL, Bwotle FY. Hypoglycemic and hypolipidemic effect of bitter kola (Garcinia kola) seed extract on alloxan-induced diabetic albino rats. Journal of Biosciences and Medicines. 2020 Jun 10;8(06):127.
24. Ozioma EO, Chinwe OA. Herbal medicines in African traditional medicine. Herbal medicine. 2019 Jan 30;10:191-214.
25. Onasanwo SA, Rotu RA. Antinociceptive and anti-inflammatory potentials of kolaviron: mechanisms of action. Journal of Basic and Clinical Physiology and Pharmacology. 2016 Jun 1;27(4):363-70.
26. Arndts K, Specht S, Debrah AY, Tamarozzi F, Klarmann Schulz U, Mand S, Batsa L, Kwarteng A, Taylor M, Adjei O, Martin C. Immunoepidemiological profiling of onchocerciasis patients reveals associations with microfilaria loads and ivermectin intake on both individual and community levels. PLoS neglected tropical diseases. 2014 Feb 20;8(2):e2679.
27. Panche AN, Diwan AD, Chandra SR. Flavonoids: an overview. Journal of nutritional science. 2016;5:e47.
28. Okolonkwo BN, Amadi CF, Chikwubike UO. Nyenke CU. The Comparative Effect of Vitamin E + C on the Chronic Toxicity of Paraquat in Albino Rats (Rattus norvegicus). European Journal of Medicinal Plants. 2022;33(6):7-13
29. Okolonkwo BN, Chukwubike UO, Amadi CF, Nyenke CU. Therapeutic Effects of Vitamin E on Paraquat Induced Liver Toxicity in Male Albino Rats (Rattus Norvegicus). Journal of Advances in Medicine and Medical Research. 2022;34(17):1-7
30. Nyebuchi J, Adjekuko CO, Okur AM, Ken-Ezihuo SU, Jumbo MI. Evaluation of the effect of working hours on haematological parameters among cement loaders in Port Harcourt. Asian Journal of Biochemistry, Genetics and Molecular Biology. 2022;12(1):48-54
31. Lowry DB, Modliszewski JL, Wright KM, Wu CA, Willis JH. The strength and genetic basis of reproductive isolating barriers in flowering plants. Philosophical Transactions of the Royal Society B: Biological Sciences. 2008 Sep 27;363(1506):3009-21.
32. Adeoye GO, Alimba CG, Oyeleke OB. The genotoxicity and systemic toxicity of a pharmaceutical effluent in Wistar rats may involve oxidative stress induction. Toxicology Reports. 2015 Jan 1;2:1265-72.
33. Nwozo SO, Osunmadewa DA, Oyinloye BE. Anti-fatty liver effects of oils from Zingiber officinale and Curcuma longa on ethanol-induced fatty liver in rats. Journal of Integrative Medicine. 2014 Jan 1;12(1):59-65.
34. Awad E, Awaad A. Role of medicinal plants on growth performance and immune status in fish. Fish & shellfish immunology. 2017 Aug 1;67:40-54.
35. Rao GH. Non-traditional approaches to diagnosis and management of type-2

diabetes mellitus: Point of view. J Diabetes Metab. 2015 Jan 1;6(489):2.
36. Ahumobe AA, Braide VB. Effect of gavage treatment with pulverised Garcinia kola seeds on erythrocyte membrane integrity and selected haematological indices in male albino wistar rats. Nigerian Journal of Physiological Sciences. 2009;24(1).
37. Alabi Q, Akomolafe R, Olukiran O, Nafiu A, Omole J, Adefisayo A, Oladele A. Assessment of haematological and biochemical effects of kolaviron in male Wistar rats. British Journal of Pharmaceutical Research. 2017 Jan 10;16(3):1-4.
38. Atsukwei D, Daniel E, Odeh S, Adams M, Malgwi I, Olih P. Efficacy of Garcinia kola seed ethanolic extract on haematological parameters in male Wistar rats. Journal of Advances in Medical and Pharmaceutical Sciences. 2015 Jan 10;3(1):10-22