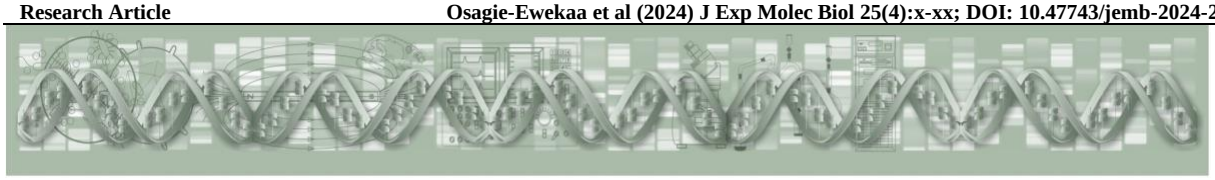

PROPHYLACTIC ANTIHYPERTENSIVE EFFECT OF EXTRACT OF SIMAROUBA GLAUCA ON SALT-LOAD INDUCED HYPERTENSION IN NORMOTENSIVE MALE WISTAR RAT

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Abstract

Simarouba glauca has been reported to demonstrate a wide range of medicinal properties; including folklore management of hypertension disorder. The current study focused on the application of aqueous leaf extract of *Simarouba glauca* (AESG) as a potential prophylactic anti-hypertensive agent in male *Wistar* rats, following salt-load induced hypertension. A total of 15 experimental adult male *Wistar* rats weighing between 184 and 244 g were used for the study. The rats were allotted into five (5) groups of 25, 50, and 100 $mgkg^{-1}$ body weight AESG; group that received 8 % NaCl for one week to induce hypertension; replaced with 0.9 % NaCl daily in drinking water for 4 weeks; the normotensive group, received food and water only *ad libitum*. Body weights and relevant hemodynamics were obtained weekly for four weeks, using the non-invasive (tail-cuff) MRBP system according to the method described by Bunag and Butterfield. Biochemical evaluation and histopathology investigation were conducted on blood plasma and relevant tissues respectively after 4 weeks according to previously established and reported methods; data were analyzed with *GraphPad* Prism, version 9 and presented as mean ± Standard Deviation. The results indicated that salt-load elicited significant weight loss; elevated hemodynamics; particularly, systolic and diastolic blood pressures; altered relevant biochemical indicators of hepatic and renal functions. Inversely, groups pre-treated with respective dose of AESG exponentially gained weight, significantly prevented alterations of hemodynamics and mitigated relevant biochemical indicators and pathological changes in relevant organs. Pre-treatment with AESG; particularly at 50 $mgkg^{-1}$, remarkably demonstrated significant anti-hypertensive potential.

Keywords: Medicinal Plants, Blood Pressure, Antihypertensive, *Simarouba glauca*

Introduction

Hypertension is a disorder of public health importance characterized by elevated systolic blood pressure of ≥ 140 mmHg and a diastolic Blood Pressure of ≥ 90 mmHg after repeated visits to a qualified health practitioner. The World Health Organization has estimated that high blood pressure causes one in every eight deaths; reported that approximately seven hundred million people are currently living with untreated hypertension; making hypertension the third leading cause of mortality in the world. The health Organization further stated that the number of people

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living with hypertension has doubled to 1.28 billion since 1990 as at the time of the report (Campbell *et al*., 2022). Globally, there are one billion hypertensive people; four million people die annually as a direct result of complications associated with hypertension (Campbell *et al*., 2022).

Hypertension is categorized in two types, primary or essential (90-95 % of hypertensive cases) and secondary (5-10 %) hypertension. Although, the specific cause of primary hypertension is yet to be linked to any known or diagnosed related cause; is yet to be explicated. However, pathophysiological contributors such as stress, dysregulation of the sympathetic nervous system, over-stimulation of vasoconstrictors, dysregulation of the Renin-Angiotensin-Aldosterone system (RAAS); resulting to excessive sodium ions retention; decreased production of vasodilators such as prostacyclin, nitric oxide (NO), and natriuretic peptides; resulting from endothelial dysfunction, oxidative stress-induced vascular remodeling; obesity; and diabetes mellitus, atherosclerosis and amongst others are speculated to be responsible for the development of essential hypertension; whereas, secondary hypertension has been strongly reported to be associated with known underlying factors such as apnea, drug-induced, neurological, and/or endocrine dysfunction (such as aldosteronism) (National Clinical Guideline Centre (UK), 2011). Secondary hypertension develops less rapidly, when compared with primary or essential hypertension. It has been reported that non-essential or secondary hypertension has been linked to identifiable; modifiable causes; when not properly managed can provoke cardiovascular complications; multi-organ dysfunction and failure such as peripheral arterial diseases, cardiac dysfunction and cardiomegaly, renal failure, retinal hypertension; amongst others (British Heart Foundation, 2015).

Blumenthal *et al*. (2015) also reported that lifestyle modifications, such as exercises, weight reduction, Dietary Approach to Stop Hypertension (DASH); which includes low-salt diets have demonstrated a 70 % prospect to lowering blood pressure, enhance antihypertensive drug efficacy and decrease risk of cardiovascular complications.

The reports of Campbell *et al*. (2022) and Lovibond *et al*. (2011), have equally revealed that the cost of managing hypertension with allopathic medicine such as angiotensin II receptor antagonists, alpha blockers, angiotensin converting enzyme inhibitors, Beta blockers, calcium channel blockers, diuretics, direct rennin inhibitors, vasodilators has raised concern for decades. This is coupled with comorbidities and the adverse effects such as muscle cramps, dizziness, extreme tiredness, dehydration, blurred vision, abnormal heart rate, skin rash, cough, vomiting, kidney failure, fever, sore throat, diarrhea, fatigue, headache, diarrhea, constipation, edema, swollen ankle, weakness, depression, hallucinations, insomnia, impotence, palpitation and a host of others associated with the application of antihypertensive drugs (Park *et al*., 2017; Lovibond *et al*., 2011).

The recent cross-sectional studies reported by Zhao *et al*. (2020); Lorbeer *et al*. (2017) and Qian *et al*. (2016) have implicated the Non-Alcoholic Steatohepatitis as an independent risk factor to the onset of hypertension and related cardiovascular complications; as well as demonstrated clinical evidence that hypertension may promote the onset of the Non-Alcoholic Fatty Liver Disease (NAFLD) and the progression of liver injury; amongst others. Studies have equally reportedly suggested that NAFLD may promote hypertension by initiating systemic inflammation, capable of triggering triglyceride and lipid intermediate accumulation in the liver that may result to hepatocellular lipotoxicity and (or) hepatocyte damage (Zhao *et al*., 2020; Lorbeer *et al*., 2017; Qian *et al*., 2016).

The study of Landazuri *et al*. (2017) reported the application of numerous pharmacological agents of medicinal plants origin. The study revealed that inherent biological compound(s) in medicine

plants have prospects in management and possible treatment of hypertension. According to the 1977 World Health Assembly (WHA) report, approximately 70-80% of the world's population rely on non-conventional medicine mainly from herbal sources for primary health care, particularly in the developing countries where the cost of consulting orthodox medical practitioners and the price of antihypertensives are beyond the reach of a majority of the affected category; hence, the experimental application of potential bioactive compounds inherent in *Simarouba glauca* leaf. Considering the cost implication of managing hypertension and related complications; the adverse effects of allopathic agents; the paucity of data on the cardiovascular potential effect of *Simarouba glauca* (*S. glauca*), the study seek to evaluate the potential anti-hypertensive efficacy of *S. glauca* on experimental salt-load induced hypertensive of male *Wistar* rat hitherto pre-treated with respective dose of *Simarouba glauca* aqueous leaf extract; as well as the pathophysiological impact of induced-hypertension on liver metabolism and the electrolyte homeostatic function of the kidney.

Patil and Gaikwad (2011) reported that *Simarouba glauca*, commonly known as "Paradise tree" or "Laxmitaru" belongs to the family Simaroubaceae. *S. glauca* has a long history of herbal medicine applications, considering its numerous pharmacological properties. Joshi and Joshi (2002), equally reported that the bioactive chemicals present in leaf, fruit, pulp and seed of *S. glauca* have demonstrated analgesic, antimicrobial, antiviral, astringent, emmenagogue, stomachic, tonic, vermifuge properties. The major active groups of phytochemicals in *S. glauca* are the quassinoids, which belong to the triterpene chemical family. Ailanthinone, glaucarubinone and holacanthone are considered as some of the main active quassinoids available in genus, *Simarouba*. Other chemicals include benzoquinone, canthin, dehydroglaucarubinone, glaucarubine, glaucarubolone, melianone, simaroubidin, simarolide, simaroubin, simarubolide, sitosterol and tirucalla (Technical Data Report, 2002).

Materials & Methods

Collection of *S. glauca* leaves and Aqueous Extraction of Compounds

Fresh leaves of *S. glauca* was harvested from *Cercobela* Farms® located at Ubiaza, Esan South East Local Government Area of Edo State. Nigeria. A fresh plant specimen was deposited at the Department of Plant Biology and Biotechnology Herbarium, University of Benin, Benin City. Nigeria and authenticated with a voucher N0. UBHs382. The leaves were rinsed with distilled water and air-dried at room temperature at the Department of Biochemistry, University of Benin, for twenty-eight (28) days. According to the extraction method previously described by Osagie-Eweka *et al*. (2016), the leaves were pulverized and sieved at the Department of Pharmacognosy, Faculty of Pharmacy, University of Benin, to obtain fine a fine powder. A 500 g leaf powder was soaked in 2.5 L of distilled water and stirred at intervals for 24 h, and filtered. The extract plant material was re-macerated in another portion of 2.5 L of distilled water and stirred at intervals for another 24 h. Both filtrate portions were pooled and freeze-dried at the Department of Biochemistry, Adekunle Ajasin University, Akungba-Akoko, Ondo State to obtain a fine powdered aqueous leaf extract (AESG); with a yield of 6 % w/w extraction. The extract was stored in a sterile bottle in the refrigerator at 18^{o}C until required for analyses.

Plate 1.1. *S. glauca* plant in its natural habitat (Cercobela Farms®, Ubiaja), Osagie-Eweka photo library (2014)

Experimental Animals

A total of 15 experimental adult male *Wistar* rats weighing between 184 and 244 g were used for the study. The animals were housed in metabolic cages; fed and were maintained under laboratory conditions of 12 h light/ 12 h dark cycle and were acclimatized for two weeks prior to commencement of studies. *Wistar* rats in this study were handled in accordance with the international, natural and institution guidelines for care and use of laboratory animals in Biochemical and Biomedical research as promulgated by the Canadian Council of Animal Care (1984). The study protocols on animal use were approved by the Faculty of Pharmacy, University of Benin Ethics Committee with reference number EC/FP/021/11.

Hypertension induction

Hypertension was induced in accordance with the method described by Simchon *et al*. (1991). Eight (8) % NaCl solution was prepared and administered to test rats with the calibrated (150 mL) drinking bottles. Drinking bottles were washed daily and refilled with 8 % NaCl for one (1) week, after which rats were placed on Normal saline (0.9 % NaCl) for the duration of the study to sustain the attained hypertensive state in test rats.

Administration of AESG & Antihypertensive Prophylactic Study

The experiment rats were orally administered AESG as prescribed in the OECD (2010), No. 425 test guidelines; as described by Oliveira *et al*. (2016) and Rout *et al*. (2014). The rats were randomly allotted into five (5) groups of n = 3. Test rats received doses of 25, 50, and 100 $mgkg^{-1}$ body weight respectively of AESG, the untreated group received 8 % to induce hypertension and subsequently received 0.9 % NaCl in drinking water daily for 4 weeks to sustain hypertensive state; while the control group received only water *ad libitum*. The initial body weights of rats were obtained before commencement of the study; weekly for 4 weeks. Baseline systolic (SBP), diastolic Blood Pressures (DBP), mean arterial pressure (MAP) and heart rate (HR) were obtained using the non-invasive (tail-cuff) blood pressure measurement system (MRBP, IITC LIFE SCIENCE). Experimental rats were pre-administered AESG 25, 50 & 100 mg/kg body weight respectively for one week, after which experimental rats were exposed to 8 % NaCl load (induced hypertension) (Simchon *et al*., 1991) for one week, as treatment with extract continued. After one week, 8 % NaCl was withdrawn and replaced with 0.9 % NaCl to sustain hypertension for 4 weeks.

Collection of data and Samples/Specimen

Test rats in each group were weighed weekly; change in body weight was recorded. SBP, DBP, MAP and HR of rats exposed to 8 % NaCl (Induced Hypertension) and (or) already hypertensive rats were measured weekly and recorded. At the end of the *in vivo* experimental study; on the 30th day, final hemodynamic variables were obtained; rats were fasted overnight. The following day, rats were anesthetized with 1.5 g/kg intraperitoneal injection of urethane according to a method previously described by Bilanda *et al*. (2019). A 5 ml sample of venous blood was

withdrawn into heparinized specimen bottles. The blood samples were centrifuged at 3,500 rpm for 10 min to obtain plasma samples which were stored at 18 ℃ and used for biochemical analyses within a few days. The liver and kidneys were harvested from each rat, cleared off connective tissues and fixed in formal saline (0.9 g of NaCl salt in 90 mL of distilled water, mixed with10 mL of 40 % formalin to obtain a final volume of 100 mL) for histological evaluation. The fixed-excised organs were trimmed into 5 mm thickness; dehydrated with graded concentrations of ethanol (70, 95 & 99 %, absolute ethanol); cleared in xylene and embedded in paraffin wax. The embedded tissues were further sectioned into 6 µm thickness, stained with haematoxylin and Eosin (H & E); examined under the light microscope; according to the methods described by Windsor (1994) and Gurr (1959). The sections were photographed at a magnification of x400 with the Vanox-T Olympus photographic microscope.

Blood Pressure Measurement

Principle

Blood pressure measurements were obtained weekly according to the principle of Nikolai Korotkov; method described by Bunag and Butterfield (1982). Each rat was subjected to acclimatization in the restrainer for 1-2h/day for one (1) week before the commencement of blood pressure measurement. This was essential to reduce movement artifact, improve pulse and produce desired recordings/electronic traces

Biochemical analyses

Alanine Amino Transferase (ALT) was measured by monitoring the concentration of pyruvate hydrazone formed with 2, 4-dinitrophenylhydrazine, Aspartate Amino Transferase (AST) was measured by monitoring the concentration of oxaloacetate hydrazone formed with 2, 4-dinitrophenylhydrazine, Alkaline Phosphatase (ALP) was measured by monitoring the concentration of P-nitrophenol formed when P-nitrophenylphosphate is hydrolyzed by ALP in the presence of H_2O, Gamma Glutamyl Transferase (γGT) was measured by monitoring the absorbance values of 5-amino-2-nitrobenzoate produced by the reaction of L-γ-glutamyl-3-carboxy-4-nitroanilide and glycylglycine in the presence of γGT in the specimen/sample, Lactate was measured in the plasma sample; according to the methods described by Reitman and Frankel, Englehardt *et al.*; Tietz; Deutsche Geselleschaft fur Klinische Chemie respectively using commercial test kits (Randox® Laboratories, United Kingdom) (Reitman and Frankel, 1957; Englehardt, 1970; Teitz, 1987; Weisshaar, 1975). The Total Proteins, Albumin and Globulins were estimated according to the methods described by Tietz, Doumas *et al.* and calculated by subtraction of albumin from the total proteins respectively using commercial test kits (Randox® Laboratories, United Kingdom) (Tietz, 1995; Doumas *et al*., 1971). Lipid profile tests which include total cholesterol (TC), high density lipoprotein cholesterol (HDL-C), triglyceride (TG) and low-density lipoprotein cholesterol (LDL-C) were done using colorimetric methods and calculative method described by Roeschlau *et al.*; Jacobs and Van Denmark; Friedewald *et al.* respectively using commercial test kits (Randox® Laboratories, United Kingdom) (Roeschlau *et al.* 1974; Jacob and VanDenmark, 1960; Friedewald *et al*., 1972); the cardiac risk ratio was calculated using relevant lipid profile indices by dividing the total cholesterol value by the HDL-C value. The Total Bilirubin and Conjugated (Direct Bilirubin) were estimated according to a colorimetric method described by Jendrassik and Grof (1938); whereas, the unconjugated (indirect) bilirubin was calculated by subtracting the value of the conjugated bilirubin from the total bilirubin value, using commercial test kits (Randox® Laboratories, United Kingdom). The plasma sodium, chloride ions were estimated according to the methods described by Maruna (1958); Tietz *et al.* (1986), respectively using commercial test kits (Randox® Laboratories, United Kingdom). The plasma Urea and

Creatinine were estimated according to the methods described by Weatherburn (1967) and Bartels and Bohmer (1972).

Statistical Analysis

Data are expressed as mean ± SD (standard deviation). Differences between means of test groups were evaluated by one-way analysis of variance (ANOVA) followed by Tukey's multiple comparison test. Differences were considered significant at $P < 0.05$. All statistical analyses were conducted using GraphPad Prism®, version 9.

Results & Discussions

Results

Prophylactic Effect of varying dose of AESG on Body Weight (g) of Male *Wistar* Rats Exposed to 8 % NaCl, Normal Saline (N.S) for 4 weeks

The data presented in figure 1 indicate significant reduction in body weights of experimental rats pre-treated with varying dose of Aqueous Leaf Extract of *S. glauca* (AESG); including the untreated Hypertensive group at week 2, upon exposure to 8 % NaCl to induce hypertension. Within week 2-4, there was observed reversal of weight loss; significant increases in body weights of experimental rats following continuous administration of varying dose of AESG; including untreated hypertensive group, although there was observed weight loss, upon replacement of the 8 % NaCl with 0.9 % NaCl to sustain desired hypertensive state. Statistical analysis of the data obtained for weight of rats indicate significant mean weight gain at 11.43 ± 1.35, 16.45 ± 0.98, 11.60 ± 0.57 % of rats administered 25, 50 & 100 $mgkg^{-1}$ body weight of AESG respectively after 4 weeks against a significant loss in mean body weight of -6.72 ± 0.01 % of untreated hypertensive group vis-à-vis the normotensive group which equally gained 18.00 ± 0.07 % mean body weight after 4 weeks. The group pre-treated with AESG 50 $mgkg^{-1}$ demonstrated the highest gain in percentage body weight when compared with other pre-treated groups vis-à-vis the untreated hypertensive group. The group pre-treated with AESG 50 $mgkg^{-1}$ competed favorably with the normotensive group.

Prophylactic Effect of varying doses of AESG on Hemodynamics Indices of Male *Wistar* Rats Exposed to 8 % NaCl, Normal Saline (N.S) for 4 weeks.

The data presented in figure 2 indicate significant ($P \leq 0.05$) exponential increases in systolic blood pressure (SBP) of untreated hypertensive group through week 1 (123.500 ± 2.8 mmHg), week 2 (142.59 ± 1.9 mmHg), week 3 (147.17 ± 7.5 mmHg); particularly at week 4 (148.58 ± 2.5 mmHg); when compared to the Normotensive group with SBP through week1 (126.67 ± 1.2 mmHg), week 2 (127.38 ± 5.6 mmHg), week 3 (131.0 ± 6.7 mmHg) and week 4 (130.33 ± 3.9 mmHg). Accordingly, the data presented in figure 2 further revealed groups pre-treated with 25, 50 or 100 $mgkg^{-1}$ AESG significantly prevented systolic hypertension through week 2 to 4, when compared with untreated hypertensive group. The Diastolic Blood Pressure (DBP) and the Mean Arterial Pressure (MAP) data presented in figures 3 and 4 respectively show that 25 and 50 mg/kg AESG demonstrated significant DBP and MAP lowering effect; compared favorably with the normotensive group; particularly at week 2 and 3 respectively; when compared to the untreated hypertensive group's week 2 and 3.

Remarkably, pre-treatment with respective doses of AESG (25, 50 and 100 $mgkg^{-1}$) significantly prevented SBP elevations at week 2 of 8 % NaCl high salt load; when compared with the untreated hypertensive group. Notably, AESG 50 $mgkg^{-1}$ demonstrated most effective feature; which significantly ($P \leq 0.05$) prevented elevation of SBP at week 2 (127.75 ± 7.5 mmHg); further

significantly ($P \leq 0.05$) reversed SBP elevation at week 3 (115.08 ± 5.6 mmHg), when compared to AESG 25 & 100 $mgkg^{-1}$; the untreated hypertensive group respectively. The data presented in fig. 5 indicate obviously that there was no significant difference ($P \geq 0.05$) in the Heart Rates (HRs) of groups pre-treated with respective dose of AESG, when compared with the HR of untreated hypertensive group through week 2 - 4.

Furthermore, the data presented in figures 6-10 illustrate the SBP, DBP, MAP and HR recorded polygraph charts of normotensive, and untreated hypertensive groups vis-à-vis groups pre-treated with 25, 50 or 100 $mgkg^{-1}$ AESG.

Prophylactic Effect of varying Dose of AESG on Liver Function of Male Wistar Rats exposed to 8 % NaCl, Normal Saline for 4 weeks

The data presented in figure 11 indicate that induced hypertension (8 % NaCl load) elicited significant ($P \leq 0.05$) elevation in plasma Alanine transaminase (ALT) activity; followed by significant ($P \leq 0.05$) reduction in plasma Aspartate transaminase (AST) and ɣ-glutamyl transferase (GGT) activities respectively; whereas, the plasma Alkaline Phosphatase (ALP) and Lactate dehydrogenase (LDH) activities appeared non-significantly ($P \geq 0.05$) affected by the induced hypertension; when compared with the enzyme activities of the normotensive group. Consequently, the group pre-treated with AESG 50 $mgkg^{-1}$ significantly ($P \leq 0.05$) prevented elevated plasma ALT activity; when compared with the enzyme activity of the untreated hypertensive group; whereas, the groups pre-treated with AESG 25 & 100 $mgkg^{-1}$ respectively exhibited observed elevated ($P \leq 0.05$) plasma ALT activities; when compared with the enzyme activity of the untreated hypertensive group.

The groups pre-treated with AESG 50 & 100 $mgkg^{-1}$ respectively significantly ($P \leq 0.05$) prevented and further reduced plasma AST activity; when compared with the plasma AST activity of untreated hypertensive group. Neither the plasma ALP activity of the untreated hypertensive group nor the groups pre-treated with varying dose of AESG were altered by induced hypertension (8 % NaCl load); whereas, the plasma LDH activity of the groups pre-treated with 25 and 50 $mgkg^{-1}$ respectively was significantly ($P \leq 0.05$) elevated; when compared with the plasma LDH activity of the untreated hypertensive group. The data presented in figure 12 illustrate the liver synthesizing functional ability; which featured plasma Total Proteins, Albumin and Globulins. The data highlight significant ($P \leq 0.05$) reductions in plasma total proteins and globulins concentrations; significant ($P \leq 0.05$) elevation in plasma Albumin concentrations of untreated hypertensive group; when compared with the normotensive group. Strikingly, it was observed that the groups pre-treated with AESG 25 & 50 $mgkg^{-1}$ respectively significantly ($P \leq 0.05$) attempted to prevent the lowering of plasma total proteins and globulins concentrations respectively, when compared with the untreated hypertensive group; whereas, the group pre-treated with AESG 100 $mgkg^{-1}$ demonstrated a non-significant ($P \geq 0.05$) and significant ($P \leq 0.05$) effects at preventing the lowering of plasma total proteins and plasma globulin concentrations respectively; when compared with the untreated hypertensive group. The group pre-treated with varying dose of AESG (25, 50 and 100 $mgkg^{-1}$) demonstrated significant ($P \leq 0.05$) effect at preventing and further lowing the plasma albumin concentrations; when compared with the untreated hypertensive group.

Prophylactic effect of varying Doses of AESG on Plasma Lipid Profile & Cardiac Risk Ratio of Male *Wistar* Rats Exposed to 8 % NaCl, Normal Saline for 4 Weeks.

The data presented in figure 13 indicate that the untreated hypertensive group featured significant ($P \leq 0.05$) elevations in plasma Total Cholesterol (TC), High-Density Lipoprotein Cholesterol (HDL-C), Low-Density Lipoprotein Cholesterol (LDL-C), Triglycerides (TG) concentrations; a non-significant ($P \geq 0.05$) increase in the plasma Cardiac Risk Ratio (CRR), when compared with the of the normotensive group. Pre-treatment with varying dose of AESG; particularly, the 25 & 50 $mgkg^{-1}$ significantly ($P \leq 0.05$) prevented and further reversed elevations in plasma TC, LDL-

C and TG concentrations respectively, when compared with the untreated hypertensive group; with a corresponding significant ($P \leq 0.05$) elevation in the HDL-C concentrations of the groups pre-treated with AESG 25 & 50 $mgkg^{-1}$; when compared with the untreated hypertensive group.
Whereas, the group pre-treated with AESG 100 $mgkg^{-1}$; however, demonstrated poor prophylactic effect; evidenced by significant ($P \leq 0.05$) elevations in TC, LDL-C; a significant ($P \leq 0.05$) reduction in HDL-C concentrations when compared with the untreated hypertensive group. Furthermore, the group pre-treated with AESG 100 $mgkg^{-1}$ demonstrated poor prophylactic effect, although significantly ($P \leq 0.05$) prevented further elevations in plasma TG concentrations when compared to the untreated hypertensive group.
The plasma CRR level of the untreated hypertensive group was quite elevated ($P \geq 0.05$); when compared with the plasma CRR level of the normotensive group. Impressively, groups pre-treated with AESG 25 and 100 $mgkg^{-1}$ prevented CRR; characterized by significantly ($P \leq 0.05$) lower CRR levels; when compared with the untreated hypertensive. Whereas, the group pre-treated with AESG 100 $mgkg^{-1}$ featured elevated CRR levels, when compared with the untreated hypertensive group.

Prophylactic Effect of varying Dose of AESG on Plasma Total Bilirubin, Conjugated & Unconjugated Bilirubin of Male Wistar Rats Exposed to 8 % NaCl, Normal Saline for 4 weeks

The data presented in figure 14 indicate that the untreated hypertensive group demonstrated significantly ($P \leq 0.05$) elevated plasma conjugated bilirubin; markedly ($P \leq 0.05$) lowered plasma Total Bilirubin and unconjugated bilirubin concentrations; when compared with the normotensive group. However, pre-treatment with varying dose of AESG (25, 50 and 100 $mgkg^{-1}$) significantly ($P \leq 0.05$) prevented elevated plasma conjugated bilirubin levels; prevented and further significantly ($P \leq 0.05$) reduced plasma total bilirubin and unconjugated bilirubin levels respectively; when compared to the untreated hypertensive group.

Prophylactic Effect of varying Dose of AESG on Plasma Sodium & Chloride ions of Male Wistar Rats Exposed to 8 % NaCl, Normal Saline for 4 weeks

The data presented in figure 15 indicate that induced hypertension, that is the untreated hypertensive group demonstrated significant ($P \leq 0.05$) elevation and reduction in plasma sodium and chloride ions concentrations respectively; when compared with the normotensive group. Furthermore, it was observed that pre-treatment with varying dose of AESG (25, 50 and 10 $mgkg^{-1}$) did not prevent elevations in plasma sodium ion concentration. That is, there were significant ($P \leq 0.05$) elevations of plasma sodium ions concentrations in groups pre-treated with AESG 25, 50 and 100 $mgkg^{-1}$ respectively; when compared with the sodium ion concentration of untreated hypertensive group. Likewise, groups pre-treated with AESG 25 and 50 $mgkg^{-1}$ respectively demonstrated corresponding increases ($P \leq 0.05$) in plasma chloride ion concentrations, a non-significant corresponding increase in chloride ion concentration of group pre-treated with AESG 100 $mgkg^{-1}$; when compared with the chloride ion concentration of the untreated group.

Prophylactic Effect of varying Dose of AESG on Plasma Urea & Creatinine levels of Male Wistar Rats Exposed to 8 % NaCl, Normal Saline for 4 weeks

The data presented in figure 16 feature significant ($P \leq 0.05$) elevation and reduction of plasma Urea and Creatinine concentrations respectively in untreated hypertensive group, when compared with the normotensive group. Whereas, pre-treatment with respective doses of AESG (25, 50 and 100 $mgkg^{-1}$) significantly ($P \leq 0.05$) prevented elevated plasma urea concentration; when compared with the urea concentration of untreated hypertensive group. Similarly, groups pre-treated with AESG 25 and 50 $mgkg^{-1}$ respectively, significantly ($P \leq 0.05$) prevented and further

lowered the creatinine concentrations, when compared with the creatinine level of the untreated hypertensive group. Contrariwise, the group pre-treated with AESG 100 $mgkg^{-1}$ demonstrated significant ($P \leq 0.05$) rise in creatinine level; when compared with the creatinine level of the untreated hypertensive group.

Discussion

Hypertension, being a condition characterized by asymptomatic cardiovascular complications; is often not unconnected with comorbidities, such as obesity, diabetes *mellitus*, likely compromised liver functioning, cardiac disorder, renal dysfunction; amongst others; perhaps arising from underlying conditions. Hence, the study equally examined relevant biochemical indicators; taking under consideration, the preliminary investigative study on blood pressure lowering potential of the aqueous leaf extract of *Simarouba glauca* (Osagie-Eweka *et al*., 2023).

The study adopted the salt-sensitive hypertensive model, characterized by salt-load induced Na^+ and water retention (Rodriguez-Iturbe *et al*., 2007; Lifton *et al*., 2001; Guyton, 1992); consequently, resulted to elevated systolic (SBP) and diastolic blood pressure (DBP); accompanied with alterations in mean arterial pressure (MAP) and heart rates (HR) of experimental animals. Although, scientific review has reported contrary findings, that salt-resistant subject may not often present with alterations in relevant hemodynamic indicators due to their extra ability to pass out high salt load from system circulation, when compared to the salt-sensitive subject (Guyton, 1990). The study highlighted the remarkable impact of Na^+-load induced hypertension on body weight of experimental models. The outcome of the study revealed significant weight loss in the untreated hypertensive group. While the data of the present study is at variance with considered epidemiological and experimental studies respectively (Guyton, 1990; Iida *et al*., 2019; Guo *et al*., 2017), which indicated that high salt-induced hypertension did not elicit hypertension-related weight loss. Conversely, pre-treatment with respective doses of AESG; particularly at 50 $mgkg^{-1}$ remarkably prevented hypertension induced weight loss, associated with high salt load on experimental models. The present study indicates that prophylactic management of salt-load induced hypertension strongly correlated with geometric increase in the body weight of experimental models administered respective doses of AESG. In other words, it demonstrated convincing findings that pre-treatment with AESG strongly mitigated high salt-load induced weight loss in experimental models.

According to the World Health Organization Joint News Report in Geneva, Switzerland, a subject is said to be hypertensive, after several ambulatory blood pressure measurement of relevant hemodynamics, of elevated SBP of ≥ 140 mmHg and DBP of ≥ 90 mmHg (Campbell *et al*., 2022). In the study being reported, experimental animal exposed to 8 % NaCl load demonstrated marked elevation in SBP and DBP of 142.59 ± 1.9 mmHg and 91.79 ± 6.8 mmHg respectively, which obviously indicated that salt-load elicited desired hypertension (see figures 2, 3 & 7 (MRBP Polygraph)). The elevated hemodynamics were sustained in the experimental models by withdrawal of 8 % NaCl and prompt replacement with 0.9 % NaCl through week 2-4. The outcome of the salt-load, which resulted in hypertension in the present study is consistent with the reports of Rodriguez-Iturbe *et al*. (2007), Lifton *et al*. (2001) and Guyton (1992) perhaps suggestively attributed to high salt-load sensitivity of experimental animals. Furthermore, it is imperative to state that 8 % salt load, resulted to enlarged coronary artery of the heart, as presented in micrograph figure 19b; which is consistent with a prominent feature associated with hypertension-induced vascular alterations. Interestingly, the experimental groups pre-treated with respective doses of AESG; particularly, the AESG 25 and 100 $mgkg^{-1}$ respectively demonstrated remarkable reduction

in systolic, diastolic and mean arterial blood pressures when compared to the untreated hypertensive model. Studies have reportedly implicated the potential antihypertensive effect of biologically active phytocompounds in experimental models; coupled with natriuretic agonistic effect. A similar study equally reported the potent anti-hypertensive properties of triterpenoids isolated from the *reishi* or *lingzhi* mushroom of the *G. lucidum* (Onyema-iloh1 *et al*., 2018; Boh *et al*., 2007). Alkaloid (Aloperine) from plant source has been reported to demonstrate a vasorelaxant effect on isolated smooth muscle tissue indicating a possible hypotensive agent (Yang *et al*., 2018). Likewise, the Aloperine isolated from *S. flavescens* root strongly indicated KCNQ5 potassium channel activation; which demonstrated both voltage-dependent and voltage-independent; vascular-expressed KCNQ5 opening; subsequently reversed blood volume/pressure (Manville *et al*., 2019). Previous study reported from our laboratory has equally revealed the abundant presence of bioactive phytomedicinal compounds in *S. glauca* (Osagie-Eweka *et al*., 2016); supported by the report of Gurupriya *et al*. (2017). The data of the present study clearly indicate that AESG possess potential antihypertensive effect; perhaps attributed to inherent phytocompounds such as alkaloid(s), as previously reported (Gurupriya *et al*., 2017).

The data presented in figure 11 indicate that untreated hypertensive group demonstrated elevated plasma alanine aminotransferase (ALT); with apparently normal levels of plasma aspartate aminotransferase (AST), Alkaline Phosphatase (ALP), ɣ-glutamyl aminotransferase (GGT) and Lactate dehydrogenase (LDH); likewise, the data presented in figure 12 show that plasma total proteins, albumin and globulin levels of untreated hypertensive group appeared equally unaltered. Studies conducted by Ikewuchi (2013) and Ayalogu *et al*. (2011) reported significant elevations in biomarkers of liver enzymes; particularly, the plasma ALT in salt load induced hypertensive models. Considering that other relevant indicator enzymes of liver function, such as GGT and others were not altered in the present study; including the total proteins, albumin and globulins; there was no obvious, significant pathological liver damage capable of compromising its integrity and function. It's most likely and strongly adduced that the observed elevated plasma ALT in untreated hypertensive group and other groups pre-treated with respective dose of AESG, may have resulted from inflammatory hepatic stenosis; arising from high salt load-induced hypertension, as evidenced in micrograph figures 17b-e. Additionally, It is important to note that pe-treatment with respective dose of AESG did not demonstrate significant hepato-protective effect from high salt-load induced inflammatory stenosis. In a similar evaluation of the pathological and biochemical conditions associated with high-salt activated hypertension, it is observed in the data presented in figure 14 that untreated hypertensive group presented with hyper-conjugated bilirubin, which obviously indicate that salt load elicited inflammatory stenosis; may have resulted in constricted bile excretion, leading to accumulated conjugated bilirubin in the blood. Previous studies conducted by Ikewuchi (2013) and Ayalogu *et al*. (2011) reported indicate that high salt-induced hypertension elicited markedly elevated serum conjugated bilirubin; which agrees with the data of the present study. The observed inflammation may have equally resulted to complications linked with symptoms of steatohepatitis. Interestingly, respective dose of AESG effectively prevented elevation of plasma conjugated bilirubin, characterized by inflamed stenosis; suspected to have been evoked by high salt-induced hypertension.

The data presented in figure 13 indicate marked elevation of plasma total cholesterol, low density lipoprotein-Cholesterol and triglycerides; including increased cardiac risk ratio in untreated hypertensive group. The outcome of the present study clearly demonstrated that high-salt load was associated with mechanisms capable of eliciting disturbances in cholesterol homeostasis in hypertensive state; considering the role of the liver in cholesterol metabolism and the observed

inflammatory stenosis presented in micrographs figures 17b-e, there are convincing indications that elevated cholesterols may not be unconnected with the state of the liver. Prevention of elevated levels of cholesterols; particularly, the triglycerides is key to the management of hypertension related cardiovascular complications such as vascular hardening and arteriosclerosis. The review report of Nordestgaard and Varbo, underscores the role of triglycerides in cardiovascular diseases. The review further implicated triglycerides in hypertensive condition pointing that "the cholesterol content of triglyceride-rich lipoprotein (remnant cholesterol) is more likely to be the cause of atherosclerosis and cardiovascular disease rather than raised triglycerides" (Nordestgaard and Varbo, 2014). The review reported by Zhao *et al*. (2020), highlighted the pathophysiological contribution of Non-Alcoholic Fatty Liver disorder (NAFLD) to the progression of hypertension related cardiovascular conditions. The review also emphasized the possibility of hypertension induced NAFLD-linked to steatohepatitis; resulting to symptomatic biochemical alterations and complications associated with multisystemic adverse effects (Zhao *et al*., 2020). The data of the present study agrees with the report of the review published by Zhao *et al*., with respect to the possibility of hypertension-elicited steatohepatitis (Zhao *et al*., 2020). However, the outcome of the present study revealed that pre-treatment at 25 & 50 $mgkg^{-1}$ significantly prevented elevations in total cholesterol, low-density lipoprotein cholesterols and triglycerides; with marked reduction in cardiac risk ratio at 25 & 50 $mgkg^{-1}$ respectively. The marked reduction in plasma lipoprotein cholesterols and triglycerides; low cardiac risk observed, may not be unconnected with the extract's ability to potentially mitigate the predisposing effect of dyslipidemia associated with the etiology of hypertension and cardiovascular diseases at low doses. Although, previous study in our laboratory reported that oral administration of aqueous leaf extract at higher doses of 500, 1000 and 2000 $mgkg^{-1}$ respectively elicited hyperlipidemia (Osagie-Eweka *et al*., 2021). However; at lower doses in the present study, respective doses of AESG significantly mitigated hypertension-induced dyslipidemia. Review and several studies on the roles of bioactive compounds of medicinal plants origin, have reported evidence of anti-hyperlipidemic effect; the regulatory potential of these biologically active compounds on lipid metabolism; particularly at the level of cholesterol synthesis and re-uptake at the liver and enterocytes respectively (Patti *et al*., 2018; Gil-Ramírez *et al*., 2016; Hu *et al*., 2016; Derosa *et al*., 2013; Cicero *et al*., 2007). The review report of Ji *et al*. (2019) highlighted the possible regulatory mechanisms capable of preventing hypercholesterolemia and the potential anti-dyslipidemic activities of numerous bioactive compounds of medicinal plants origin; ranging from Curcumin, a polyphenol isolated from *Curcuma longa Linn* which inhibits cholesterol absorption through its binding to the Niemann-Pick C1 like1 (NPC1L1-related transporter) (a transmembrane protein which facilitates the transfer of cholesterol in diets and bile from the lumen into the brush border of the membrane of the enterocytes) (Feng *et al*., 2017; Feng *et al*., 2010; Altmann *et al*., 2004). Free cholesterol (FC) is esterified to cholesterol esters (CEs) by acyl CoA: cholesterol acyltransferase-2 (ACAT)-2. ACAT2 can catalyze the formation of cholesteryl ester in intestinal epithelial cells [61]. Triterpenic acid (oleanolic acid (OA) and ursolic acid (UA)) isolated from hawthorn; berberine (extracted from Coptis chinensis) are responsible for the cholesterol-lowering effect of these plants by inhibiting intestinal ACAT activity (Wang *et al*., 2014; Lin *et al*., 2011). Inhibition of microsomal triglyceride transfer protein (MTTP) leads to decreased apolipoprotein B (APOB) secretion and chylomicron assemblage; a number of in vitro studies have demonstrated that several flavonoids, such as taxifolin, quercetin, naringenin and tangeretin have Microsomal Triglyceride Transfer Protein (MTTP) inhibitory activities Casaschi *et al*., 2004; Kurowska *et al*., 2004; Casaschi *et al*., 2002; Wilcox *et al*., 2001). The review of Ji *et al*. (2019) further highlighted numerous herbal medicinal plants with bioactive compounds such alkaloids (berberine), saponins (ginsenoside),

polyphenols (pomegranate) and flavonoids (taxifolin, quercetin), that have significantly demonstrated anti-hypercholesterolemic and anti-dyslipidemic properties and potentials in attenuating atherosclerosis-related cardiovascular complications. Interestingly, previous study conducted in our laboratory reported the significant presence of numerous phytocompounds in the leaf extracts of *S. glauca*; some of which have been implicated in cholesterol homeostasis and lipid regulation (Osagie-Eweka *et al*., 2016). Therefore, the data presented in figure 13 clearly indicate that respective doses of AESG demonstrated significant lipid and lipoprotein cholesterol-lowering activity; which may likely be attributed to the availability of relevant phytocompounds.

Electrolyte homeostasis and fluid volume is tightly regulated by the kidneys; strictly monitored under hypertensive condition. The overall assessment of the optimum functionality of the kidneys is directly proportional to the amount of sodium and water balance either excreted or reabsorbed. The role of the kidneys in regulation of blood volume; by extension, blood pressure can't be over emphasized. The increased reabsorption of sodium by the kidneys is accompanied by corresponding retention of water which increases blood volume or blood pressure. The homeostatic regulatory mechanism of the kidneys equally maintains required levels of chloride ions and other relevant ions necessary to regulate electrolyte-water balance. The measurement of creatinine concentration in blood or plasma is a functional assessment of the integrity and the filtration capacity of the glomerulus (Glomerular filtration rate, GFR); the experimental and clinical significance of elevated concentration of measured creatinine in plasma or serum is indicative of poor filtration capacity or compromised renal function. Urea is a non-protein nitrogenous waste (NPN) obtained from deaminated amino acids of breakdown of proteins; which is available as ammonia, converted to urea by the liver enzymes activities; excreted by the kidneys. Therefore, characteristically elevated levels of urea in blood equally indicates suspected compromised renal functioning associated with poor glomerular filtration capacity (Salazar, 2014). In the present study, the data presented in figure 15 reveals marked elevations in plasma sodium ions in untreated hypertensive group; including groups pre-treated with respective doses of AESG. However, the chloride ion concentrations indicate non-significant alterations in untreated hypertensive group and groups pre-treated with respective doses of AESG. The elevated plasma sodium ion is attributed to salt-load induced hypertension on experimental models; leading to increased sodium absorption and water retention; whereas, it appeared there was a corresponding efflux of chloride ions that resulted to non-significant elevations in plasma chloride ions. The measured plasma creatinine levels of untreated hypertensive and other groups pre-treated with respective doses of AESG where not elevated, which indicate that salt-load induced hypertension did not impact on the kidney's ability to effectively clear creatinine from the blood; whereas, there was observed elevated urea concentration in the untreated hypertensive group; which indicate that the kidney's urea excretory function was impaired; perhaps attributed to inflammation observed in the histological evaluation of the renal tubules. Interestingly, groups pre-treated with respective doses of AESG demonstrated reno-protective anti-inflammatory responses; effectively attempted to prevented elevated plasma urea concentrations.

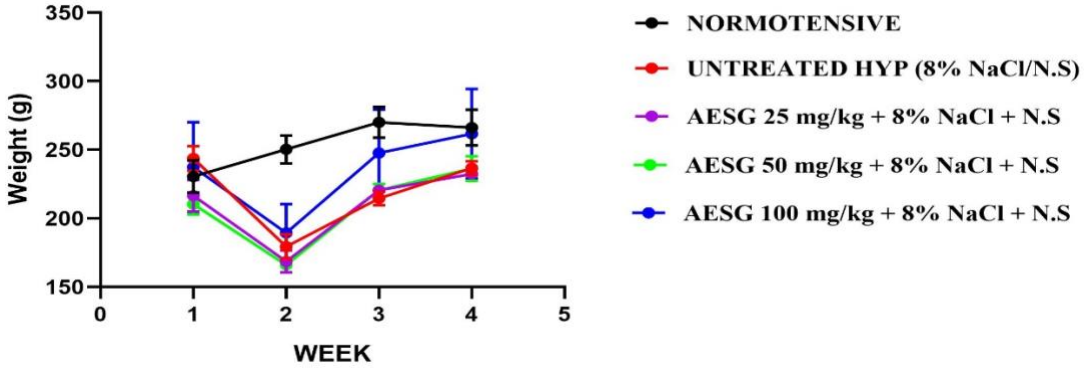


Figure 1. Prophylactic effect of varying doses of AESG on Mean Body Weight (g) of Male *Wistar* Rats Exposed to 8 % NaCl, Normal Saline (N.S) for 4 weeks against Normotensive & Hypertensive groups respectively. Data are Mean ± SD (n ≥ 3). Data are Mean ± SD (n ≥ 3)

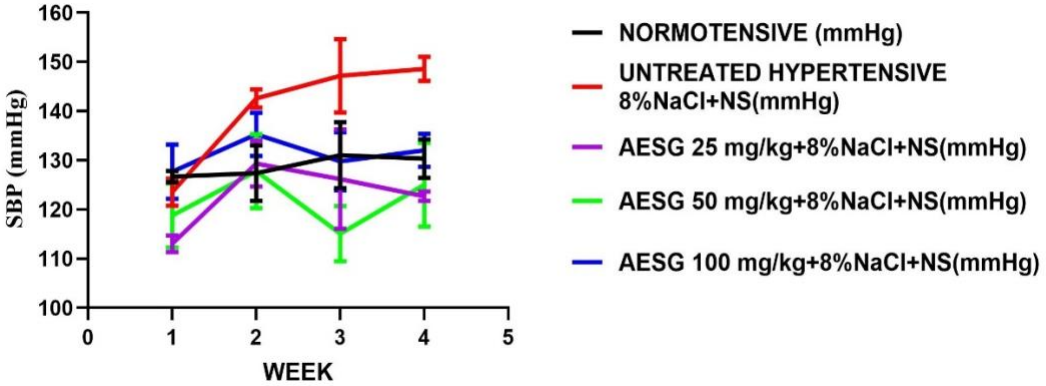


Figure 2. Prophylactic effect of varying doses of AESG on Mean SBP (mmHg) of Male *Wistar* Rats Exposed to 8 % NaCl, Normal Saline (N.S) for 4 weeks against Normotensive & Hypertensive groups respectively. Data are Mean ± SD (n ≥ 3). Data are Mean ± SD (n ≥ 3)

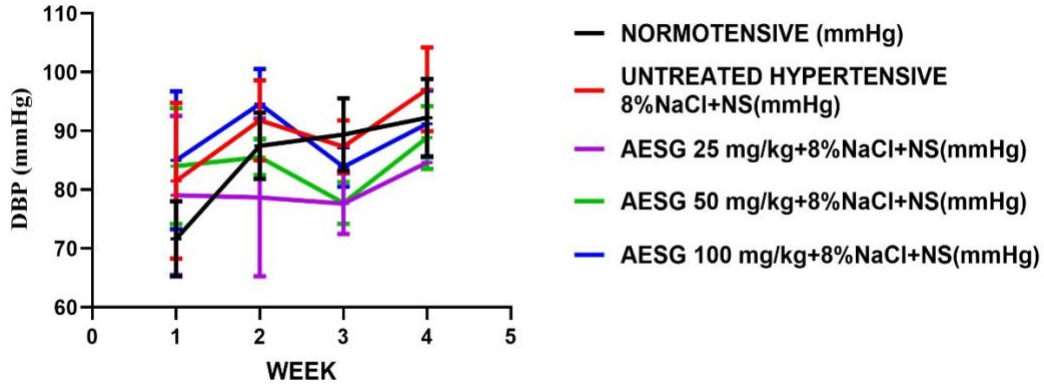


Figure 3. Prophylactic effect of varying doses of AESG on Mean DBP (mmHg) of Male *Wistar* Rats Exposed to 8 % NaCl, Normal Saline (N.S) for 4 weeks against Normotensive & Hypertensive groups respectively. Data are Mean ± SD (n ≥ 3). Data are Mean ± SD (n ≥ 3)

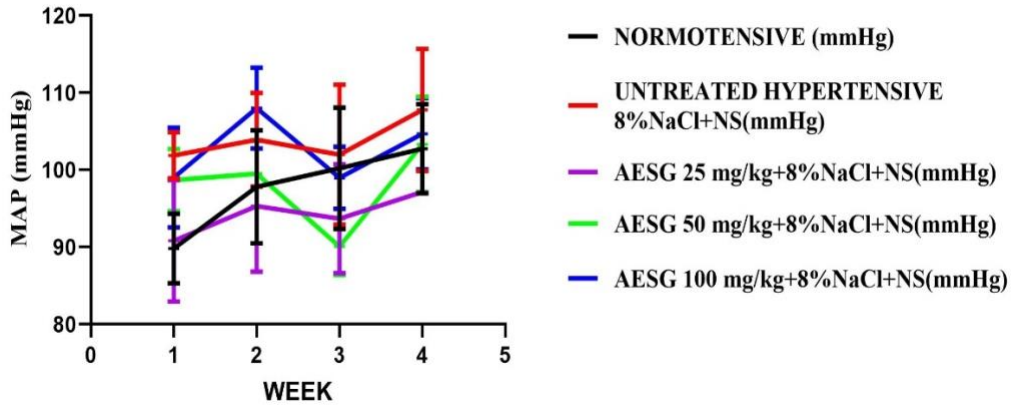


Figure 4. Prophylactic effect of varying doses of AESG on MAP (mmHg) of Male *Wistar* Rats Exposed to 8 % NaCl, Normal Saline (N.S) for 4 weeks against Normotensive & Hypertensive groups respectively. Data are Mean ± SD (n ≥ 3). Data are Mean ± SD (n ≥ 3)

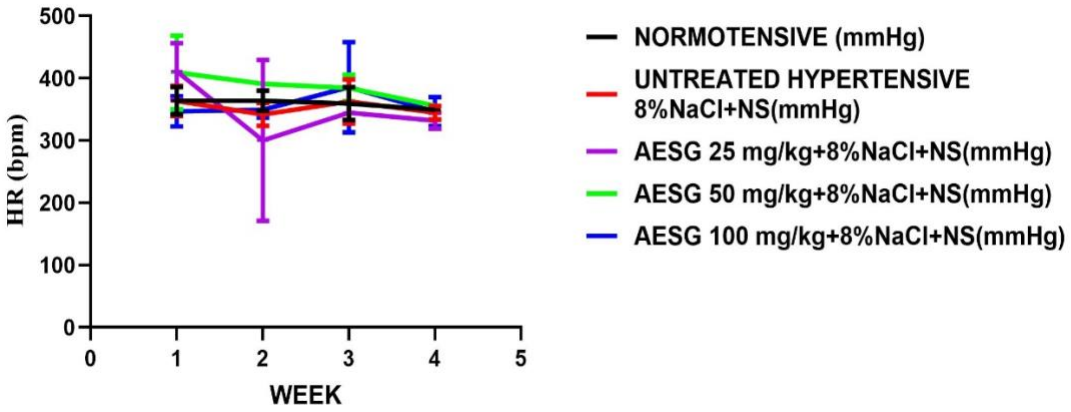


Figure 5. Prophylactic effect of varying doses of AESG on Mean Heart Rate (HR) (bpm) of Male *Wistar* Rats Exposed to 8 % NaCl, Normal Saline (N.S) for 4 weeks against Normotensive & Hypertensive groups respectively. Data are Mean ± SD (n ≥ 3). Data are Mean ± SD (n ≥ 3)

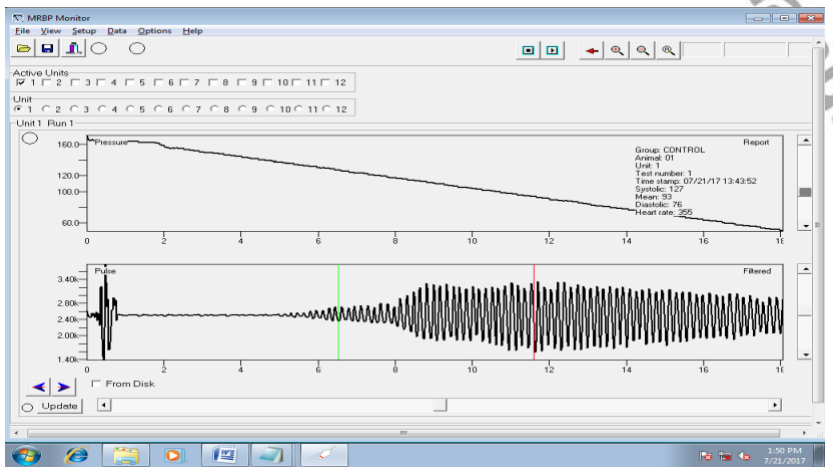


Figure 6. MRBP Record polygraph chart of Normotensive *Wistar* Rat

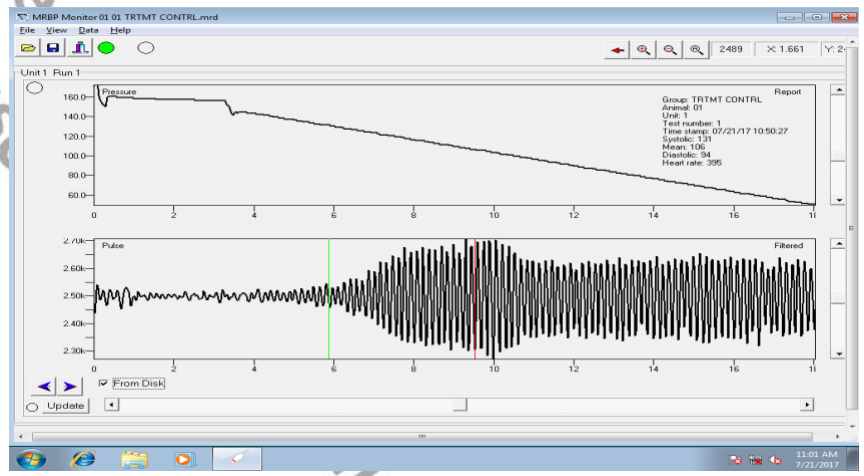


Figure 7. MRBP Record polygraph chart of untreated Hypertensive *Wistar* Rat

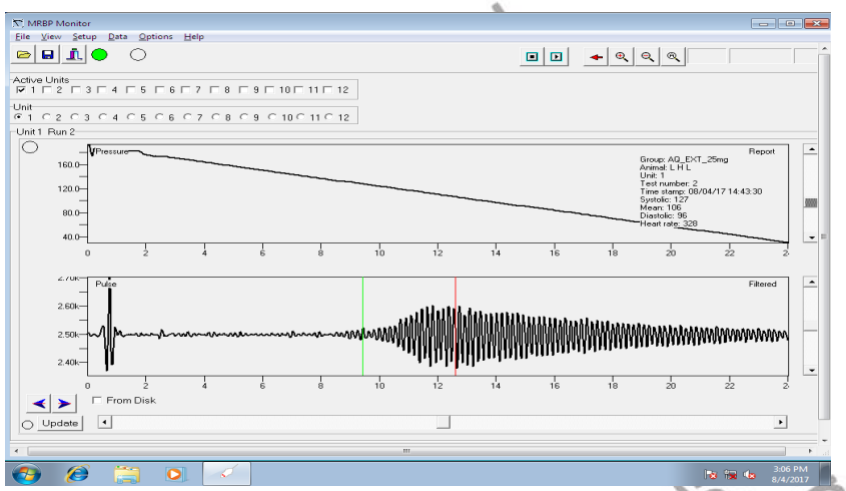


Figure 8. MRBP Polygraph chart of *Wistar* Rat Administered AESG 25 mgkg^{-1}

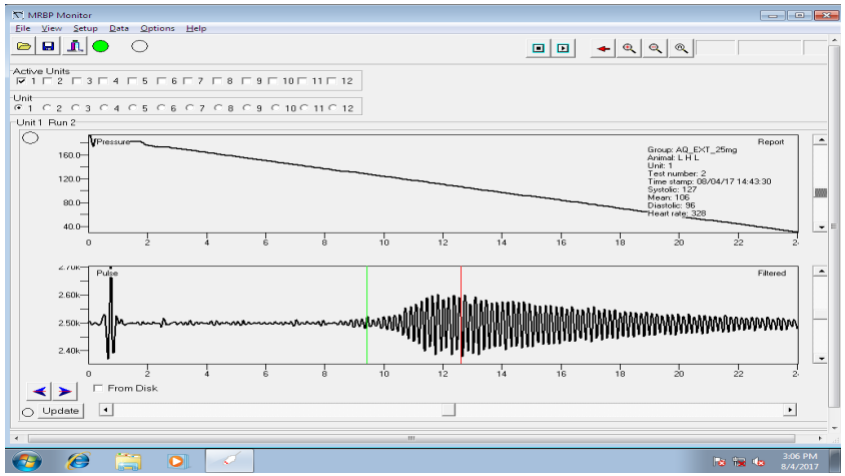


Figure 9. MRBP Polygraph chart of *Wistar* Rat Administered AESG 50 mgkg^{-1}

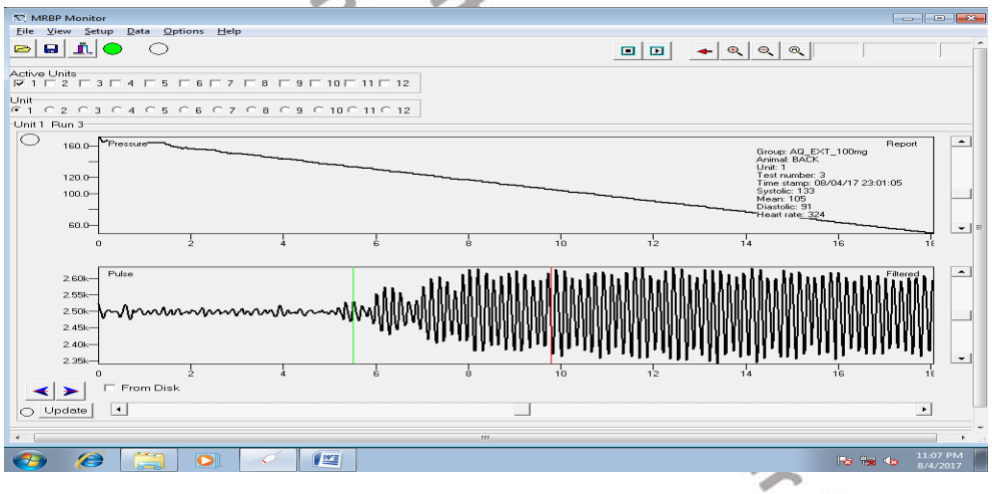


Figure 10. MRBP Record polygraph chart of *Wistar* Rat Administered AESG 100 mgkg^{-1}

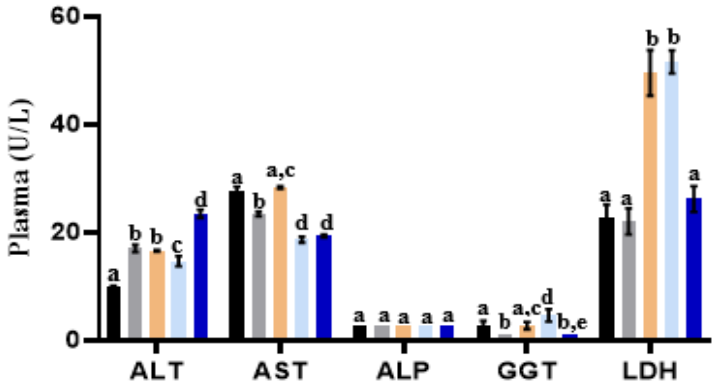


Figure 11. Prophylactic effect of varying Doses of AESG on Liver Function of Male Wistar Rats Exposed to 8 % NaCl, Normal Saline for 4 weeks, against the Normotensive & Hypertensive groups respectively. Data with similar lowercase alphabets are not significantly different amongst mean ($P \geq 0.05$); whereas, data with different lowercase alphabets are significantly different amongst mean ($P \leq 0.05$). Data are Mean ± SD ($n \geq 3$).

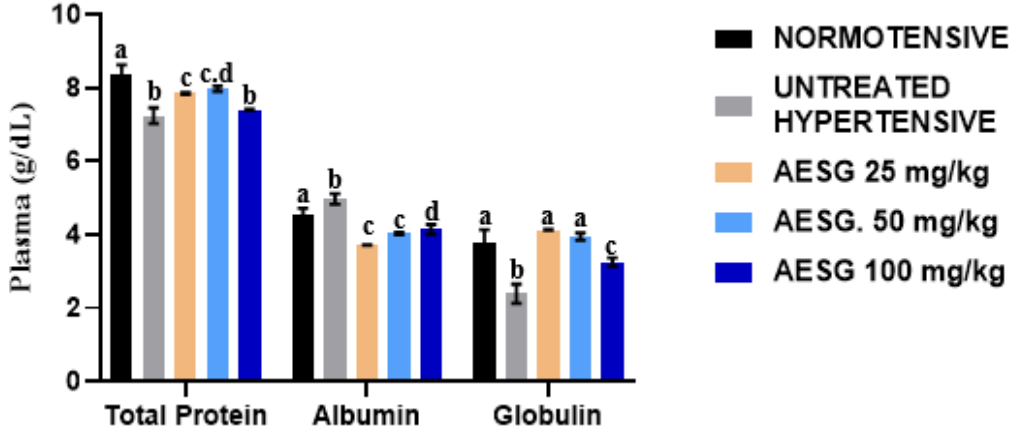


Figure 12. Prophylactic effect of varying Doses of AESG on Liver synthesizing Function of Male Wistar Rats Exposed to 8 % NaCl, Normal Saline for 4 weeks, against the Normotensive & Hypertensive groups respectively. Data with similar lowercase alphabets are not significantly different amongst mean ($P \geq 0.05$); whereas, data with different lowercase alphabets are significantly different amongst mean ($P \leq 0.05$). Data are Mean ± SD ($n \geq 3$).

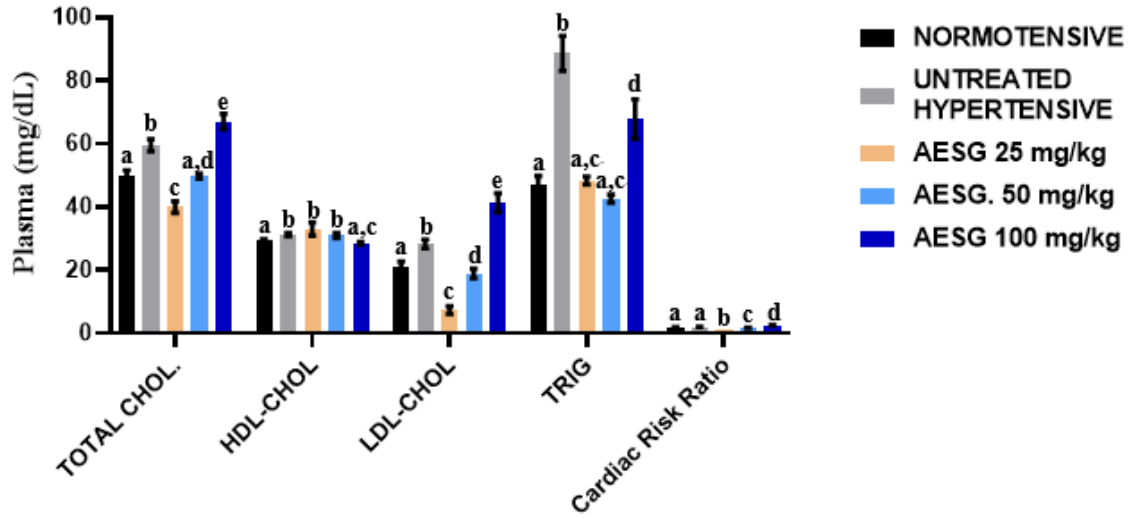


Figure 13. Prophylactic effect of varying Doses of AESG on Plasma Lipid Profile & Cardiac Risk Ratio of Male Wistar Rats Exposed to 8 % NaCl, Normal Saline for 4 weeks, against the Normotensive & Hypertensive groups respectively. Data with similar lowercase alphabets are not significantly different amongst mean ($P \geq 0.05$); whereas, data with different lowercase alphabets are significantly different amongst mean ($P \leq 0.05$). Data are Mean ± SD ($n \geq 3$).

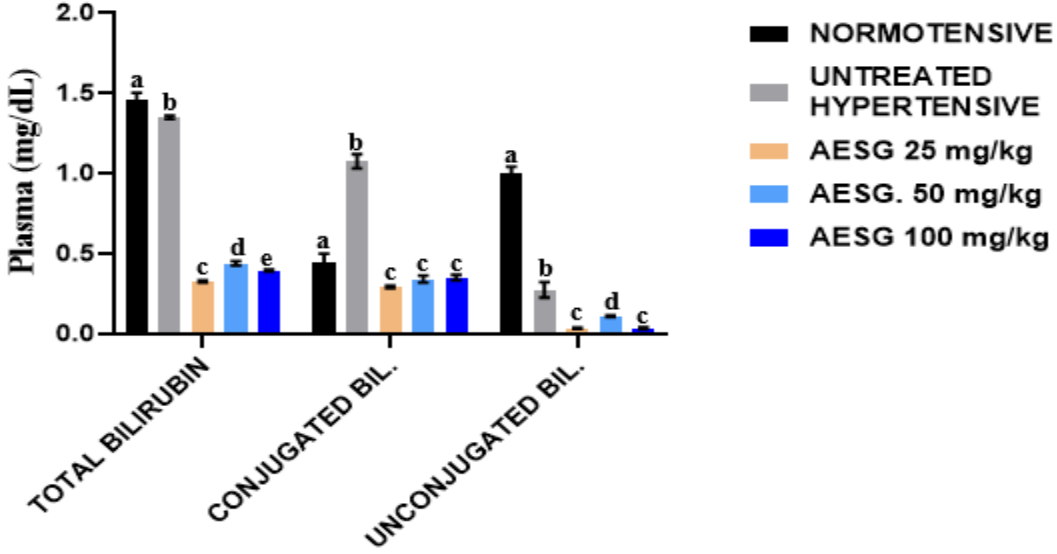


Figure 14. Prophylactic Effect of varying Doses of AESG on Plasma Total Bilirubin, Conjugated & Unconjugated Bilirubin of Male Wistar Rats Exposed to 8 % NaCl, Normal Saline for 4 weeks, against the Normotensive & Hypertensive groups respectively. Data with similar lowercase alphabets are not significantly different amongst mean ($P \geq 0.05$); whereas, data with different lowercase alphabets are significantly different amongst mean ($P \leq 0.05$). Data are Mean ± SD ($n \geq 3$).

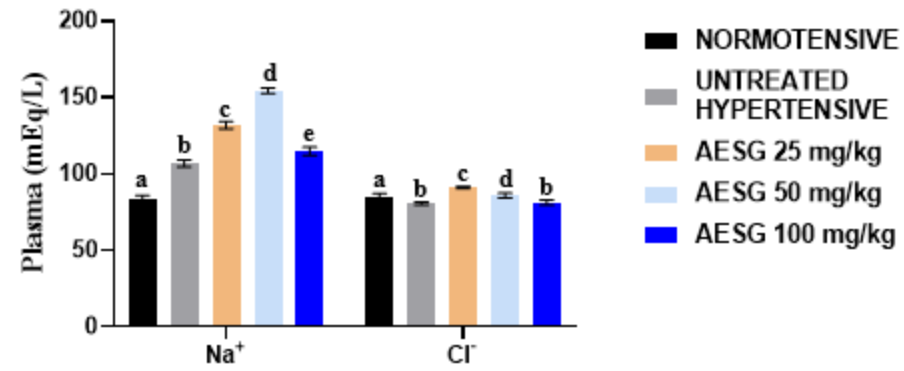


Figure 15. Prophylactic Effect of varying Doses of AESG on Plasma Sodium & Chloride ions of Male Wistar Rats Exposed to 8 % NaCl, Normal Saline for 4 weeks, against the Normotensive & Hypertensive groups respectively. Data with similar lowercase alphabets are not significantly different amongst mean ($P \geq 0.05$); whereas, data with different lowercase alphabets are significantly different amongst mean ($P \leq 0.05$). Data are Mean ± SD ($n \geq 3$).

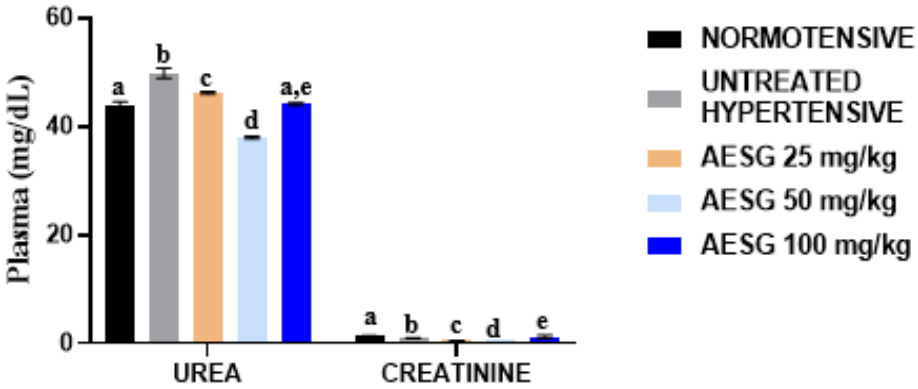
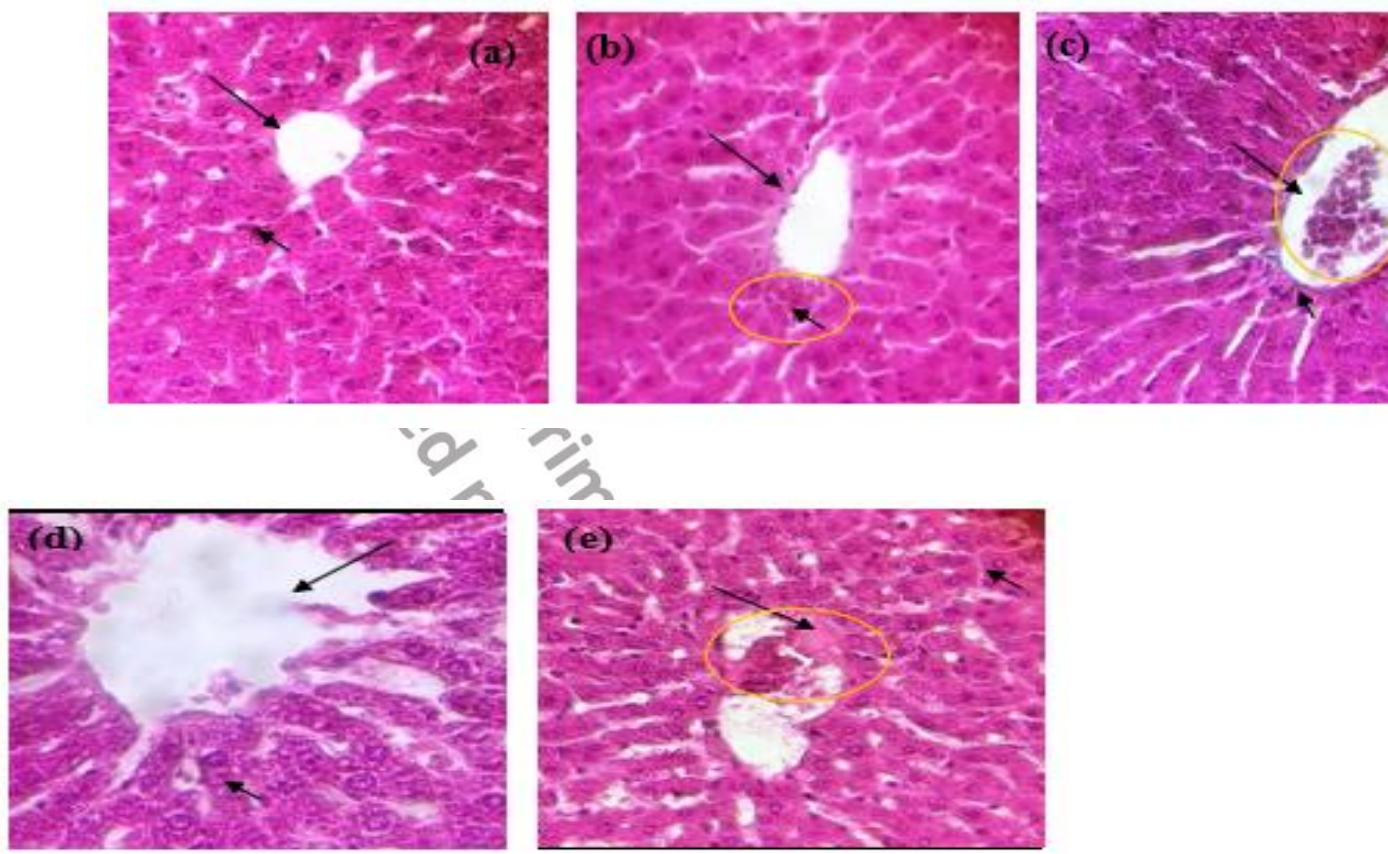


Figure 16. Prophylactic Effect of varying Doses of AESG on Plasma Urea & Creatinine levels of Male Wistar Rats Exposed to 8 % NaCl, Normal Saline for 4 weeks, against the Normotensive & Hypertensive groups respectively. Data with similar lowercase alphabets are not significantly different amongst mean ($P \geq 0.05$); whereas, data with different lowercase alphabets are significantly different amongst mean ($P \leq 0.05$). Data are Mean ± SD ($n \geq 3$).

Histology Evaluation

Liver Tissue

Figure 17a. Normotensive group: Illustrates distinct central venule (long arrow), hepatocytes with quite healthy nucleus (short arrow); well fenestrated sinusoids. Figure 17b. Untreated Hypertensive group: Illustrates distinct central venule (long arrow) with noticeable inflammatory cells surrounding it (short arrow) the hepatocytes show mild stenosis. Figure 17c. AESG 25 mgkg^{-1} group: Illustrates centriole with thickened wall surrounding by inflammatory cells (long arrow). There is quite a stenosis (short arrow). Figure 17d. AESG 50 mgkg^{-1} group: Illustrates distinct centriole (long arrow). There is prominent stenosis of fatty changes (short arrow). Figure 17e. AESG 100 mgkg^{-1} group: Illustrates slightly congested central venule (long arrow). There is prominent stenosis and mild fatty changes (short arrow).



(a) **Normotensive group**: Illustrates distinct central venule (**long arrow**), hepatocytes with quite healthy nucleus (**short arrow**); well fenestrated sinusoids.

(b) **Untreated Hypertensive group**: Illustrates distinct central venule (**long arrow**) with noticeable inflammatory cells surrounding it (**short arrow**); the hepatocytes show mild stenosis.

(c) **AESG 25 mgkg-1 group**: Illustrates centriole with thickened wall surrounding by inflammatory cells (**long arrow**). There is quite a stenosis (**short arrow**)

(d) **AESG 50 mgkg^{-1} group**: Illustrates distinct centriole (**long arrow**). There is prominent stenosis of fatty changes (**short arrow**)

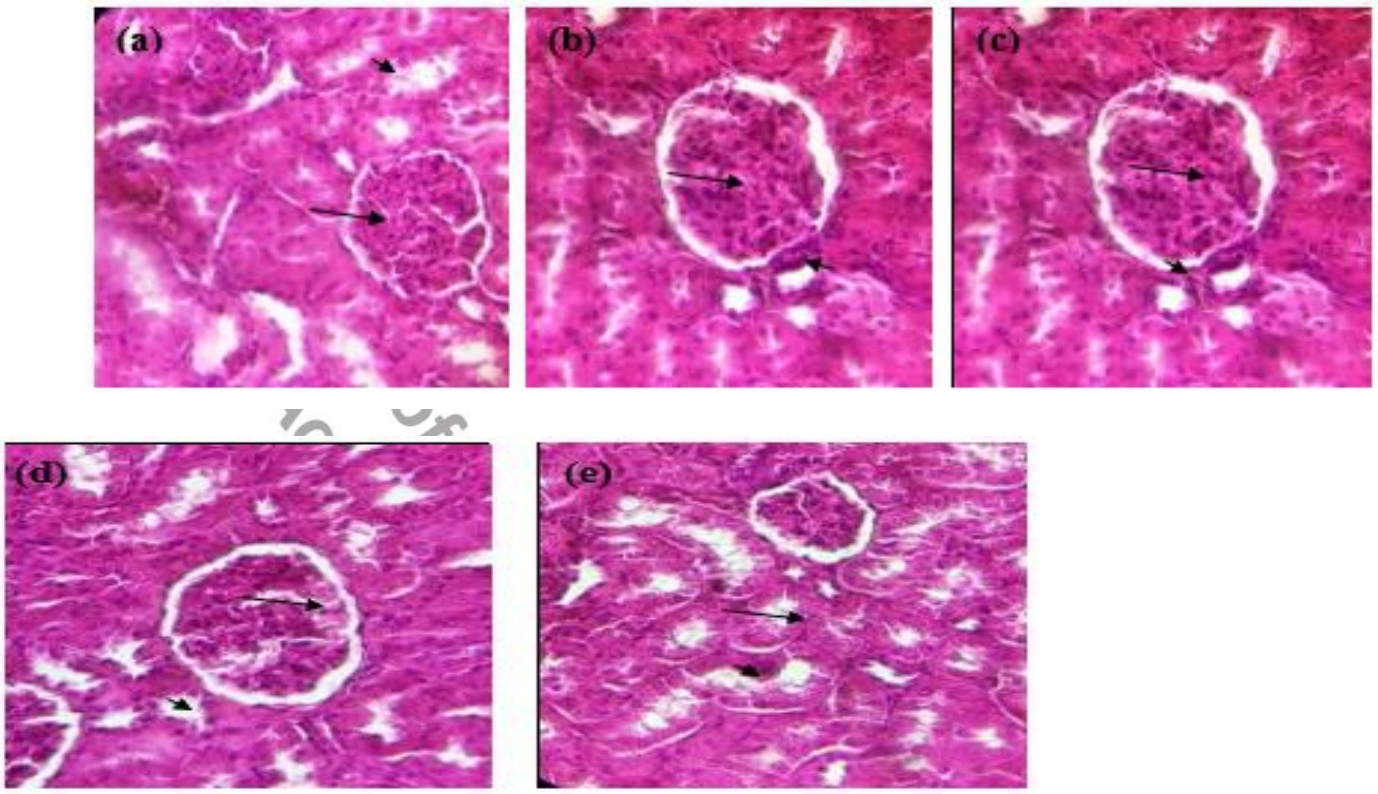
(e) **AESG 100 mgkg^{-1} group**: Illustrates slightly congested central venule (**long arrow**). There is prominent stenosis and mild fatty changes (**short arrow**)

Kidney Tissue

Figure 18a. Normotensive group: reveals visible renal corpuscle (long arrow) and distinct interstitial space (short arrow) and tubules. Figure 18b. Untreated Hypertensive group: reveals visible renal corpuscle (long arrow) with mild inflammatory cells surrounding it (short arrow) the tubules appear not so distinct. Figure 18c. AESG 25 mgkg^{-1} group: reveals renal

corpuscles with slight granulation (long arrow). There is mild distortion in the tubules (short arrow). Figure 18d. AESG 50 mgkg^{-1} group: reveals renal corpuscles with slight granulation (long arrow). There is visible interstitial and tubules (short arrow). Figure 18e. AESG 100 mgkg^{-1} group: reveals visible atrophied renal corpuscle (long arrow) with distortion in the tubules (short arrow).

Fig. 18. Kidney x400 (H & E)



(a) Normotensive group: reveals visible renal corpuscle (**long arrow**) and distinct interstitial space (**short arrow**) and tubules.

(b) Untreated Hypertensive group: reveals visible renal corpuscle (**long arrow**) with mild inflammatory cells surrounding it (**short arrow**); the tubules appeared not so distinct.

(c) AESG 25 mgkg^{-1} group: reveals renal corpuscles with slight granulation (**long arrow**). There is mild distortion in the tubules (**short arrow**)

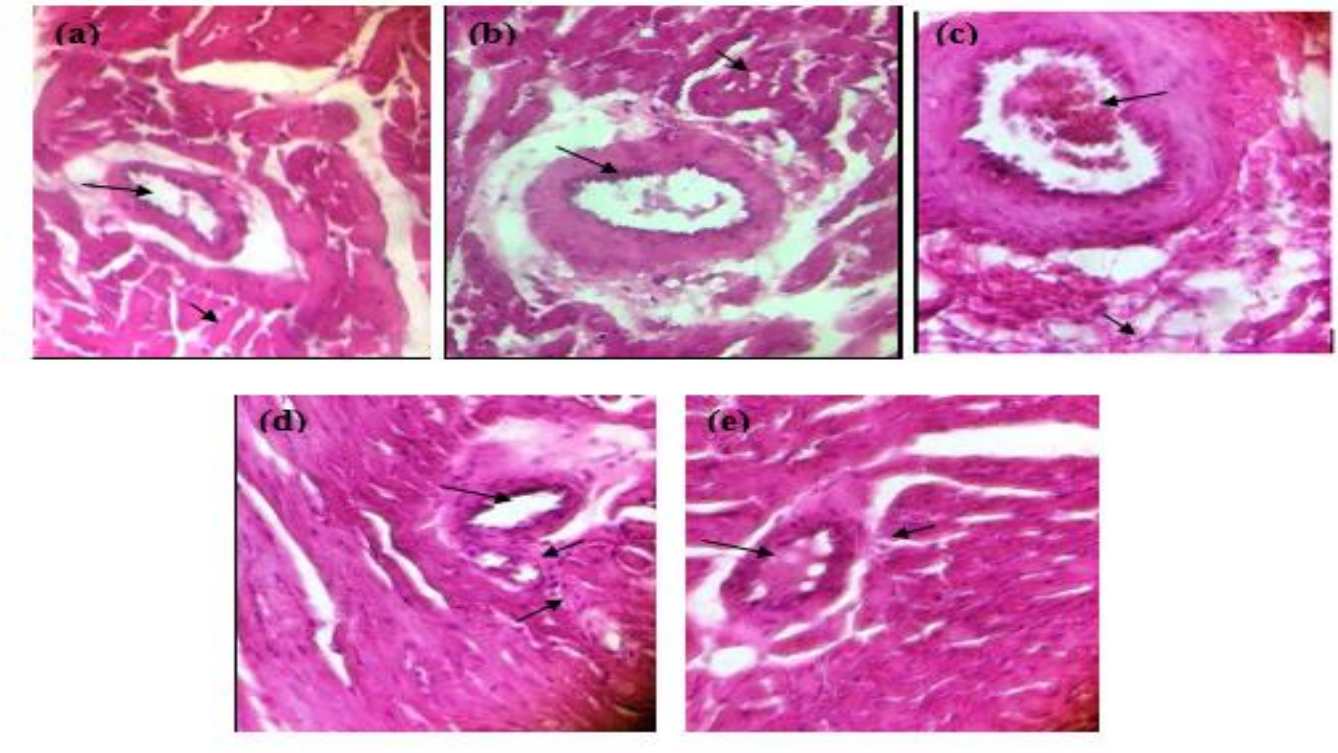
(d) AESG 50 mgkg^{-1} group: reveals renal corpuscles with slight granulation (**long arrow**). There is visible interstitial and tubules (**short arrow**)

(e) AESG 100 mgkg^{-1} group: reveals visible atrophied renal corpuscle (**long arrow**) with distortion in the tubules (**short arrow**).

Heart Tissue

Figure 19a. Normotensive group: illustrates visible bundles of myofibrils (short arrow); interstitial space with prominent coronary artery (long arrow). Figure 19b. Untreated Hypertensive group: reveals bundles of myofibrils (short arrow); interstitial space and visibly enlarged coronary artery (long arrow). Figure 19c. AESG 25 mgkg^{-1} group: reveals enlarged congested coronary artery (long arrow) with indistinct bundles of myofibrils (short arrow). Figure 19d. AESG 50 mgkg^{-1} group: reveals prominent coronary artery (long arrow), with visible myofibrils (short arrow); interstitial space with mild inflammatory infiltrates (short arrow). Figure 19e. AESG 100 mgkg^{-1} group: reveals visibly congested coronary artery (long arrow) with not so distinct myofibrils and interstitial; characterized by mild inflammation and fatty changes (short arrow).

Fig. 19. Heart x400 (H & E)



(a) Normotensive group: illustrates visible bundles of myofibrils (**short arrow**); interstitial space with prominent coronary artery (**long arrow**)

(b) Untreated Hypertensive group: reveals bundles of myofibrils (**short arrow**); interstitial space and visibly enlarged coronary artery (**long arrow**)

(c) AESG 25 mgkg^{-1} group: reveals enlarged congested coronary artery (**long arrow**) with indistinct bundles of myofibrils (**short arrow**).

(d) AESG 50 mgkg^{-1} group: reveals prominent coronary artery (**long arrow**), with visible myofibrils (**short arrow**); interstitial space with mild inflammatory infiltrates (**short arrow**)

(e) AESG 100 mgkg^{-1} group: reveals visibly congested coronary artery (**long arrow**) with not so distinct myofibrils and interstitial; characterized by mild inflammation and fatty changes (**short arrow**).

Conclusion

Considering the analyzed data, the outcome of the study revealed that pre-treatment with respective doses of AESG prevented salt-load induced weight loss; elevated systolic, diastolic blood pressures, mean arterial blood pressure and heart rates. We equally observed that the plant extract did not cause injury on the liver and kidney. Prophylactic treatment with respective doses of AESG prevented dyslipidemia and cardiovascular risk associated with elevated lipoprotein cholesterols in salt-load induced hypertensive experimental models. On the overall, it was noted that the AESG demonstrated a significant antihypertensive effect; although, with little or no observed adverse effect on relevant metabolic organs and tissue considered in the study. Therefore, it is strongly recommended that further studies be conducted on other extracts of *S. glauca* to ascertain its antihypertensive efficacy; possible isolation of compounds and mechanism(s) of action.

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Authors' Contributions

SD.E, designed the work, conducted the analysis and interpretation of the data; reported the outcome of the analyzed data obtained from the study. **N.E.J** contributed to the concept of the study and approved the submission of the corrected version.

Conflict of Interest Declaration

The Author(s) declares and disclose that there are no competing or conflicts of interests before, during the course of the work and after.

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