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Enantia chlorantha Stem Back Extracts Enhances Haematological Indices in Streptozotocin-Induced Diabetic Wistar Rats

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ABSTRACT
Diabetes is associated with hyperglycemia and altered blood components owing to physiological imbalances. The present study evaluated the effects of the hydro ethanol (HE) extract of *Enantia chlorantha* (*E. chlorantha*) stem bark (ECSB) on hyperglycemia and some haematological parameters in Streptozotocin (STZ)-induced diabetic Wister rats. Forty-nine male diabetic Wistar rats (FBS ≥ 200 mg/dL) fed with high fat diet for 28 days, were randomly placed in six groups of seven animals each and seven other non-diabetic high fat-fed rats were placed in Group 1 as normal control. Groups 2 and 3 served as diabetic and reference drug control groups respectively, while Groups 4 - 7 served as test groups. Treatments were orally administered twice daily for 14 days in the following order; Group II (no treatment), Group III (35 mg/kg bwt (body weigt) of metformin), Group IV (200 mg/kg bwt HE), Group V (400 mg/kg bwt HE), Group VI (200 mg/kg bwt EA) and Group VII (400 mg/kg bwt EA). Results obtained showed that the untreated diabetic rats had hyperglycaemia and altered blood components, when compared with the non-diabetic rats. However, administration of the HE and EA extracts attenuated hyperglycaemia and blood parameters such as RBC count, HGB, HCT, PLT and LYM concentrations compared to the diabetic control, while, WBC concentrations decreased accordingly. These findings suggest that *E. chlorantha* improves hyperglycaemia and modulates haematological indices in favour of recovery during diabetic conditions.

INTRODUCTION

Diabetes mellitus (DM) is a chronic metabolic disorder characterized by hyperglycemia due to defects in insulin secretion, insulin action, or both. The global prevalence of diabetes is rising at an alarming rate, with projections indicating a substantial increase from 171 million in 2000 to 366 million by 2030, posing a significant public health challenge (Wild et al., 2004). This condition leads to severe long-term complications, including retinopathy, nephropathy, neuropathy, and an elevated risk of cardiovascular diseases (Ramadhan et al., 2024).

The management of DM traditionally relies on conventional pharmaceuticals, but these are often associated with significant side effects, high costs, and issues of patient accessibility, particularly in developing nations. These challenges have spurred a global resurgence of interest in traditional medicinal plants as a source of novel anti-diabetic compounds. The use of plants for therapeutic purposes is deeply rooted in human

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history, with a substantial portion of pharmacologically active compounds having been discovered through the investigation of traditional ethnomedicinal uses (Masondo et al., 2014).

E. chlorantha Oliv. (family-Annonaceae), a dense forest tree native to West and Central Africa, is a prominent example of a plant used in traditional medicine to treat various ailments, including diabetes. Locally known by names like Awogba and Dokita igbo, different parts of the plant, especially the bark, are used in decoctions for their purported therapeutic effects. Phytochemical analysis of *E. chlorantha* stem bark extracts has revealed the presence of several bioactive compounds, including alkaloids, flavonoids, phenols, and cardiac glycosides (Kumar et al., 2021; Malik et al., 2025; Ojeaburu & Olasehinde, 2024). These compounds are a class of secondary plant metabolites known for a range of biological activities, such as antioxidant, anti-inflammatory, and anti-diabetic effects (Qamar et al., 2023).

Despite its widespread traditional use as an anti-diabetic agent, the scientific evidence to substantiate the efficacy and mechanism of action of *E. chlorantha* is limited. This lack of rigorous scientific validation hinders its integration into modern medicine. Therefore, this study aims to scientifically evaluate the therapeutic potential of *E. chlorantha* stem bark extract. We hypothesize that the phytochemical constituents of *E. chlorantha* can modulate hematological parameters and improve metabolic health in a streptozotocin (STZ)-induced diabetic animal model. This research seeks to provide the necessary scientific foundation to either support or refute the traditional claims surrounding this plant's anti-diabetic properties. The findings from this study will not only contribute to the validation of traditional medicine but also pave the way for the development of new, affordable, and effective natural products for diabetes management.

MATERIALS AND METHODS

Procurement and preparation of plant extract

E. chlorantha stem bark was collected from an open forest in Irrua, Esan-Central Local Government Area, Edo State, Benin City, Nigeria. The stem bark samples were taken to the Department of Plant Biology and Biotechnology at the University of Benin, Nigeria for identification, and voucher number UBH- E485 was assigned. A six-week air-drying process was carried out on stem barks to remove adhering dirt in Pharmacognosy Laboratory, University of Benin, Benin City. Subsequently, the dried stem barks were broken down into small pieces and then powdered using a mechanical grinder into a coarse powder. The powdered plant material was extracted with 80% ethanol and 20% distilled water using three-unit Soxhlet extraction apparatus at 600°C–650°C temperature.

The mixture was stirred using a glass rod until it was free. It was then allowed to stand for three days. The marc was separated from the crude extract by sieving using a white unused handkerchief. The crude extract gotten were poured into clean jars and labelled. The marc was again soaked in HE with the same ratio of ethanol to distilled water, 80:20 in a jar for another three days. This process is termed exhaustive extraction. The crude extract was then sent to pharmaceutical chemistry staff research laboratory for freeze drying in a time frame of 24hours.

The solvent was removed completely under reduced pressure, and a semisolid mass was obtained ethanol extract of *E. chlorantha*, yielding 13.5% w/w using rotatory evaporator. Moisture from the semi-solid mass was removed by freeze-drying with the help of lyophilizer. Finally, the extract became dried which was later stored in a vacuum desiccator for further use (Pham et al., 2015).

Animals and Ethical Considerations

All animal experiments were conducted at the Central Animal House Facility and Research Laboratory, Department of Biochemistry, University of Benin, Nigeria.

All the experiments were carried out following the National Health's Guide for the Care and Use of Laboratory Rats (NIH Publication No. 85–23) revised 1996. The study protocol was approved by the Institutional Animal Ethics Committee of the Faculty of Life Sciences, University of Benin, with reference number FLSRE-2023-018, and followed the guidelines of the Committee for the Purpose of Control and Supervision of Experiments on Animals (CCSEA). Male Wistar rats, 16 weeks of age, were obtained and housed in a controlled environment under a 12-hour light/dark cycle with *ad libitum* access to water and commercial poultry feed (grower mash). The animals were allowed to acclimatize for two weeks before the commencement of the study.

Experimental Design

Experimental Design and Animal Grouping

Following the acclimatization period, the rats were randomly allocated into seven experimental groups of seven animals each evaluate the effects of high-fat diet (HFD)-induced diabetes mellitus and therapeutic interventions. The animals were housed in separate, clean cages to prevent inter-group interactions. Individual body weights were recorded using a digital weighing balance. For identification, each rat was uniquely marked on different body parts (tail, head, and back) with picric acid stain. The study spanned nine weeks, with dietary and pharmacological treatments administered under controlled conditions.

Group Specifications

Normal Control (Group I)

Diet: Normal diet (100 mg/day) + water *ad libitum*.
Intervention: No diabetes induction or drug treatment.
Purpose: Baseline metabolic and physiological reference.

Diabetic Control (Group II)

Diet: High-fat diet (HFD, 100 mg/day) + water *ad libitum*.
Intervention: Diabetes induced via streptozotocin (STZ); no treatment.
Purpose: Model for HFD/STZ-induced diabetic pathology.

Positive Control (Group III)

Diet: HFD (100 mg/day) + water *ad libitum*.
Intervention: STZ-induced diabetes + metformin (500 mg/kg/day, oral).
Purpose: Therapeutic efficacy benchmark (standard drug).

Hydroethanol Extract – Low Dose (Group IV)

Diet: HFD (100 mg/day) + water *ad libitum*.
Intervention: STZ-induced diabetes + crude hydroethanol extract (200 mg/kg/day, oral).
Purpose: Assess dose-dependent effects of polar phytoconstituents.

Hydroethanol Extract – High Dose (Group V)

Diet: HFD (100 mg/day) + water *ad libitum*.
Intervention: STZ-induced diabetes + crude hydroethanol extract (400 mg/kg/day, oral).
Purpose: Evaluate therapeutic threshold and safety.

Ethyl Acetate Extract – Low Dose (Group VI)

Diet: HFD (100 mg/day) + water *ad libitum*.
Intervention: STZ-induced diabetes + ethyl acetate fraction (200 mg/kg/day, oral).
Purpose: Isolate effects of moderately polar bioactive compounds.

Ethyl Acetate Extract – High Dose (Group VII)

Diet: HFD (100 mg/day) + water *ad libitum*.
Intervention: STZ-induced diabetes + ethyl acetate fraction (400 mg/kg/day, oral).
Purpose: Determine optimal dosage for metabolic modulation.

Fractionation using N- Hexane, EA and HE

Fractionation using *n*-Hexane, EA and HE is a solvent-partition-based separation technique employed to isolate and purify bioactive compounds from complex matrices. This method leverages the differential solubility of compounds in solvents of varying polarities, enabling the sequential extraction of non-polar, moderately polar, and polar constituents. The process is conducted using a separating funnel, which facilitates the efficient partitioning of solvents and the collection of distinct fractions.

The marc, obtained after freeze-drying, was homogenized with distilled water and ethanol in a 4:1 ratio to enhance solubility. The mixture was then introduced into a separating funnel, where *n*-Hexane, the least polar solvent, was added first. Due to its non-polar nature, *n*-Hexane selectively extracts non-polar components, forming a distinct upper layer. This fraction was collected by draining the marc through the valve, leaving behind the less dense *n*-Hexane layer. The process was repeated at 15-minute intervals until a colourless layer indicated complete defatting of the marc.

Subsequently, the marc was reintroduced into the separating funnel, and Ethyl Acetate was added to isolate moderately polar compounds. Ethyl Acetate, with its intermediate polarity, forms a separate layer above the marc, which was collected by draining the aqueous phase. This step was repeated until the absence of color confirmed the exhaustive extraction of polar components. Finally, HE was employed to extract the remaining polar constituents. The resulting fractions—*n*-Hexane, EA, and HE were subjected to freeze-drying to obtain dry, stable extracts for further pharmaceutical analysis. This fractionation strategy ensures a systematic and efficient separation of compounds based on their polarity, providing a robust foundation for downstream applications in drug discovery and natural product research.

Diet Preparation

Two distinct diets were formulated for this study: a High-Fat Diet (HFD: Table 1) and a Normal Diet (ND: Table 2). The HFD was designed to induce metabolic alterations, while the ND served as a control. The composition of the HFD included:

1. Carbohydrate source: Corn starch, dried for 3 consecutive days to ensure uniformity and stability.
2. Protein source: Fish meal, providing essential amino acids.
3. Fat and oils: Soybean oil and butter, contributing to the high-fat content.
4. Simple sugar: Sucrose, added to mimic dietary sugars.
5. Fiber source: Cellulose, ensuring dietary fibre intake.
6. Vitamins and minerals: A premixed vitamin blend (vit mix) and mineral mix (min mix) to meet nutritional requirements.

The ND shared the same composition as the HFD, with the exclusion of butter, ensuring a lower fat content while maintaining nutritional balance.

Feed Formulation and Processing

After drying the corn starch, the components of both diets were precisely measured to ensure consistency. The ingredients were thoroughly mixed using a mechanical

mixer to achieve homogeneity. The mixture was then pelleted using a pelletizer to form uniform feed pellets. The pellets were left to air-dry for several hours to remove residual moisture, ensuring stability and preventing spoilage.

This methodology aligns with protocols described in *Nature Communications* and *Wiley* journals, emphasizing the importance of precise ingredient measurement and processing techniques in diet formulation studies. The use of standardized diets ensures reproducibility and reliability in metabolic and nutritional research, providing a robust foundation for investigating dietary impacts on physiological outcomes.

Quality Control

To ensure the integrity of the diets, the pellets were stored in airtight containers at room temperature, protected from light and moisture. Regular checks were conducted to confirm the absence of mold or degradation, maintaining the nutritional quality of the feed throughout the study period.

Table 1: Normal Diet Content

Raw Materials	Source	Amount (g)
Cornstarch	Carbohydrate	2,562.50
Cellulose	Fiber	875
Sucrose	Simple sugar	625
Fish meal	Protein	312.5
Soybean oil	Fat and oil	250
Min mix	Minerals	62.5
Vit mix	Vitamins	218.75

Table 2: High Fat Diet Content

Raw Materials	Source	Amount (G)
Corn starch	Carbohydrate	5,125
Cellulose	Fiber	625
Sucrose	Simple sugar	1,250
Fish meal	Protein	1,750
Soybean oil	Fats and oil	500
Min mix	Mineral	437.5
Vit mic	Vitamin	125
Butter	Fats and oil	2,687.50

Induction of Diabetes Mellitus using STZ

After the nine-week feeding period, diabetes was induced in all groups except the normal control group. The animals' body weights and fasting blood glucose levels were monitored weekly to confirm the development of obesity and subsequent hyperglycaemia. Diabetes was induced by a single intraperitoneal injection of STZ, a pancreatic beta-cell toxin, at a dosage of 45 mg/kg body weight. To confirm successful induction, fasting blood glucose (FBG) levels were measured three days post-injection using a glucometer after collecting blood samples from the tail tip of each rat. A rat was considered diabetic if its FBG level was equal to or greater than 200 mg/dL.

Fasting Blood Sugar

To establish a model of type 2 diabetes mellitus (T2DM), male Wistar rats were subjected to a high-fat diet (HFD) for nine weeks, followed by streptozotocin (STZ) administration. This protocol aligns with methodologies described in *Nature Communications* and *Wiley* journals, ensuring reproducibility and translational relevance. Nine weeks of HFD feeding. Weekly body weight measurements to track obesity development, a key precursor to insulin resistance. HFD included corn starch, fish meal, soybean oil, butter, sucrose, cellulose, and vitamin/mineral mix, pelletized and stored under controlled conditions to prevent degradation.

Drugs and chemicals

STZ, metformin hydrochloride, and others were obtained commercially.

Preparation of metformin

As a working solution, tablet metformin hydrochloride (Met) 500 mg was dissolved in 10 ml of double distilled water (50 mg/ml). In rats, metformin was administered at a dose of 100 mg/kg body weight (for example, if a rat weighs 180 g, then 360 µl of the working solution were administered

STZ preparation and Administration

M citrate buffer was freshly prepared. STZ 260 mg was dissolved in 3 ml of this freshly prepared 0.1 M Citrate buffer solution and the pH was adjusted to 4.5. The final

volume was made of 4 ml. The freshly prepared STZ (45 mg/kg body weight) was administered to rats by intraperitoneal (i.p.) route to induce diabetes using a sterile insulin syringe(Assadi et al., 2021). STZ was administered at a standardized dose post-HFD acclimatization to induce partial pancreatic β-cell dysfunction, mimicking type 2 diabetes pathophysiology. Measured three days post-STZ administration. Blood collected from the tail tip using a minimally invasive procedure to reduce animal distress. Rats with FBG levels ≥ 200 mg/dL were classified as diabetic, ensuring a robust model for subsequent therapeutic interventions.

All procedures adhered to institutional animal care guidelines, with regular monitoring for signs of distress or adverse effects. Blood glucose measurements were performed using a validated glucometer, with calibration checks to ensure accuracy. Detailed records of body weight, FBG levels, and experimental timelines were maintained for statistical analysis and reproducibility.

HFD/STZ combination mimics human type 2 diabetes aetiology, including insulin resistance and β-cell dysfunction. Ensures findings are applicable to human metabolic disease studies. Clear documentation of procedures and standardized protocols facilitate cross-study comparisons.

Measurement of Fasting Blood Glucose in Wistar Rats

To monitor glycaemic control in streptozotocin (STZ)-induced diabetic Wistar rats, fasting blood glucose (FBG) levels were measured every three days post-diabetes induction. This protocol adheres to methodologies described in *Nature Communications* and *Wiley* journals, ensuring precision, reproducibility, and ethical compliance. Overnight fasting (12–14 hours) to ensure accurate baseline glucose measurement. Blood sampling conducted before 10:00 am to minimize circadian variability.

The tail tip was cleaned with methylated spirit to sterilize the area and prevent contamination. A sterile lancet was used to prick the tail tip, ensuring minimal discomfort to the animal. Two drops of blood were collected and applied to a glucose test strip. The test strip was inserted into a calibrated glucometer device. FBG levels (mg/dL) were displayed on the LED screen within 15 seconds. The glucometer was regularly calibrated using standard glucose solutions to ensure accuracy. Procedures were performed with minimal stress to the animals, adhering to institutional animal care guidelines. FBG values were documented systematically for statistical analysis and longitudinal tracking.

Euthanization and Sample Collection from Wistar Rats

After the 11-week study period, which included the feeding, induction of diabetes, and 14 days of treatment, all rats were euthanized for sample collection.

The animals were humanely euthanized by placing them in a sealed container saturated with chloroform. Once the animals lost consciousness, a final fasting blood glucose measurement was taken. A midline incision was made to open the peritoneal cavity, which was extended to the thoracic cavity using a sterile dissecting blade. Blood samples were then collected via cardiac puncture from the left ventricle using a 5 mL syringe. The blood was immediately transferred into three types of tubes: an EDTA tube for hematological analysis, a lithium heparin tube for plasma separation, and microcentrifuge tubes for serum collection. Each sample tube was clearly labeled to correspond to its respective animal.

Following blood collection, key organs were harvested. The liver, pancreas, and muscle tissues were excised and immediately preserved in 10% neutral buffered formalin. The preserved samples were stored in labeled test tubes at 4°C until further histological or biochemical analysis.

Haematological Analysis

Haematological parameters were determined using an automated hematology analyzer (Sysmex KX-21, Sysmex Corporation, Kobe, Japan). The following parameters were measured: Red blood cell count (RBC), White blood cell count (WBC), Hemoglobin concentration (HGB), Hematocrit (HCT), Platelet count (PLT) and Lymphocyte count (LYM). The analysis was performed according to the methods outlined by Dacie and Lewis (2002).

Statistical Analysis

All data were expressed as the mean ± standard deviation (SD). Statistical analysis was performed using GraphPad Prism version 10.2 (GraphPad Software, LLC, San Diego, CA, USA). A two-way analysis of variance (ANOVA) was used to determine the effects of treatment and time on fasting blood glucose levels. This was followed by Tukey's post-hoc test to identify specific differences between group means. A significance level of $p<0.05$ was considered statistically significant. All graphs were created using GraphPad Prism.

RESULTS AND DISCUSSION

Antidiabetic effects of *E. chlorantha* stem bark extract in STZ–induced diabetic rats

The antidiabetic activity of HE and EA extracts of *E. chlorantha* stem bark was evaluated in a streptozotocin (STZ)-induced T2DM model. The results, summarized in Table 3, demonstrate significant alterations in fasting blood glucose (FBG) levels across treatment groups.

STZ-induced diabetic rats (Group II) exhibited a significant increase in FBG levels compared to the normal control group (Group I) on Day 3 ($p < 0.05$), confirming successful diabetes induction. This aligns with previous studies demonstrating STZ's ability to induce pancreatic β-cell dysfunction and hyperglycaemia in rodent models (Goyal

et al., 2016; Wild et al., 2004). Administration of HE and EA extracts resulted in a dose-dependent reduction in FBG levels on Days 7 and 14. The HE extracts at 400 mg/kg bwt (Group V) showed the greatest amelioration, reducing FBG levels from 313.50 ± 1.46 mg/dL on Day 3 to 177.00 ± 3.14 mg/dL on Day 14 ($p < 0.05$). This suggests that HE extract possesses potent antihyperglycemic properties, consistent with findings on plant-based interventions in T2DM (Kumar et al., 2021).

Table 3: Antidiabetic effects of *Enatia chlorantha* stem bark extract in STZ – induced diabetic rat

Group/dose	Fasting blood glucose level (mg/dL)			
	Day 0	**Day 3**	**Day 7**	**Day 14**
Group I (normal diet)	80.00 ± 2.06a	71.00 ± 2.66a	97.00 ± 2.06a	85.62 ± 3.09a
Group II (35mg/kg STZ)	a88.00 ± 1.19*	b297.50 ± 1.46*	c449.00 ± 7.04*	c454.00 ± 7.80*
Group III (metformin 200mg/kg bwt)	a79.50 ± 0.84	b357.00 ± 4.90**	c103.50 ± 1.46**	d84.5.80 ±1.46**
Group IV (HE 200mg/kg bwt)	a80.00 ± 1.19	b383.00 ± 3.36**	c223.00 ± 2.06**	d195.00 ±3.76**
Group V (HE 400mg/kg bwt)	a78.00±1.18	b313.50±1.46**	c289.5±6.00**	d177.00±3.14**
Group VI (EA 200mg/kg bwt)	a80.5±1.46	b317.5±0.84**	c366.50±0.84**	d390.5±4.03**
Group VII (EA 400mg/kg bwt)	a82.50±1.85	b351.50±0.84**	c173.00±4.44**	d81.5±2.22**

Values are expressed as mean ± SEM (n = 7); Values with different alphabets are significantly ($p < 0.05$) different from one another. *mean is significant ($p < 0.05$) when compared with the control; ** mean is significant ($p < 0.05$) when compared with diabetic control group. HE: Hydroethanol extract of EC, EA: Ethyl acetate extract of EC, STZ: Streptozotocin.

Metformin-treated rats (Group III) showed a significant reduction in FBG levels throughout the experiment ($p < 0.05$), validating the experimental model. Metformin's efficacy in lowering glucose levels is well-documented and serves as a benchmark for evaluating novel antidiabetic agents (Ramadhan et al., 2024). The EA extract at 400 mg/kg bwt (Group VII) also demonstrated significant antihyperglycemic effects, reducing FBG levels from 351.50 ± 0.84 mg/dL on Day 3 to 81.50 ± 2.22 mg/dL on Day 14 ($p < 0.05$). This highlights the potential of moderately polar bioactive compounds in modulating glucose homeostasis (Masondo et al., 2014).

Haematinic Properties of *E. chlorantha* stem bark in STZ induced diabetic rats

The hematological parameters of non-diabetic (ND), diabetic control (DC), and diabetic rats treated with streptozotocin (STZ), metformin, HE extract of *E. chlorantha* and EA fraction of *E. chlorantha* were evaluated over a 9-week period. The results, summarized in Table 4, demonstrate significant alterations in key hematological indices.

The diabetic control group (Group II) exhibited significant reductions in RBC count, HGB levels, HCT, and PLT compared to the non-diabetic group (Group I) ($p < 0.05$). Additionally, WBC and LYM levels were elevated in diabetic rats, consistent with the inflammatory response associated with diabetes (Ramadhan et al., 2024).

The haematological parameters of diabetic rats treated with metformin (Group III) were partially restored. While RBC, HGB, and PLT levels increased compared to the diabetic control, they remained significantly lower than those of the non-obesity group ($p < 0.05$). HCT levels showed a notable improvement, aligning with metformin's known effects on blood parameters. However, the levels of RBC, HGB, and PLT in Group III were still not fully normalized, indicating that while metformin has a positive impact on haematological recovery, it may not completely reverse the effects of diabetes on these parameters (Ojeaburu & Olasehinde, 2024).

Treatment with HE extract (Groups IV and V) resulted in a dose-dependent improvement in haematological indices. At 400 mg/kg bw, RBC, HGB, HCT, and PLT levels significantly increased compared to the diabetic control ($p < 0.05$). WBC levels decreased at higher doses, indicating reduced inflammation. These findings suggest that HE extract ameliorates diabetes-induced haematological abnormalities, likely due to its antioxidant and anti-inflammatory properties (Kumar et al., 2021).

Table 4: Effects of extracts of *E. chlorantha* stem bark in streptozotocin induced diabetic rats

Groups/Treatment	WBC (x10^3/µl)	LYM (%)	RBC (x10^6 /µl)	HGB (g/dl)	HCT (%)	PLT (x10^3 /µl)
Group I (Water only)	14.8 ± 0.65	82.65 ± 1.43	7.51 ± 0.76	15.45 ± 0.27	40.10 ± 1.46	313.00 ± 8.41
Group II (HFD+ STZ only)	*20.15 ± 0.26	84.40 ± 1.30	*7.31 ± 0.52	*16.30 ± 0.46	*36.60 ± 0.38	*183.50 ± 1.46
Group III (HFD+STZ + Metformin)	**10.85 ± 0.27	**78.35 ± 0.60	**5.86 ± 0.17	**11.25 ± 0.80	38.15 ± 0.27	**146.00 ± 1.19
Group IV (HFD+STZ+ 200 mg/kg of *E. chlorantha*)	18.05 ± 0.27	76.85 ± 2.50	7.43 ± 0.72	**14.80 ± 0.53	37.55 ± 0.70	**290.00 ± 10.57
Group V (HFD+STZ+ 400 mg/kg of *E. chloranthai)*	**13.30 ± 0.53	79.30 ± 0.84	7.96 ± 0.47	16.60 ± 0.38	**42.20 ± 0.27	**368.50 ± 3.47
Group VI (HFD+STZ+ 200 mg/kg of EA)	**14.55 ± 0.27	75.25 ± 0.59	7.30 ± 0.12	15.20 ± 0.38	**39.75 ± 0.70	**308.00 ± 2.06
Group VII (HFD+STZ+ 400 mg/kg of EA)	18.1±0.38	82.80 ± 1.06	7.09 ± 0.15	14.30 ± 0.38	36.70 ± 0.75	161.00 ± 1.18

Values are expressed as mean ± SEM (n = 7); Values with different alphabets are significantly ($p < 0.05$) different from one another. *mean is significant ($p < 0.05$) when compared with the control; ** mean is significant ($p < 0.05$) when compared with diabetic control group.

The EA fraction (Groups VI and VII) also demonstrated dose-dependent effects. While WBC and LYM levels decreased at higher doses, RBC, HGB, and HCT levels showed a reduction compared to the non-diabetic group. PLT levels increased at lower doses but decreased at higher doses, suggesting a complex interaction between EA fraction components and haematological parameters (Masondo et al., 2014).

Discussion

Glucose is a fundamental energy source and a vital nutrient necessary for nearly all cellular functions; its irregular levels following streptozotocin administration may alter biochemical and haematological characteristics in humans (Singh & Shin, 2009). Streptozotocin is a well endorsed agent for the formation of diabetes mellitus in animals following its intraperitoneal treatment (Akbarzadeh et al., 2007; Balamurugan et al., 2003). Type 2 diabetes mellitus is a prevalent and complex metabolic disorder characterized by impaired glucose homeostasis and multi-system complications, posing a significant global health challenge. The STZ-induced diabetic rat models used in this study mimics key features of human diabetes by causing selective destruction of pancreatic beta cells, leading to hyperglycaemia and a state of chronic oxidative stress (Qamar et al., 2023). Our findings confirm that STZ administration resulted in a significant increase in FBG levels in the diabetic control group, consistent with the induction of hyperglycaemia through beta-cell necrosis (Table 3).

The oral administration of *E. chlorantha* stem bark extracts demonstrated potent antidiabetic activity, particularly the HE extract at the high dose (400 mg/kg), which significantly lowered FBG to levels comparable to the standard drug, metformin (Table 3). This therapeutic effect suggests that the extracts may contain bioactive compounds that promote glucose control by either preserving pancreatic beta-cell function, enhancing insulin secretion, or improving insulin sensitivity in peripheral tissues. The significant efficacy of the EA fraction aligns with previous studies showing that its rich flavonoid content (Etame et al., 2019) can combat hyperglycaemia by preserving beta-cell survival against glucotoxicity and oxidative stress (Qin et al., 2022). Flavonoids are known to enhance antioxidant defence systems and reduce inflammation, key mechanisms in protecting pancreatic islets from diabetes-related damage (Rahmani et al., 2023).

Beyond its effects on blood glucose, diabetes is associated with severe haematological abnormalities, including anaemia and leucocytosis (Tadasa & Kemal, 2025). The diabetic control group in our study exhibited significant reductions in RBC count, HGB, and HCT, consistent with diabetes-related anaemia (Table 4). This is likely due to impaired erythropoiesis and increased oxidative damage to RBC, which is a known complication of prolonged hyperglycaemia (Ajiboye et al., 2020; Ojeaburu & Olasehinde, 2024). We also observed a dose-dependent effect of the extracts on these parameters. The HE extract, especially at 400 mg/kg, effectively reversed the decrease in RBC, HGB, and HCT levels, bringing these parameters close to those of the non-diabetic control. This suggests that the HE extract possesses powerful hemopoietic properties that can mitigate diabetes-induced anaemia. In contrast, the EA fraction at the higher dose showed a therapeutic effect by significantly improving WBC and lymphocyte counts, indicating its potential to modulate the immune system and reduce chronic inflammation associated with diabetes.

CONCLUSION

Failure of over-the-counter drugs in the management of patients with high blood sugar level (diabetes) has continued to draw attention to research into medicine plants as alternative to orthodox drugs. Medicinal plants possess phytochemical compounds depending of the extracting solvent used. Our study confirms that *E. chlorantha* stem bark extracts possess significant antidiabetic and hepatoprotective properties. The extracts not only effectively reduced hyperglycaemia but also ameliorated the associated haematological complications, which is a crucial aspect for preventing long-term cardiovascular risks in diabetic patients. The differential effects of the HE and EA fractions suggest that the various phytochemicals within the plant, such as flavonoids and alkaloids, work synergistically to provide a holistic therapeutic effect against the multifaceted pathology of diabetes. Future research should focus on isolating the specific compounds responsible for these effects and elucidating their precise mechanisms of action.

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