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Original Research Article

Evaluation of Antioxidant and Anti-Diabetes Properties of *Celba pentandra* L. Gaertn. on *Drosophila melanogaster*

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Abstract

Antioxidants are bioactive molecules that safeguard cells against oxidative stress, a condition that induces cellular injury and is implicated in the development of various chronic diseases. Diabetes mellitus is a long-term metabolic disorder marked by persistent hyperglycemia resulting from inadequate insulin secretion, impaired insulin action, or both. This study aimed to investigate the *in vitro* antioxidant and antidiabetic potentials of the methanolic leaf extract of *C. pentandra* using an experimental model of *Drosophila melanogaster* subjected to a high-sucrose diet. The antioxidant potential of the extract was evaluated using several assays, including 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging activity, ferric reducing antioxidant power (FRAP), lipid peroxidation inhibition, reducing power capacity, and reduced glutathione (GSH) quantification. The antidiabetic activity of the plant was assessed through *in vitro* α-amylase inhibition and *in vivo* analysis using *D. melanogaster* exposed to a high-sucrose diet. A total of thirty-five (35) *D. melanogaster* were utilized, divided into five experimental groups. Metformin (16 mg) was employed as the reference drug, while *C. pentandra* extract was administered at concentrations of 2 and 4 mg. The experiment was conducted over a ten-day period, after which the flies were analyzed for glucose and total protein levels. The *in vitro* antioxidant evaluation of *C. pentandra* methanolic extract demonstrated substantial activity across all assays, with notably higher lipid peroxidation inhibition (84.73%) and reducing power (20.52%) compared to the standard antioxidant, butylated hydroxytoluene (BHT) (50.13% and 17.26%, respectively) at 500 µg/mL. The extract exhibited a concentration-dependent increase in α-amylase inhibitory activity (21.83%, 22.55%, and 23.60%), although the values remained lower than those of the standard drug, acarbose (52.76%, 54.20%, and 68.04%) at 500, 750, and 1000 µg/mL, respectively. These findings suggest that the extract's ability to modulate glucose metabolism may improve with increasing concentrations. In *in vivo* assays, metformin produced the lowest glucose concentration (40 mg/dL), while the 4 mg *C. pentandra* extract-treated group showed reduced glucose levels (60 mg/dL) compared to the 2 mg group (150 mg/dL). Protein concentration analysis revealed no significant differences ($P < 0.05$) among the extract-treated groups, whereas both the normal (non-diabetic) and metformin-treated controls exhibited similar protein levels (80 mg/dL). Overall, *C. pentandra* methanolic extract demonstrated potent lipid peroxidation inhibition and superior reducing power, indicating its potential to mitigate oxidative stress. Furthermore, its glucose-lowering effect suggests a promising antidiabetic activity, possibly through enhanced insulin sensitivity.

Keywords: Antioxidant, Diabetes mellitus, *Ceiba pentandra, Drosophila melanogaster*.

INTRODUCTION

Antioxidants are bioactive molecules that protect cellular components from oxidative damage induced by free radical formation. Oxidation involves the transfer of electrons from a donor molecule to an oxidizing agent, a process that can generate highly reactive free radicals (Teresa *et al.*, 2011). These reactive species contain one or more unpaired electrons in their outermost orbitals, enabling them to initiate chain reactions that damage lipids, proteins, and nucleic acids. Antioxidants neutralize these radicals by donating electrons or hydrogen atoms, thereby terminating the chain reactions and preventing further oxidative injury. Living organisms possess intricate antioxidant defense systems comprising non-enzymatic compounds, such as vitamins C and E, and enzymatic antioxidants including catalase (CAT), superoxide dismutase (SOD), and peroxidases (Hamid *et al.*, 2010). Although oxidative processes are essential for numerous physiological functions, excessive oxidation results in oxidative stress, which disrupts cellular homeostasis and contributes to the pathogenesis of several diseases. Persistent oxidative stress has been implicated in the onset and progression of diverse pathological conditions, including diabetes

mellitus, cardiovascular disorders, neurodegenerative diseases such as Parkinson's and Alzheimer's, cancer, inflammatory diseases, hepatic dysfunction, and the aging process (Amit and Priyadarsini, 2011).

The term *diabetes mellitus* originates from the Greek word *diabetes*, meaning "to pass through," and the Latin word *mellitus*, meaning "sweet". Diabetes mellitus is a metabolic disorder characterized by chronically elevated blood glucose levels resulting from defects in insulin secretion, insulin action, or both. According to Andrew *et al.,* (2018), it represents a complex metabolic condition of chronic hyperglycemia associated with disturbances in carbohydrate, protein, and lipid metabolism, often leading to dysfunction in multiple organ systems.

The major forms of the disease include Type 1 diabetes, previously termed insulin-dependent diabetes mellitus (IDDM) or juvenile-onset diabetes, which results from autoimmune destruction of pancreatic β-cells and consequent insulin deficiency; Type 2 diabetes, which accounts for approximately 90% of all cases, characterized by insulin resistance and impaired insulin secretion; and gestational diabetes, which arises during pregnancy and typically resolves after childbirth. Clinical manifestations commonly include polyuria, polydipsia, polyphagia, blurred vision, and unexplained weight loss. In severe cases, diabetic ketoacidosis may develop, leading to stupor, coma, and, without prompt intervention, death.

Conventional therapeutic management involves exogenous insulin administration for Type 1 diabetes and oral hypoglycemic agents, such as biguanides and sulfonylureas, for Type 2 diabetes (Chijioke *et al.*, 2010). Although these treatments effectively regulate blood glucose levels, they often fail to prevent long-term complications and may produce adverse effects or diminished efficacy with prolonged use. This has prompted increasing interest in plant-derived bioactive compounds as alternative or complementary therapies for diabetes management.

Medicinal plants remain a vital source of therapeutic agents in both traditional and modern medicine. Approximately 80% of rural populations rely on plant-based remedies as their primary form of healthcare (Akinyemi, 2000). Historically, plants have served as fundamental sources of treatment for numerous ailments across all continents, particularly in Africa, where traditional medicine continues to play a significant role. Despite advances in synthetic drug development during the twentieth century, more than 25% of prescribed pharmaceuticals in industrialized nations are derived directly or indirectly from plant sources (Newman *et al.*, 2000).

Ceiba pentandra (L.) Gaertn., commonly known as the White Cotton Tree or Kapok Tree, belongs to the family Malvaceae. It is native to Central and South America, Africa, and Southeast Asia, and is widely distributed across tropical regions, including the warmer parts of India. The species is characterized by a massive trunk, buttressed roots, and flowers that range in color from white and cream to pink or red. Flowering and fruiting typically occur between January and June, although some trees may remain non-flowering for 5–10 years. Various parts of *C. pentandra*—including the leaves, fruits, roots, bark, and flowers—have been reported to possess diverse medicinal properties. The plant is traditionally used in the treatment of diabetes, inflammation, asthma, tumors, cough, and fever. Phytochemical analysis have identified the presence of phenols, alkaloids, flavonoids, tannins, glycosides, saponins, resins, and terpenoids, which contribute to its pharmacological activities.

Previous studies have demonstrated the plant's cardioprotective, gastroprotective, hepatoprotective, antiallergic, antimicrobial, and antidiarrheal properties (Elumalai *et al.*, 2012; Peter and Adebayo, 2012). Given these diverse bioactivities, *C. pentandra* represents a promising source of natural compounds for therapeutic applications. Therefore, the present study aimed to evaluate the *in vitro* antioxidant and antidiabetic potentials of the methanolic leaf extract of *C. pentandra* using *Drosophila melanogaster* exposed to a high-sucrose diet as an experimental model.

MATERIALS AND METHODS

Collection of Plant Material

Fresh leaves of *Ceiba pentandra* were collected from the Cocoa Research Institute of Nigeria (CRIN), Ibadan (latitude 7.4740° N, longitude 5.7379° E). The plant was taxonomically identified and authenticated by Dr. O. A. Obembe, a plant taxonomist in the Department of Plant Science and Biotechnology, Adekunle Ajasin University, Akungba-Akoko, Ondo State, Nigeria. A voucher specimen (No. PSBH-269) was prepared and deposited in the Departmental Herbarium for future reference.

Preparation of Plant Extract

Fresh *C. pentandra* leaves (1 kg) were carefully washed, air-dried, and ground using a mechanical grinder. The resulting powder was soaked in methanol for seven days, with occasional stirring. After maceration, the mixture was filtered through muslin cloth, and the filtrate was left to evaporate at room temperature until dry. The dried methanolic extract was then stored in a desiccator until needed for experimental procedures.

In vitro Antioxidant Assay

Antioxidant activity of the plant extract was evaluated using assays for reduced glutathione levels, 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging activity, ferric reducing antioxidant power

(FRAP), lipid peroxidation, and reducing power capacity.

Determination of reduced glutathione level

The reduced glutathione (GSH) level was determined following the method of Beutler *et al.,* (2017). *C. pentandra* methanolic extract (50 mg) was dissolved in 5 mL of dimethyl sulfoxide (DMSO), thoroughly mixed, and used for the assay. Glutathione standard solutions (100–400 µL) were pipetted into separate test tubes, and the volume in each tube was adjusted to 1 mL with distilled water. Subsequently, 3 mL of phosphate buffer was added to each tube and mixed thoroughly. Thereafter, 0.5 mL of 5,5'-dithiobis-(2-nitrobenzoic acid) (DTNB; Ellman's reagent) was added to all tubes, followed by incubation at room temperature for 5 minutes. Absorbance was recorded at 412 nm within 10 minutes. The same procedure was repeated for the *C. pentandra* extract (100 µL), and absorbance was similarly measured at 412 nm.

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

The DPPH radical scavenging activity of the *Ceiba pentandra* methanol extract was determined following the procedure described by Omoboyowa *et al.,* (2020), with minor modifications. Briefly, 1 mL of butylated hydroxytoluene (BHT) solution at concentrations ranging from 250 to 1000 µg/mL was mixed with 1 mL of DPPH solution (2.4 mg of DPPH dissolved in 200 mL of methanol). The resulting mixture was vortexed thoroughly and incubated in the dark for 30 minutes. The absorbance was subsequently measured at 517 nm, and the percentage of DPPH inhibition was calculated using the following formula:

$$Percentage\ DPPH\ inhibition = \frac{Absorbance\ of\ control\ -\ Absorbance\ of\ sample}{Absorbance\ of\ control} \times 100$$

Determination of Ferric Reducing Antioxidant Potential (FRAP)

The ferric reducing antioxidant power (FRAP) assay of the *Ceiba pentandra* methanol extract was performed following the method described by Benzie and Strain (1996). The freshly prepared FRAP working reagent consisted of 25 mL of acetate buffer, 2.5 mL of 2,4,6-tripyridyl-s-triazine (TPTZ) solution, and ferric chloride solution in a 10:1:1 ratio. The reagent mixture was warmed to 37°C and subsequently heated at 100°C for 15 minutes. After cooling, the solution was centrifuged for 10 minutes. Then, 0.2 mL of the *C. pentandra* methanol extract was mixed with 2.8 mL of the FRAP reagent and incubated in the dark for 30 minutes at 27°C. The absorbance was measured at 593 nm, and the FRAP value was determined from a ferrous sulfate calibration curve, expressed as mM Fe^{2+} equivalents.

Estimation of Lipid Peroxidation

The lipid peroxidation inhibitory activity of the *C. pentandra* methanol extract was assessed following the method described by Ruberto *et al.,* (2000), with slight modifications. Briefly, 300 µL of the *C. pentandra* methanol extract or the standard antioxidant (BHT) was mixed, in triplicate, with 500 µL of egg homogenate (10% v/v in phosphate-buffered saline, pH 7.4) and the volume was adjusted to 1.0 mL with distilled water. Subsequently, 50 µL of $FeSO_4$ (0.075 M) and 20 µL of L-ascorbic acid (0.1 M) were added sequentially to initiate lipid peroxidation. The reaction mixture was incubated at 37°C for 1 hour. Thereafter, 0.2 mL of ethylenediaminetetraacetic acid (EDTA, 0.1 M) and 1.5 mL of thiobarbituric acid (TBA) reagent were added. The mixture was centrifuged at 3000 × g, and the absorbance of the supernatant was measured at 532 nm against a blank. Control tubes containing all reagents except the plant extract were also prepared. The percentage inhibition of lipid peroxidation was calculated using the following equation:

$$\text{Scavenging activity} = \frac{\text{Absorbance of control} - \text{Absorbance of sample}}{\text{Absorbance of control}} \times 100$$

Estimation of Reducing Power Ability

The reducing power of the *Ceiba pentandra* methanol extract was evaluated following the method described by Omoboyowa *et al.,*(2020), with minor modifications. Aliquots of the extract and the standard antioxidant, butylated hydroxytoluene (BHT), at concentrations of 500, 750, and 1000 µg/mL, were prepared in triplicate test tubes. Methanol was added to each tube to obtain a final volume of 1 mL, while a separate tube containing only methanol served as the blank. Subsequently, 2.5 mL of phosphate buffer (0.2 M, pH 6.6) and 2.5 mL of potassium ferricyanide (1%) were added sequentially to each tube, including the blank. The mixtures were incubated at 50°C for 20 minutes, after which 2.5 mL of trichloroacetic acid (10%) was added. The resulting solutions were centrifuged at 3000 × g for 10 minutes at room temperature. From each supernatant, 2.5 mL was transferred into a new tube, mixed with 2.5 mL of distilled water, and 0.5 mL of ferric chloride (0.1%) was added. The absorbance of the resulting solution, exhibiting a green coloration, was measured at 700 nm. An increase in absorbance indicated greater reducing power.

In Vitro Anti-Diabetes Assay

The in vitro antidiabetic potential of the *C. pentandra* methanol extract was evaluated using the α-amylase inhibition assay.

Determination of α amylase inhibition assay

The α-amylase inhibitory activity of the *C. pentandra* methanol extract was evaluated using the 3,5-dinitrosalicylic acid (DNSA) method as described by Worthington (1993) and Wickramaratne *et al.,* (2016), with minor modifications. The extract was dissolved in 10% dimethyl sulfoxide (DMSO) to prepare the desired concentrations. A reaction mixture containing 500 µL of the extract and 500 µL of 0.02 M sodium phosphate buffer (pH 6.9, containing 0.006 M NaCl) with 0.5 mg/mL α-amylase solution was incubated at room temperature for 10 minutes. Subsequently, 500 µL of 1% (w/v) starch solution prepared in the same buffer was added, and the mixture was further incubated at 25°C for 10 minutes. The reaction was terminated by adding 200 µL of DNSA reagent (composed of 12 g sodium potassium tartrate tetrahydrate in 8 mL of 2 M NaOH and 20 mL of 96 mM 3,5-dinitrosalicylic acid solution), followed by boiling for 5 minutes in a water bath at 85–90°C. After cooling to room temperature, the mixture was diluted with 5 mL of distilled water, and the absorbance was measured at 540 nm using a UV–Visible spectrophotometer.

A control representing 100% enzyme activity was prepared by replacing the extract with 200 µL of buffer. A corresponding blank was prepared for each concentration using the extract in the absence of the enzyme solution. Acarbose (2–100 µg/mL) served as the positive control. The α-amylase inhibitory activity was expressed as the percentage of inhibition, calculated using the following equation:

$$\text{Inhibition percentage} = \frac{\text{Absorbance of control} - \text{Absorbance of sample}}{\text{Absorbance of control}} \times 100$$

In Vivo Anti-Diabetes Assay

Culture of fly stock

The *Drosophila melanogaster* (Harwich strain) used in this study was obtained from the *Drosophila* Laboratory, Department of Biochemistry, University of Ibadan, Oyo State, Nigeria. The flies were maintained at 25 ± 2°C in the Phytomedicine and Functional Food Laboratory, Department of Biochemistry, Adekunle Ajasin University, Akungba-Akoko (AAUA). The culture medium consisted of cornmeal, agar-agar, brewer's yeast, and Nipagin (as a preservative).

Induction of diabetes in flies using sucrose

To induce type 2 diabetes in Drosophila melanogaster (Harwich strain), the protocol of Omale *et al.,* (2021) and Omoboyowa *et al.,* (2021) were followed with minor adjustments. The normal fly diet was supplemented with 2.5 g sucrose per 10 g of food, while maintaining constant levels of other components (1% agar, 3.4% yeast, 8.3% corn meal, and 1% nipagin). Flies were exposed to the sucrose- enriched diet for 10 days, after which diabetes-like symptoms such as reduced (L3) larvae emergence rate, decreased body size and impaired locomotor activity were observed.

Locomotor assay

The negative geotaxis assay was employed to evaluate the locomotor performance of *Drosophila melanogaster*. A total of thirty-five (35) flies from both the control and experimental groups were used. The flies were briefly anesthetized on ice and subsequently transferred into a 15 × 15 cm glass column. After full recovery, the flies were gently tapped to the bottom of the column, and the number of flies that successfully climbed to the 6 cm mark within a fixed time interval was recorded. The negative geotaxis index was calculated using the following formula:

$$\text{Percentage (\%) negative geotaxis} = \frac{\text{Total number of flies} - \text{Number of flies that climb above 6 cm}}{\text{Total number of flies}} \times 100$$

Treatment of sucrose-induced (diabetic) flies

Thirty-five (*Drosophila melanogaster*) flies per group, in triplicate, were assessed for antidiabetic activity as described below:

- **Group 1 (Normal control):** Non-diabetic flies maintained on a standard diet.
- **Group 2 (Negative control):** Diabetic flies fed with 10 mg dimethyl sulfoxide (DMSO) per 10 g of diet.
- **Group 3 (Positive control):** Diabetic flies treated with 16 mg metformin per 10 g of diet.
- **Group 4 (Treatment 1):** Diabetic flies treated with 2 mg *Ceiba pentandra* methanol extract per 10 g of diet.
- **Group 5 (Treatment 2):** Diabetic flies treated with 4 mg *C. pentandra* methanol extract per 10 g of diet.

After 10 days of treatment, the flies were anesthetized with ethanol, blotted dry, and weighed. Whole-fly homogenates were prepared in 0.1 M phosphate buffer (pH 7.0) at a 1:10 ratio (1 mg of flies per 10 mL of buffer). The homogenates were centrifuged at 4000 × g for 10 minutes, and the resulting supernatants

were collected for in vitro antioxidant assays and glucose concentration determinations.

Estimation of Glucose Concentration

Glucose concentration in *Drosophila melanogaster* homogenates was determined using the LiquiCHEK Glucose Kit (Agappe), according to the method described by Trinder (1969). This assay is based on the enzymatic oxidation of glucose-by-glucose oxidase, producing hydrogen peroxide, which subsequently reacts with phenol and 4-aminoantipyrine in the presence of peroxidase to yield a pink-colored quinoneimine complex.

For the analysis, 10 µL of fly homogenate supernatant was added to 1 mL of reagent solution and incubated at 37°C for 10 minutes. The absorbance of the resulting solution was measured at 505 nm against a reagent blank using a UV–Visible spectrophotometer. Glucose concentrations were calculated from a standard calibration curve prepared using known concentrations of glucose and expressed as millimoles of glucose per gram (mmol/g) of tissue.

Estimation of Protein Concentration

A series of test solutions of the *C. pentandra* methanol extract were prepared at concentrations of 40, 60, and 80 µg/mL. To each sample, 200 µL of 1 M Folin–Ciocalteu reagent was added, followed by thorough mixing and incubation at room temperature for 30 minutes. The absorbance of the resulting blue-colored complex, corresponding to the concentration of reduced Folin reagent, was measured at 750 nm using a UV–Visible spectrophotometer (Yulia *et al*., 2025). The total phenolic content was subsequently quantified from a gallic acid standard calibration curve and expressed as milligrams of gallic acid equivalents (mg GAE) per gram of extract.

Statistical Analysis

All experimental data were expressed as mean ± standard error of the mean (SEM). Statistical analysis was performed using the Statistical Package for the Social Sciences (SPSS) software, version 22. Differences among group means were analyzed using one-way analysis of variance (ANOVA), followed by Duncan's New Multiple Range Test (DNMRT) for post hoc comparison of means. Statistical significance was considered at $p < 0.05$.

RESULTS

Antioxidant Assay

Reduced glutathione level of *C. pentandra* methanol extract on *Drosophilia melanogaster*

Figure 1 shows result on reduced glutathione level of *C. pentandra* methanol extract on *D. melanogaster*. The result revealed significant reduction of glutathione level in metformin treated group (group 3; 15 nM/ml) when compared to all other groups, while negative control (group 2) showed highest level of glutathione (25 nM/ml). No significant difference ($P< 0.05$) was seen in the activities of groups 4 (21 nM/ml) and 5 (22 nM /ml).

DPPH (1, 1-diphenyl-2-picrylhydrazyl) inhibition activity of *C. pentandra*

Figure 2 shows the results of DPPH inhibition activity of *C. pentandra* methanol extract with butylated hydroxytoluene (BHT) used as the standard. The results showed 36.31%, 30.90% and 22.82% in 500, 750 and 1000 ug/ml respectively but highest percentage of DPPH inhibition was shown in standard, BHT as 82.07%, 82.78% and 83.04% at 500, 750 and 1000 ug/ml respectively.

Ferric-reducing antioxidant power (FRAP) of *Ceiba pentendra* extract

Figure 3 shows the results of ferric-reducing antioxidant power (FRAP) of *C. pentandra* methanol leaves extract with butylated hydroxytoluene. (BHT) used as the standard. The results showed lowest FRAP activity in *C. pentandra* as 11.14% at 500 ug/ml while BHT had 36.26%.

Lipid peroxidation scavenging activity of *Ceiba pentandra* extract

Figure 4 shows the result of lipid peroxidation scavenging activity of *C. pentandra* methanol extract with BHT used as standard. *C. pentandra* indicated higher percentage (84.73%) of scavenging activity than the standard (50.13%).

Reducing power ability of *Ceiba pentandra* extract

Figure 4 shows the results of the reducing power ability of *C. pentandra* leaves methanol extract with BHT used as standard. The results reviewed that BHT (17.26% at 500 ug/ml, 17.15% at 750 ug/ml, 17.12% at 1000 ug/ml) had lowest activity when compared to *C. pentandra* (20.52% at 500 ug/ml, 20.76% at 750 ug/ml, 20.82% at 1000 ug/ml) across the concentration.

***In Vitro* Anti-Diabetes Results**

Inhibitory activity of alpha-amylase of *Ceiba pentandra* extract

Figure 6 shows the inhibitory activity of alpha-amylase of *C. pentandra* extract in different concentrations. The results revealed that the activity of *C. pentandra* (21.83% at 500 ug/ml, 22.55% at 750 ug/ml, 23.6% at 1000 ug/ml) was significantly lower than the standard, acarbose (52.76% at 500ug/ml, 54.2% at 750ug/ml and 68.04% at 1000ug/ml).

***In Vivo* Anti-Diabetes Assay Results**

Locomotor assay of *C. pentandra* methanol extract

Result of locomotor assay using negative geotaxis of *C. pentandra* methanol extract on sucrose induced diabetic flies is shown in Figure 7. This result showed that group 3 (Positive control: 16 mg metformin) had highest negative geotaxis (70%) while group 2

(Negative control: 10 mg DMSO) gave lowest negative geotaxis (18%) when compared to normal control group (90%). Group 5 (4 mg *C. pentandra* methanol extract) revealed higher locomotor activity (38%) than group 4 (2 mg *C. pentandra* methanol extract) (27%).

Glucose concentration of *C. pentandra* methanol extract on *Drosophila melanogaster*

Figure 8 shows total glucose level of sucrose-induced *D. melanogaster* treated with *C. pentandra* methanol extract. Negative control (group 2; 175 mg/dl) showed increase in glucose level (P< 0.05) compared to the normal control (group 1; 70 mg/dl). Positive control (group 3; 40 mg/dl) gave significant lowest glucose concentration (P > 0.05) when compared to all other groups while 4 mg *C. paradisi* methanol extract (group 5; 60 mg/dl) had lower level of glucose compared to 2 mg *C. paradisi* methanol extract (group 4; 150 mg/dl).

Protein concentration of *Ceiba pentandra* methanol extract on *Drosophila melanogaster*

Protein concentration's result of *C. pentandra* methanol extract on *D. melanogaster* is shown in Figure 9. There was no significant difference (P< 0.05) in level of protein in the extract treated groups but normal and positive controls (groups 1 and 2) had the same concentration of 80 mg/dl.

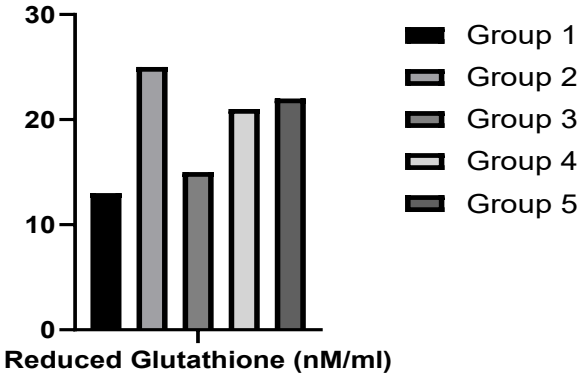

Figure 1: Reduced glutathione level (nM/ml) of *C. pentandra* methanol extract on *D. melanogaster.* Group 1= Normal control; Group 2 = Negative control; Group 3 = Positive control; Group 4 = 2 mg *C. paradisi* extract; Group 5 = 4 mg *C. paradisi* extract.

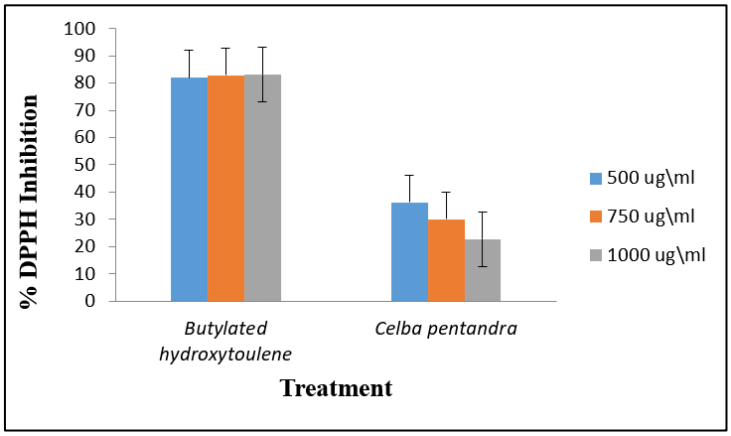

Figure 2: DPPH inhibition of *C. pentandra* methanol extract

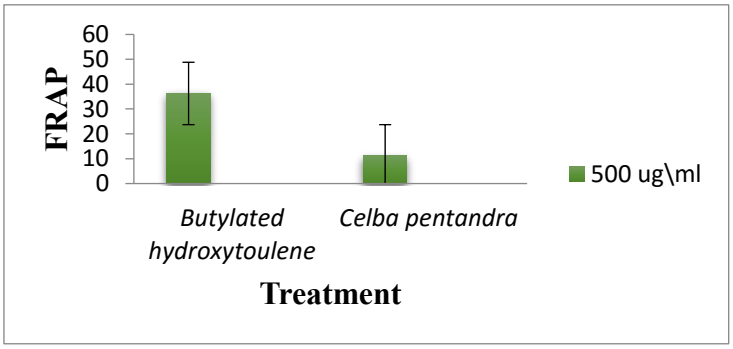

Figure 3: Ferric reducing antioxidant power (FRAP) of *C. pentandra* methanol extract

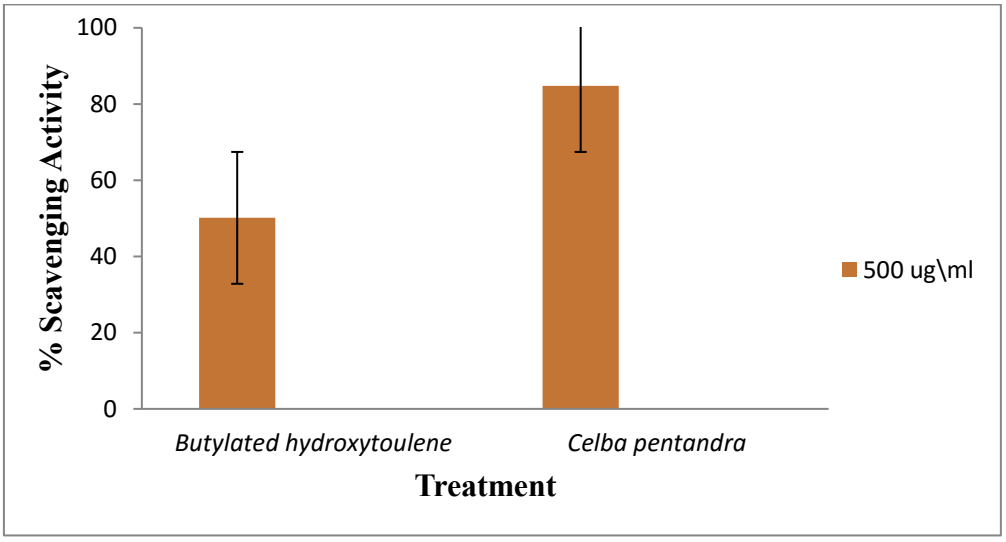

Figure 4: Lipid peroxidation scavenging activity of *C. pentandra* methanol extract

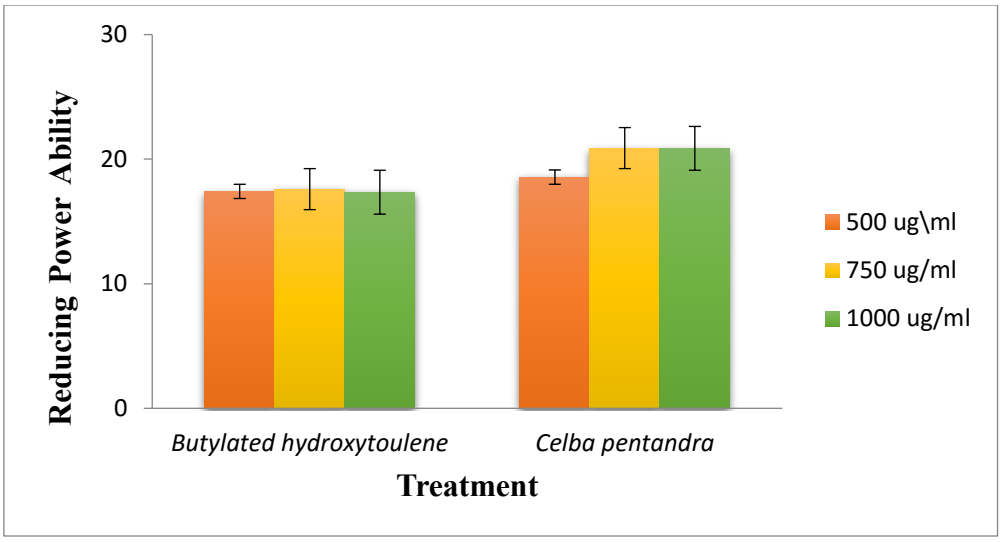

Figure 5: Result of reducing power ability of *C. pentandra* methanol extract

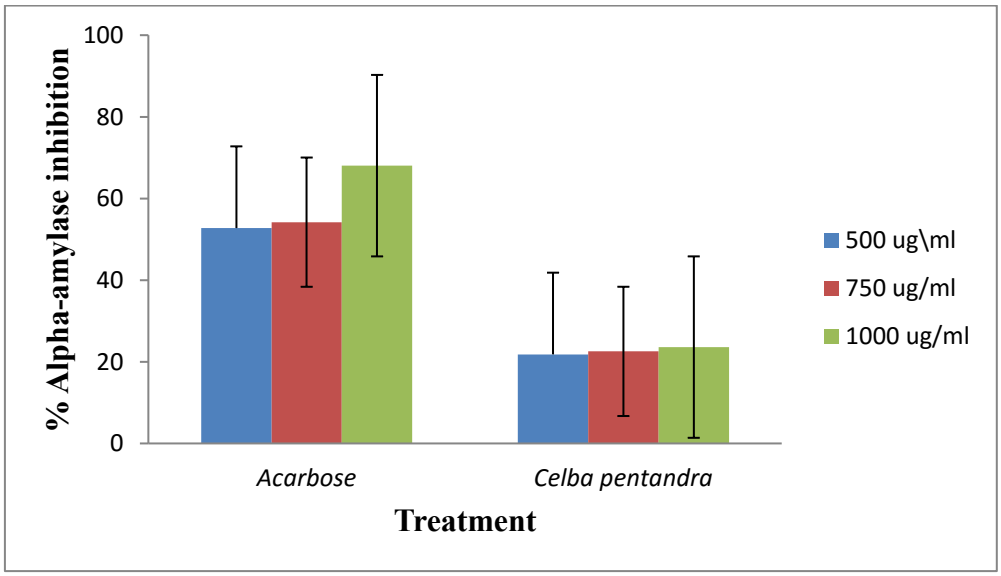

Figure 6: Inhibitory activity of alpha-amylase on *C. pentandra* methanol extract

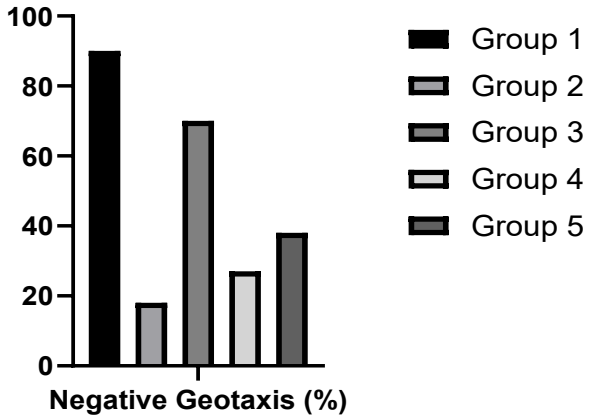

Figure 7: Result on locomotor assay using negative geotaxis (%) of *C. pentandra* methanol extract on sucrose induced diabetic flies. Group 1= Normal control; Group 2 = Negative control; Group 3 = Positive control; Group 4 = 2 mg *C. paradisi* extract; Group 5 = 4 mg *C. paradisi* extract

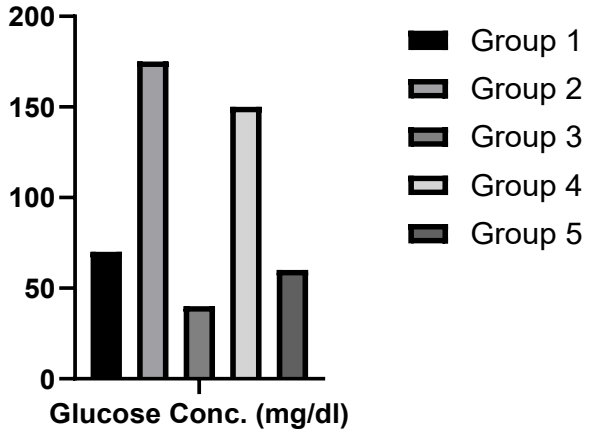

Figure 8: Total Glucose Level of Sucrose-induced *D. melanogaster* treated with *Ceiba pentandra* methanol extract. Group 1= Normal control; Group 2 = Negative control; Group 3 = Positive control; Group 4 = 2 mg *C. paradisi* extract; Group 5 = 4 mg *C. paradisi* extract

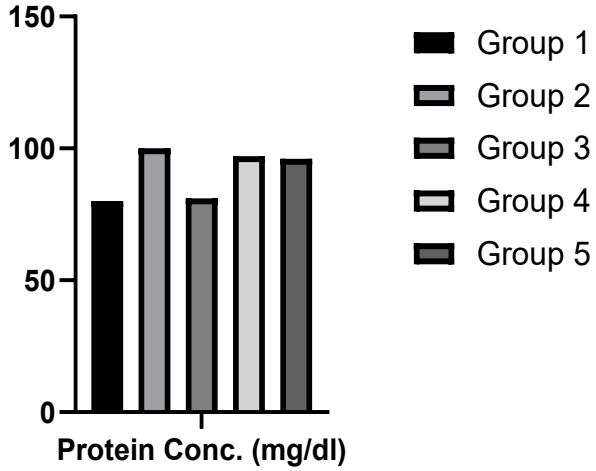

Figure 9: Result on protein concentration (mg/dl) of *Ceiba pentandra* methanol extract on *Drosophila melanogaster*. Group 1= Normal control; Group 2 = Negative control; Group 3 = Positive control; Group 4 = 2 mg *C. paradisi* extract; Group 5 = 4 mg *C. paradisi* extract

DISCUSSION

Diabetes mellitus is a metabolic disorder characterized by elevated blood glucose levels resulting from insufficient insulin secretion or impaired insulin action (Belsti *et al*., 2019). High-sucrose-induced diabetes is a form of diabetes mellitus that develops following excessive sucrose consumption, leading to insulin resistance, glucose intolerance, and pancreatic β-cell dysfunction (Kumar *et al*., 2016).

Oxidative stress plays a critical role in the pathogenesis of diabetes and its complications. Antioxidants act as key defense molecules that neutralize free radicals, thereby protecting cells from oxidative damage. Excessive free radical production can disrupt cellular function, induce inflammation, and contribute to the development of chronic diseases such as diabetes, cancer, and cardiovascular disorders.

Medicinal plants continue to serve as essential sources of therapeutic compounds for managing metabolic diseases, including diabetes. *Ceiba pentandra* has long been utilized both as a medicinal and dietary plant to promote health and manage various ailments.

The reduced glutathione level analysis (in vivo antioxidant assay) revealed varying effect of *C. pentandra* methanol extract on *D. melanogaster*. Positive control group (16 mg metformin) exhibited a significant reduction in glutathione level compared to all other groups, indicating reduction of oxidative stress. Negative control group (untreated diabetic group) showed the highest glutathione level, suggesting enhanced oxidative stress. No significant difference was observed in glutathione levels between 2 and 4 mg of *C. pentandra* methanol extract but the extract was able to shut down oxidative stress to a considerable level when compared to negative control group. The in vitro antioxidant study of *C. pentandr*a extract revealed its potency in all the methods used but higher activities were observed in lipid peroxidation scavenging activity and reducing power ability than the standard, butylated hydroxytoluene (BHT). The higher values recorded showed that the extract had ability to inhibit lipid peroxidation and prevent oxidative damage better than BHT, standard. This finding underscored the importance of medicinal plant in combating oxidative stress-related lipid damage.

Ceiba pentandra methanol extract displayed concentration dependent increase pattern in alpha-amylase inhibitory study (in vitro anti-diabetes assay) but the values were lower than those observed in the standard, acarbose. This suggested that the extract might help modulate glucose metabolism better with increased concentrations.

C. pentandra extract was further evaluated for anti-diabetes activity in high sucrose induced diabetic *D. melanogaster*. The locomotor analysis revealed significant differences among the groups. Positive control group (16 mg metformin) exhibited the highest negative geotaxis, indicating enhanced locomotor and motor function. Negative control group (untreated diabetic group) showed the lowest negative geotaxis, suggesting impaired locomotor and motor functions, demonstrating the adverse effect of high sucrose fed diet on motor function when compared to the normal control (non-diabetic group). Treatment group 1 (4 mg *C. pentandra* methanol extract) exhibited significantly higher locomotor compared to treatment group 2 (2 mg *C. pentandra* methanol extract) suggesting the extract's potential to enhance movement and motor function. This displayed enhanced negative geotaxis, indicating improved motor function and demonstrating potential neuroprotective effect which could be attributed to increase in concentration of the extract. This showed a dose-dependent relationship between *C. pentandra* extract and locomotor activity. In glucose concentration analysis, negative control group (untreated diabetic group) exhibited elevated glucose levels compared to the normal control group (non-diabetic group) indicating hyperglycemia. Positive control group (16 mg metformin) gave the lowest glucose concentration level, confirming its efficacy as a standard anti-diabetes agent followed by treatment group 1 (4 mg *C. pentandra* methanol extract) and treatment group 2 (2 mg *C. pentandra* methanol extract). *Ceiba pentandra* methanol extract (4 mg) treated group showed significant reduction in glucose levels compared to 2 mg *C. pentandra* methanol extract treated group suggesting a dose dependent anti-diabetes potential. The findings of this study corroborated to the work documented by Kumar *et al.,*(2016).

In protein concentration, there were no significant differences in protein levels observed among the groups. The positive control group (16 mg metformin) exhibited the highest protein concentration indicating a potential biological activity. The normal control group (non-diabetic group) showed the lowest protein concentration, suggesting a possible suppressive effect.

CONCLUSION

The *C. pentandra* methanol extract exhibited effective inhibition of lipid peroxidation, superior reducing power ability and also decreased glutathione levels suggesting its potential role in reducing oxidative stress. The extract also demonstrated anti-diabetes effect by reducing glucose levels and improving insulin sensitivity.

It can therefore be concluded that the plant has promising antioxidant and anti-diabetes properties, making it a potential natural remedy for managing oxidative stress-related disorders and diabetes. However, more research is needed to fully explore its therapeutic potential.

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