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Cajanus cajan Leaves and Ginger Increase Antioxidant Defence in Pancreatic Diabetic Rats: An Immunohistochemical Study

(*DAUN UNDIS DAN JAHE MENINGKATKAN PERTAHANAN ANTIOKSIDAN PADA PANKREAS TIKUS DIABETES : STUDI IMUNOHISTOKIMIA*)

Tutik Wresdiyati[1*], Bella Dinar Fauqii Cahyani[1], Hamzah Alfarisi[1], Siti Sa'diah[1], Made Astawan[2]

[1]School of Veterinary Medicine and Biomedical Science,
IPB University, IPB Darmaga Campus-Bogor 16880, Indonesia
[2]Department of Food Science and Technology,
Faculty of Agricultural Technology, IPB University,
Jl. Agathis, IPB Darmaga Campus, Bogor
West Java 16880, Indonesia
+62 8626064; Email: tutikwr@apps.ipb.ac.id

ABSTRACT

Hyperglycemia in diabetes mellitus results in stress oxidative condition. This study aims to analyze the antioxidant defence in pancreatic experimental-diabetic rats treated with a combined extract of *Cajanus cajan* leaves and ginger, using an immunohistochemical technique for copper, zinc – superoxide dismutase (Cu,Zn-SOD). A total of 25 male *Sprague Dawley* rats were used in this study. The rats were randomly divided into five groups; normal control group (NC), diabetic group (DM), two groups DM treated with a different dose combination of *Cajanus cajan* leaf extract and ginger extract (DME and DMF), and DM treated with glibenclamide (DMG). This study used 110 mg/kg BW alloxan induction to obtain diabetic conditions, and the treatments of the extract were conducted for 28 days. The pancreatic tissues were processed using the paraffin standard method and an immunohistochemical technique using monoclonal antibody Cu,Zn-SOD. The results showed that a combination of *Cajanus cajan* leaf extract and ginger extract lowered the level of blood glucose, increased body weight, and increased the level of Cu,Zn-SOD content in pancreatic tissues of experimental-diabetic rats. This study concluded that combining *Cajanus cajan* leaf extract and ginger extract increased antioxidant defence in pancreatic organs of experimental-diabetic rats.

Keywords: Diabetes, *Cajanus cajan*, Cu,Zn-SOD, ginger

ABSTRAK

Hiperglikemia pada diabetes melitus mengakibatkan kondisi stres oksidatif. Penelitian ini bertujuan untuk menganalisis pertahanan antioksidan pada pankreas tikus model-diabetes yang diberi ekstrak kombinasi daun undis (*Cajanus cajan*) dan jahe, menggunakan teknik imunohistokimia untuk copper, zinc – superoxide dismutase (Cu,Zn-SOD). Sebanyak 25 ekor tikus *Sprague Dawley* jantan digunakan dalam penelitian ini. Tikus

secara acak dibagi menjadi lima kelompok; kelompok kontrol normal (NC), kelompok diabetes (DM), dua kelompok DM yang diobati dengan perbedaan dosis kombinasi ekstrak daun undis dan ekstrak jahe (DME dan DMF), dan kelompok DM yang diobati dengan glibenclamide (DMG). Penelitian ini menggunakan induksi aloksan 110 mg/kg BB untuk mendapatkan kondisi diabetes, dan perlakuan ekstrak dilakukan selama 28 hari. Jaringan pankreas diproses menggunakan metode standar parafin, dilanjutkan dengan teknik imunohistokimia menggunakan antibodi monoklonal Cu,Zn-SOD. Hasil penelitian menunjukkan bahwa kombinasi ekstrak daun undis dan ekstrak jahe menurunkan kadar glukosa darah, meningkatkan berat badan, dan meningkatkan kadar Cu,Zn-SOD pada jaringan pankreas tikus model-diabetes. Penelitian ini menyimpulkan bahwa kombinasi ekstrak daun undis dan ekstrak jahe meningkatkan pertahanan antioksidan pada organ pankreas tikus percobaan diabetes.

Kata-kata kunci: Cu,Zn-SOD, daun undis, diabetes, jahe

INTRODUCTION

An imbalance between free radicals and antioxidants in the body is known as oxidative stress. This situation occurs when overproduction of free radicals and less number of endogenous antioxidants. The oxidative stress then resulted in decreased antioxidant defence in the body. Oxidative stress occurs in diabetic patients with high blood glucose levels, known as hyperglycemia. Hyperglycemia in diabetic patients is caused by a lack of insulin secretion, decreased insulin sensitivity, or both (Park *et al.*, 2021). Hyperglycemia leads to oxidative stress by increasing production of reactive oxygen species (ROS), reactive nitrogen species (RNS), and reactive chlorine species (RCS) through several pathways: autooxidation of glucose, increase in glycolysis, activation of the sorbitol or polyol pathway, formation of advanced glycation end products (AGEs) and others, glycation of non-enzymatic protein, NADPH oxidase, lipid oxidation, hexosamine pathway (Yaribeygi *et al.,* 2019).

In oxidative stress conditions, abundant free radicals, existed which may damage biomacromolecules of cell's structure components such as proteins, lipids, carbohydrates, and DNA. Then it leads to the oxidative stress damage occured in several organs and subsequently leading to microvascular complications (neuropathy, cataracts, and retinopathy) including macrovascular complications (stroke and heart failure) (Asmat *et al.,* 2016; Sun *et al.,* 2021). Therefore, it is important to maintain a normal blood glucose level, balance antioxidants and free radicals, and increase the antioxidant defence in the body.

It was reported that diabetic condition in rats showed decreased antioxidant Cu,Zn-SOD in the pancreatic tissues (Wresdiyati *et al.,* 2015), liver and kidney tissues (Wresdiyati *et al.,* 2010), and testicular tissues (Wresdiyati *et al.,* 2020b). These findings showed that diabetic conditions decreased antioxidant defence in several organs.

Methanolic, ethanolic, and aqueous extracts of *Cajanus cajan* leaves exhibited antioxidant, hypoglycemic, and alpha-glucosidase inhibitory activities (Aja *et al.,* 2015; Mahitha *et al.,* 2015; Wresdiyati *et al.,* 2020a; Tungmunnithum *et al.,* 2021). Aqueous and methanolic extracts of ginger extract were also reported to exhibit antioxidant and hypoglycemic activities (Wresdiyati and Astawan, 2004; Wresdiyati *et al.,* 2007; Okafor and Okafor, 2022). Wresdiyati *et al.* (2020a) also reported combination of ethanolic *C. cajan*

leaves extract and ginger extract resulted in a hypoglycemic effect in experimental hyperglycemic rats. The hypoglycemic effect was greater than that of *C. cajan* leaves or ginger extract. Based on these results and concerning the severe effect of free radicals on oxidative damage on several organs, this study was performed and aimed to analyze the antioxidant defence in pancreatic tissues of experimental diabetic rats treated with the combination extracts and visualized using an immunohistochemical technique for antioxidant Cu,Zn-SOD content profile in the pancreatic tissues.

RESEARCH METHODS

Materials

Cajanus cajan leaves used in this study were obtained from Mataram of Lombok City, West Nusa Tenggara, while ginger (*Zingiber officinale*) var. amarum was obtained from Solo City, Central Java, Indonesia. Identification and confirmation of the *C. cajan* were made by the National Research and Innovation Agency (BRIN), Republic of Indonesia. The plants were harvested in January 2023. This study used male *Sprague Dawley*-four weeks old rats.

This study used chemicals include 96% ethanol, alloxan (Sigma-Aldrich, US), Accu-Check strips, and glucometer (Roche, Germany), physiological NaCl, ketamine, xylazine, glacial acetic acid (Merck, Germany), picric acid (Sigma Aldrich, USA), absolute ethanol (Merck, Germany), xylol (Merck, Germany), paraffin, toluene (Merck, Germany), neofren®, 30% Hydrogen Peroxide (AR) (Smartlab A-1052, Indonesia), normal goat serum 10%, primary Cu,Zn-SOD-monoclonal antibody (Sigma-Aldrich, US, S2147), Starr Trek Universal Horseradish peroxidase (HRP) Detection System (Biocare Medical STU700H-KIT), hematoxylin dye, milli-q, entellan® (Merck, Germany).

Extraction of Pigeon Pea Leaves and Ginger

The *C. cajan* leaves and ginger var. amarum rhizome were dried in the oven at 45°C for two days and one night, then they were grounded and filtered separately at a size of 80 mesh. The powdered pigeon pea leaves and ginger were extracted using 96% ethanol and maceration methods separately, as described by Wresdiyati *et al.* (2015a; 2020a). The two extracts were evaporated separately to get two dry extracts.

Preparation and Treatment of The Combination of Pigeon Pea Leaf and Ginger Extracts in Rats.

The experimental procedures used in this study followed the Ethical Approval Letter from the Animal Ethic Committee School of Veterinary Medicine and Biomedical Science, IPB University, number 040/KEH/SKE/X/2022. The rats were obtained from the National Agency for Drug and Food Control of Indonesia. The rats were adapted for seven days by feeding them the rat's standard feed (AOAC 1995) and given drinking water *ad libitum*. To ensure their health, the rats were administered vitamins, antibiotics, and antiparasitic treatments. Rats were conditioned in the conditions: 22-24°C, 50-63% humidity, and 12h bright light-dark cycle. This study used 25 male four-week-old male *Sprague-Dawley* rats, divided into five groups; normal control group (NC), diabetic group (DM), DM treated with glibenclamide (DMG), and two DM groups treated orally with 300 mg/kg BW of pigeon pea leaf extract and 60 mg/kg BW of ginger extract group (DME), and DM group treated orally with 300 mg/kg BW of pigeon pea leaf extract and 125 mg/kg BW of ginger extract (DMF). The diabetic condition was obtained using alloxan induction (110 mg/kg BW) (Wresdiyati *et al.,* 2016).

All rats groups (NC, DM, DME, DMF, and DMG) were treated for 28 days (Wresdiyati *et al.,* 2016). Feed consumption was measured every day. The blood glucose level was measured every four days using a glucometer and Accu-Check strips. Bodyweight was also measured every four days (Wresdiyati *et al.,* 2016). These parameters were measured on specific days to monitor the change periodically.

Sampling Pancreatic Organs

The rats were anaesthetized using ketamine (70 mg/kgBW) and xylazine (10 mg/kgBW) and euthanized by exsanguination. Rats were necropsied and pancreatic tissues were collected for histological analysis. The pancreatic tissues were fixated using Bouin solution, followed by a stopping point using 70% ethanol. The pancreatic tissues were then processed using the paraffin standard method (Kiernan 2013). The cooper, zinc-superoxide dismutase (Cu,Zn-SOD) in the pancreatic tissues were immunohistochemically stained using a Cu,Zn-SOD-monoclonal antibody (Sigma-Aldrich, US, S2147) and Starr Trek Universal HRP Detection System (Biocare Medical STU700H-KIT) for Cu,Zn-SOD identification. The positive reaction of Cu,Zn-SOD in the acinar cells and Langerhans islet cells was microscopically visualized with brown colour (Wresdiyati *et al.,* 2016).

Data Analysis

The immunohistochemical profile of Cu,Zn-SOD content in pancreatic tissues was observed using a light microscope (Olympus CH20) and documented using a camera microscope (Olympus CX31-CCD10 USB Camera). The Cu,Zn-SOD positive immunoreactions were categorized into three levels: a strong positive reaction (+++), indicated by a dark brown color throughout the entire cell nucleus; a moderate positive reaction (++), indicated by a dark brown color in certain areas of the cell nucleus; and a weak positive reaction (+), indicated by a light brown color in the cell nucleus. The negative immunoreaction (-) was characterized by a blue color in the cell nucleus. The number of positive and negative immunoreactions of pancreatic acinar cells and pancreatic Langerhans islet cells were counted and analyzed using ImageJ software. Blood glucose level and body weight were analyzed qualitatively. The total food consumption and the number of immunoreactions of pancreatic acinar cells and pancreatic Langerhans islet cells were analyzed using the software SPSS 16 using the one-way analysis of variance method. If there was a significant difference, Duncan's test was performed.

RESULTS AND DISCUSSION

The amount of feed consumption of the treated rats (Table 1) showed no significant difference between the treatment groups ($P>0.05$). The body weight growth of the diabetic rat groups showed a different growth profile during the 28-day treatment period (Figure 1). The diabetic rats (DM) not given glibenclamide or the combination extracts showed the lowest body weight growth profile. The group of diabetic rats orally treated with 300 mg/kg BW of *C. cajan* leaves extract and 60 mg/kg BW of ginger extract group (DME) showed the best bodyweight growth profile compared to other diabetic groups, followed by the group of diabetic rats orally treated with 300 mg/kg BW of *C. cajan* leaves extract and 125 mg/kg BW of ginger extract group (DMF) and the diabetic group which was given with glibenclamide (DMG). These results showed the effectiveness of the combination of *C. cajan* leaves extract and ginger extracts in the growth of body weight of diabetic rats.

Table 1. Total feed consumption of experimental diabetic rats

Groups	Feed consumption
Normal control group (NC)	669.16±12.23
Diabetic group (DM)	673.90±116.36
DM + pigeon pea leaves extract 300mg/kg BW, and *Z. officinale* extract 60 mg/kg BW (DME)	657.56±8.23
DM + pigeon pea leaves extract 300mg/kg BW and *Z. officinale* extract 125 mg/kg BW (DMF)	654.72±44.20
DM + glibenclamide (DMG)	672.87±9.58

Note: Feed consumption is not significantly different (P≥0.05) among the treated groups

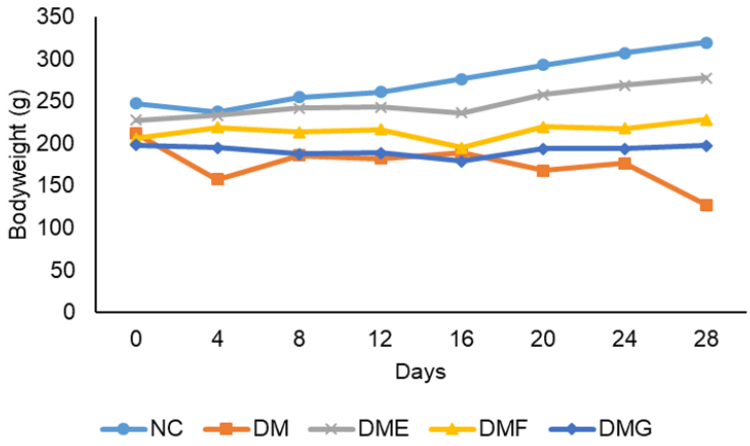

Figure 1. The body weight growth of experimental diabetic rats. The DME group showed highly increased body weight compared to other treatment groups. DM=diabetic group, NC=normal control group, DME= DM+pigeon pea leaves extract (300mg/kg BW) and ginger extract (60 mg/kg BW), DMF=DM+ pigeon pea leaves extract (300mg/kg BW) and ginger extract (125 mg/kg BW), DMG=DM+glibenclamide.

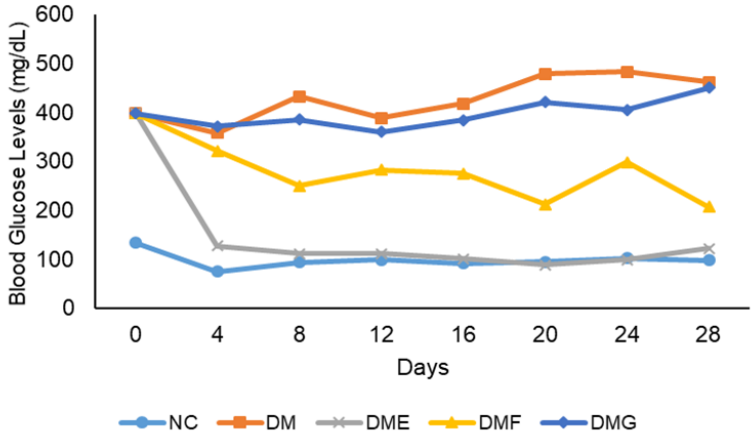

Figure 2. Blood glucose levels of experimental diabetic rats. The DME group showed highly decreased blood glucose levels compared to other treatment groups. DM=diabetic group, NC=normal control group, DME=DM+pigeon pea leaves extract (300mg/kg BW) and ginger extract (60 mg/kg BW), DMF=DM+pigeon pea leaves extract (300mg/kg BW) and ginger extract (125 mg/kg BW), DMG=DM+glibenclamide.

Table 2. The number of acinar cells in the pancreatic tissues of experimental diabetic rats per view of 400 magnification

Group	The number of acinar cells in different levels of Cu,Zn-SOD content in pancreatic tissues			
	+++	++	+	-
CN	165.00±11.00^{c}	62.33±2.51^{ab}	43.00±5.56^{a}	73.33±9.01^{a}
DM	104.00±2.64^{a}	68.00±17.43^{b}	43.33±3.21^{a}	96.33±7.23^{b}
DME	140.00±5.56^{b}	63.00±7.21^{ab}	45.33±12.85^{a}	95.66±5.03^{b}
DMF	114.66±5.03^{a}	46.33±7.09^{a}	41.66±6.80^{a}	68.33±10.40^{a}
DMG	111.33±5.50^{a}	70.00±11.53^{b}	55.33±12.58^{a}	83.00±13.45^{ab}

Note: The different letters of superscript in the same column showed a significantly different (P<0.01). The DME group treated with the combination of pigeon pea leaves extract and ginger extract showed the best result in ameliorating stress oxidative condition in the liver tissues of diabetic rats. DM=diabetic group, NC=normal control group, DME=DM+pigeon pea leaves extract (300mg/kg BW) and ginger extract (60 mg/kg BW), DMF=DM+pigeon pea leaves extract (300mg/kg BW) and ginger extract (125 mg/kg BW), DMG=DM+glibenclamide.

Table 3. The number of cells in the Langerhans islet of pancreatic tissues of experimental diabetic rats per view of 400 magnification

Group	The number of Langerhans islet cells in different levels of Cu,Zn-SOD content in pancreatic tissues			
	+++	++	+	-
CN	97.60±20.39^{c}	44.00±11.53^{c}	30.20±10.70^{b}	46.40±15.07^{b}
DM	15.08±7.59^{a}	14.80±7.91^{ab}	9.80±4.86^{a}	16.40±10.13^{a}
DME	40.20±22.35^{b}	22.40±12.03^{b}	15.60±7.89^{a}	16.80±8.64^{a}
DMF	26.00±16.14^{ab}	12.40±5.12^{ab}	9.60±5.31^{a}	7.60±1.94^{a}
DMG	13.40±8.40^{a}	6.8±4.81^{a}	7.40±3.91^{a}	5.4±2.96^{a}

Note: The different letters of superscript in the same column showed a significantly different (P<0.01). The DME group treated with the combination of pigeon pea leaves extract and ginger extract showed the best result in ameliorating stress oxidative condition in the liver tissues of diabetic rats. DM=diabetic group, NC=normal control group, DME=DM+pigeon pea leaves extract (300mg/kg BW) and ginger extract (60 mg/kg BW), DMF=DM+pigeon pea leaves extract (300mg/kg BW) and ginger extract (125 mg/kg BW), DMG=DM+glibenclamide.

The effectiveness of the combination of *C. cajan* leaves extract and ginger extract in increasing body weight profile was related to carbohydrate metabolism. The results of measuring blood glucose levels for 28 days showed that the group of diabetic rats DME showed the best profile of blood glucose levels compared to other diabetic groups (Figure 2). The blood glucose level of the DME group decreased to the normal range from the 4th day of measurement to the 28th day after being given the combination of *C. cajan* extract and ginger extract. These results show that glucose resulting from carbohydrate metabolism can be absorbed into the cells, used by cells, and stored in the liver and muscles in the rats of the DME group. It can decrease the level of blood glucose (Figure 2) and increase the body weight of the rats in the DME group (Figure 1). The rats of the DME group also do not decompose their glycogen and fat.

The profile of blood glucose levels of the DMF group also showed decreased, but it was still in a hyperglycemic range (Figure 2). The group of diabetic rats given glibenclamide (DMG) showed a profile of blood glucose levels in a hyperglycemia range (Figure 2). The group of diabetic rats that were not given glibenclamide

or combination extracts (DM) showed the highest blood glucose levels or hyperglycemia during 28 days (Figure 2). The hyperglycemia condition in the DM group is followed by the high production of free radicals through several pathways (Yaribeygi *et al.,* 2019), causing oxidative stress.

The condition of oxidative stress in the DM group was shown by the significantly lower antioxidant Cu, Zn-SOD content ($P<0.05$) in the acinar cells (Table 2 and Figure 3) compared to the normal control (CN) group. It is indicated by the significantly lower ($P<0.05$) of the number of positive reaction acinar cells and significantly higher number of negative reaction acinar cells ($P<0.05$) to the antioxidant Cu, Zn-SOD in the DM group (Table 2 and Figure 3). The condition of oxidative stress in the DM group was also indicated by the lower content of the antioxidant Cu, Zn-SOD in the islets of Langerhans cells compared to the normal control (NC) group (Table 3 and Figure 4). It is indicated by the significantly lower number of Langerhans islet cells that strongly, moderate, and weak positive reactions ($P<0.05$) compared to the normal control group (NC) (Table 3 and Figure 4). This is in line with reports that diabetes condition decreased antioxidant Cu, Zn-SOD in liver, kidney, testis and pancreatic tissues of diabetic rats (Wresdiyati *et al.,* 2008b; Wresdiyati *et al.,* 2008a; Wresdiyati *et al.,* 2010; Wresdiyati *et al.,* 2011; Wresdiyati *et al.,* 2015; Wresdiyati *et al.,* 2016; Wresdiyati *et al.,* 2020b).

The DMG diabetic group showed no significantly difference Cu, Zn-SOD antioxidant content in acinar cells ($P>0.05$) to the diabetic rat (DM) group. It was indicated by the similar number of acinar cells that had strongly, moderately, and weak positive reactions and negative reactions in the DMG group ($P>0.05$) compared to the DM group. The antioxidant Cu, Zn-SOD content of the DMG group in the islets of Langerhans cells was also not significantly different ($P>0.05$) from the DM group. The glibenclamide treatment in the diabetic rats showed no increased antioxidant Cu, Zn-SOD content in pancreatic tissues (Tables 2 and 3, Figures 3 and 4).

The DME group that showed the best blood glucose level profile (Figure 2) also showed the highest antioxidant-Cu, Zn-SOD content ($P<0.05$) compared to other diabetic rat groups (Tables 2 and 3, Figures 3 and 4). It is indicated by the significantly higher number ($P<0.05$) of acinar cells and islet cells of Langerhans that reacted positively to antioxidant-Cu, Zn-SOD content compared to other diabetic rats groups. It indicates that the combination of *C. cajan* leaves extract and ginger extract ameliorated oxidative stress by increasing antioxidant defence in the pancreatic of diabetic rats. The mechanism for increasing antioxidant defence may be through several pathways related to the function of the bioactive compounds in *C. cajan* leaves extract and ginger extract in lowering blood glucose levels and free radicals scavenger activity.

It has been reported that the 96% ethanol extract of *C. cajan* leaves extract from Lombok contains flavonoids, saponins, tannins, and steroids (Wresdiyati *et al.,* 2020a). Similarly, the 96% ethanol extract of ginger (*Z. officinale)* var. amarum from Solo, Central Java, contains flavonoids, saponins, and quinones (Wresdiyati *et al.,* 2020a).

Flavonoids suppress blood glucose levels by increasing insulin release from beta cells. Compounds in flavonoids also inhibit the activity of alpha-glucosidase and alpha-amylase better than antidiabetic drugs (Manzo and Vitor, 2017; Nguyen *et al.,* 2023). Yi *et al.* (2023) also reported that flavonoids from natural plants exhibit hypoglycemic properties by regulating blood glucose levels.

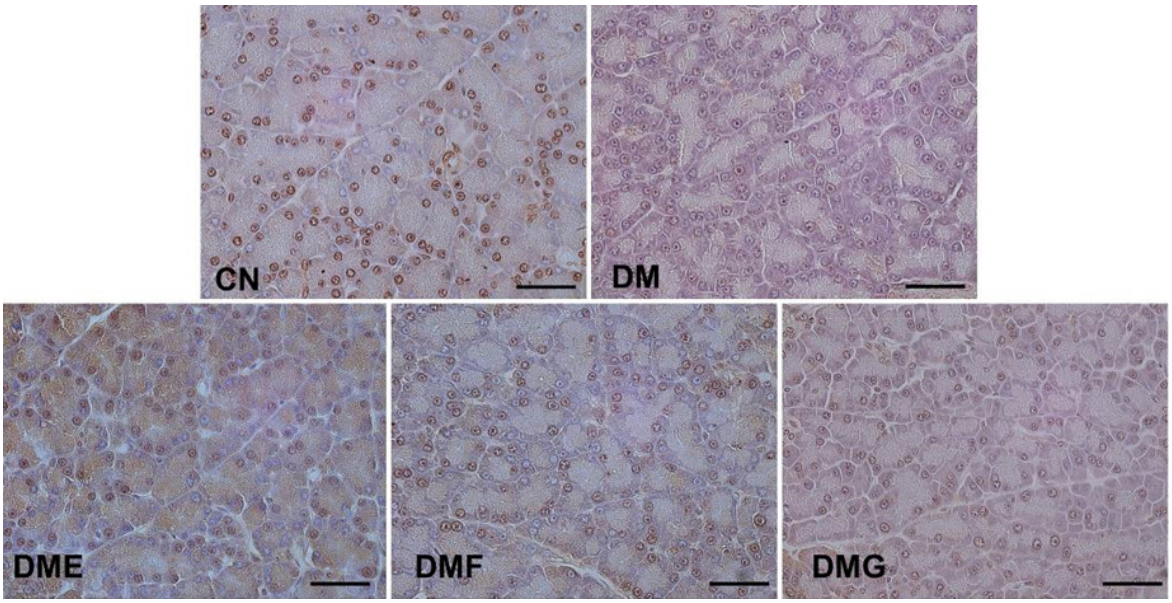

Figure 3. Photomicrograph of Immunohistochemical localization of antioxidant Cu,Zn-SOD in the acinar cells of pancreatic tissues of diabetic rats. The combination of *C. cajan* leaf extract and ginger extract (DME) increases the antioxidant Cu, Zn-SOD content in the Lanherhans islet cells of pancreatic tissues. DM=diabetic group, NC=normal control group, DME=DM+pigeon pea leaves extract (300mg/kg BW) and ginger extract (60 mg/kg BW), DMF=DM+pigeon pea leaves extract (300mg/kg BW) and ginger extract (125 mg/kg BW), DMG=DM+glibenclamide.

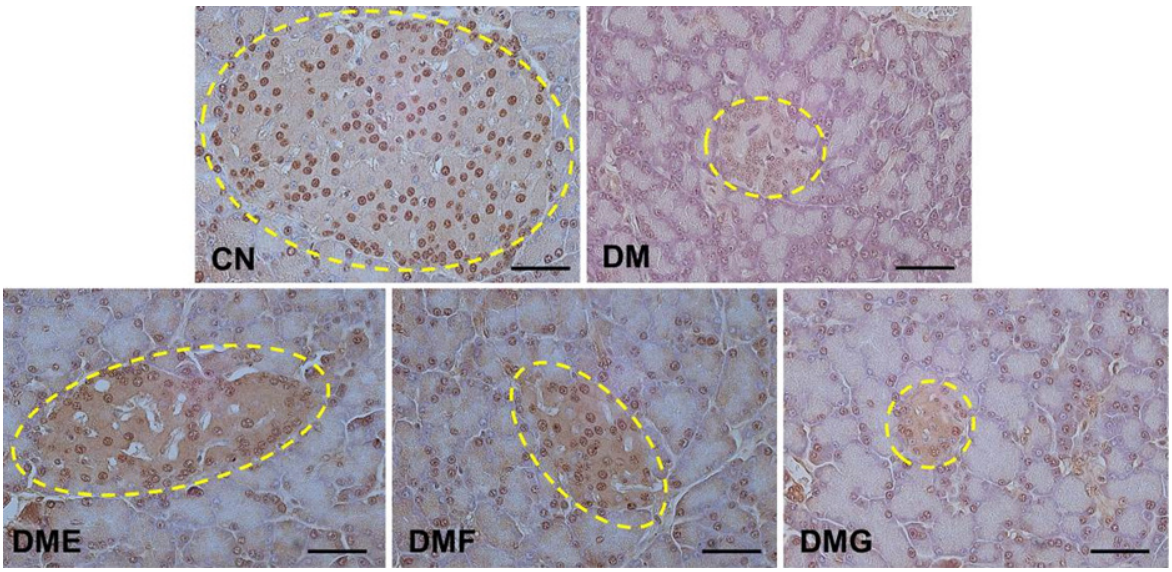

Figure 4. Photomicrograph of Immunohistochemical localization of antioxidant Cu,Zn-SOD in the Lanherhans islet cells of pancreatic tissues of diabetic rats. The combination of *C. cajan* leaf extract and ginger extract (DME) increases the antioxidant Cu, Zn-SOD content in the Lanherhans islet cells of pancreatic tissues. DM=diabetic group, NC=normal control group, DME=DM+pigeon pea leaves extract (300mg/kg BW) and ginger extract (60 mg/kg BW), DMF=DM+pigeon pea leaves extract (300mg/kg BW) and ginger extract (125 mg/kg BW), DMG=DM+glibenclamide.

Saponins lower blood glucose levels by stimulating insulin secretion from pancreatic beta cells and increasing peripheral glucose utilization. Saponins can trigger the regeneration of pancreatic beta cells and inhibition of hepatic gluconeogenesis (Yu *et al.,* 2022). El Barky *et al.* (2017) also reported saponin decreases serum blood glucose levels in diabetic patients. Saponin exhibited a hypoglycemic effect in type 2 diabetic mice (Chai *et al.,* 2021) by increasing glucose uptake activities (He *et al.,* 2023).

Tannins reduce glucose absorption in the digestive system by inhibiting the action of alpha-glucosidase and alpha-amylase enzymes so that the time needed for carbohydrate metabolism is extended and preventing hyperglycemia (Türkan *et al.,* 2019). It was also reported that tannins lowered blood glucose levels and exhibited free radicals scavengers (Sieniawska 2015; Omar *et al.,* 2022), as reported by Esmaiel *et al.,* (2019), tannins can minimize the pathological oxidative state of a diabetic situation, as well as lowered blood glucose levels by enhancing the glucose uptake through activation and GLUT-4 translocation, and insulin-signalling pathways, such as PI3K (Phosphoinositide 3-Kinase) and p38 MAPK (Mitogen-Activated Protein Kinase) (Syafril *et al*., 2019).

The bioactive compounds of ginger extract with a hypoglycemic effect are 6-gingerol, tannins, polyphenolic compounds, flavonoids, and triterpenoids. These compounds exert a hypoglycemic effect by maintaining cell function related to receptors and membrane transport (Momoh *et al.*, 2022). Compound 6-gingerol stimulates glucose uptake and glucose transporter 4 (GLUT4) translocation to cell membranes by regulating GLUT4 gene expression (Lee *et al.,* 2015).

The bioactive compound contained in *C. cajan* extract and ginger extract could suppress blood glucose levels in several pathways mentioned above. Subsequently, the extracts reduced the condition of hyperglycemia. The combination extracts ameliorated hyperglycemia condition then, reduced oxidative stress, and increased the antioxidant Cu and Zn-SOD content in pancreatic tissues.

CONCLUSION

The diabetic rat's group (DM) under alloxan induction decreased the content of Cu, Zn-SOD in the acinar cells and Langerhans islet cells of pancreatic tissues. In the diabetic rat groups, given a combination *of C. cajan* leaves (300 mg/kg BW) and ginger extracts (60 mg/kg BW) decreased the blood glucose level and increased the body weight and Cu, Zn-SOD content in pancreatic tissues. The combination *of C. cajan* leaves and ginger extracts increased antioxidant defence in diabetic rats' pancreatic organs.

SUGGESTION

The combination of *C. cajan* leaves and ginger extracts is suggested to be developed, for further research, as functional food or herb medicine to maintain blood glucose levels and ameliorate oxidative stress under diabetic conditions.

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