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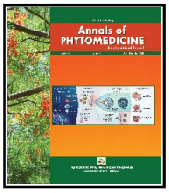

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Antidiabetic effect of palmyrah tuber (*Borassus flabellifer* L.) food products in Streptozotocin and Nicotinamide induced diabetic rats

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Abstract

Diabetes mellitus is a rapidly increasing metabolic disorder and food based therapeutic approaches are gaining importance due to their safety and affordability. The present study evaluated the antidiabetic potential of value-added palmyrah tuber (*Borassus flabellifer* L.) food products in Streptozotocin-Nicotinamide induced type 2 diabetic male Wistar rats. Diabetes was induced using Streptozotocin (65 mg/kg) and Nicotinamide (110 mg/kg) and diabetic rats were orally administered palmyrah tuber flour or palmyrah tuber based puttu mix at a dose of 200 mg/kg body weight for 28 days. Diabetic control rats showed marked hyperglycaemia (297.4 ± 7.8 mg/dl) reduced plasma insulin (5.90 ± 0.45 µl/ml), elevated HbA1c (12.90 ± 1.8 %), dyslipidaemia, impaired liver and kidney function and decreased antioxidant enzyme activities. Treatment with palmyrah tuber flour and puttu mix significantly reduced fasting blood glucose to 144.2 ± 3.6 and 149.7 ± 4.0 mg/dl, respectively, increased plasma insulin (14.5 to 15.5 µl/ml), improved HbA1c, body weight, hemoglobin, lipid profile, hepatic and renal markers, enhanced antioxidant enzymes and reduced lipid peroxidation. Histopathological analysis revealed partial regeneration and protection of pancreatic β-cells in treated rats. The findings substantiate the antidiabetic, hypolipidemic and antioxidant efficacy of palmyrah tuber based food products and support their potential use as functional foods or complementary dietary interventions in the management of type 2 diabetes mellitus.

1. Introduction

Diabetes mellitus (DM) is a metabolic disorder characterized by a loss of glucose homeostasis with disturbances of carbohydrate, fat and protein metabolism resulting from defects in insulin secretion, insulin action or both (Saeedi *et al.,* 2019). Diabetes mellitus is an epidemic health issue that is already spreading at an alarming pace throughout all parts of the world. The cases of type 2 diabetes have increased outburst in the last three decades among countries regardless of their income groupings. In India, 77 million people above the age of 18 yr are suffering from diabetes (type 2) and nearly 25 million are pre-diabetics. Good management of diabetic mellitus is considered to be a top priority to the medical fraternity. However, most pharmacotherapeutics that are already in licensure have a range of side effects. Many studies have been carried out to determine the effectiveness of new pharmacological approaches to stabilize glycaemic parameters. However, the management of diabetes mellitus is yet to be given a clear acceptance on the part of an alternative therapeutic modality.

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The Palmyrah tree has a monopodial growth habit, which appears in the form of a tall, upright tree and is especially well known by the broad, fan shaped leaves. Its fruit pericarps have historically been used as stomachic, sedative, laxative and aphrodisiac and as such they have therapeutic effects in hyperdipsia, dyspepsia, flatulence, dermatological disorders, haemorrhagic episodes, pyrexia and a range of general impediments. Modern day phytochemical studies especially those carried out by Pathberiya and Jansz (2010) have supported the antioxidant capacity within the fruit pulp of the palmyrah. In traditional medicine the extract of immature palm fruits has been widely used for the treatment of diabetes and its secondary complications (Uluwaduge *et al.,* 2005; Uluwaduge *et al.,* 2006). Recently, many studies have reported the *in vitro* antioxidant and antimicrobial properties of immature palm fruits (Renuka *et al.,* 2018; Renuka *et al.,* 2019).

The palmyrah tuber is a rich source of starch and is frequently used to prepare soups, porridge and candies. Palmyra tuber contains nutritional value like energy (384 kcal/100 g), carbohydrates (85.1 g/100 g), fat (1.73 g/100 g), protein (2.92 g/100 g), total sugar (1.7 g/100 g), dietary fiber (2.79 g/100 g), calcium (53 mg/100 g) and iron (1.7 mg/100 g) (Sandhiyadevi *et al.,* 2021). Palmyra tubers are a fiber rich root that can restore irregular bowel movements (Sathya *et al.,* 2022). The tuber forms a critical source of starch food among the rural Jaffnanese of Sri Lanka. The tubers are carefully washed to get rid of the outer sheath and basal roots and then they are boiled for about thirty min the cooked tubers are then eaten. Their organoleptic characteristics cannot be distinguished as compared to other starchy

tubers. Besides, the traditional knowledge assigns multiple therapeutic benefits to the tuber such as the alleviation of biliousness, dysentery and gonorrhoea. The tuber itself or seedling root is a diuretic, antihelminic a decoction of which is said to be used in the treatment of certain respiratory maladies. However, the tuber is often modified into different processed forms that will allow it to have a longer storage life, thus it will be available during off-season**.** An extract from immature endosperm at a concentration of 200 ml was administered against randomly selected 30 numbers of type 2 diabetic patients for twenty eight days and discovered that immature endosperm is a good diet for diabetic patients (Rahman *et al.,* 2020). Palmyrah immature endosperm could be considered as anti-hyperglycemic fruit due to its nutrient contents, especially phytochemicals, fiber, sodium, potassium, copper and zinc. Treatment of diabetes using plants based medicines is an adjunct therapy for controlling glycemic status with lesser side effects. In the present study, it is aimed to evaluate the antidiabetic and antioxidant effect of value added products from palmyrah tuber in Streptozotocin and Nicotinamide induced diabetic rats.

2. Materials and Methods

2.1 Plant material authentication

The plant material was botanically authenticated as *Borassus flabellifer* L. (Family: Arecaceae). The authenticated specimen is preserved in the Herbarium of the Botanical Survey of India (BSI), Ecology Unit, Indian Botanic Garden, Howrah, India. The Voucher Specimen was collected from Kutru, Indravati Tiger Reserve, Bastar, Madhya Pradesh, India and deposited under BSI Accession Number 16369 (CAL0000003199). The specimen was collected by Anand Kumar on 27 May 1987. The herbarium record describes the plant as an unbranched tree reaching up to 15 m in height with fan like leaves and yellow fruits commonly planted near habitations and locally known as 'Tad'. This authenticated herbarium specimen serves as a reference for the identification of the plant material used in the present study.

2.2 Preparation of palmyrah tuber flour and palmyrah puttu mix

Fully matured palmyrah tubers were procured from local farmers in and around Madurai, Tamil Nadu, India and used for the study. The tubers (young shoots) were cleaned by removing the outer layer, washed thoroughly with tap water and boiled for 30 min. After boiling the central stick-like portion was removed and the tubers were cut into small pieces. The sliced tuber pieces were dried in a cabinet dryer at 60 ± 5°C for 7 h. The dried samples were milled, ground and sieved using a 212 µm IS sieve (70 mesh) to obtain palmyrah tuber flour. The flour was packed in airtight containers and stored until further use. Palmyrah puttu mix was prepared by blending raw rice flour (50%) with palmyrah tuber flour (50%). The processed palmyrah products, namely palmyrah tuber flour and palmyrah puttu mix were utilized for the animal study. Palmyrah puttu mix is a ready-to-cook traditional breakfast product formulated from raw rice flour and palmyrah tuber flour (Figure 1).

Palmyrah tuber | Palmyrah tuber flour

Figure 1: Palmyrah tuber and flour.

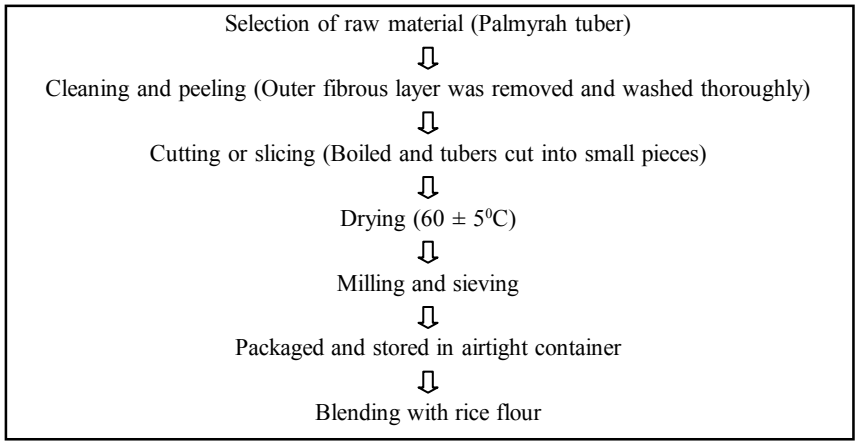

Figure 2: Flow process chart for preparation of Palmyra tuber flour.

2.3 Experimental animals and ethical approval

Adult male Wistar rats weighing 190 to 200 g were procured from the Central Animal House, K.M. College of Pharmacy, Madurai, Tamil Nadu, India. The experimental protocol was reviewed and approved by the Institutional Animal Ethics Committee (IAEC) of K.M. College of Pharmacy, Madurai, Tamil Nadu (Approval No.: IAEC/Dr. K. Shanthi/TNAU/KMCP/04/2023). The animals were housed in well ventilated polypropylene cages under standard laboratory conditions and were provided with a normal pellet diet and water ad libitum throughout the experimental period.

Figure 3: Animal study model showing male Wistar rats used for the experimental evaluation.

2.4 Induction of experimental diabetes

Streptozotocin (STZ) and Nicotinamide (NA) were introduced into overnight-fasted rats (180-200 g each) to induce diabetes mellitus. STZ was intraperitoneally injected at a dose of 65^{-1} body weight at 0.1^{-1} M citrate buffer (pH 4.5) after 15 min of the intraperitoneal injection of NA at 110^{-1} body weights in saline. Diagnosis of hyperglycemia was done by testing the level of blood glucose (BGL) at 48 h after the STZ-NA injection; rats with a BGL level above 220 mg/dl were diagnosed to be diabetic and thus used in the current study.

2.5 Experimental design

The experimental study involved five groups of rats (n=5 per group). Control rats (Group I) received 0.9% NaCl, while diabetic control rats (Group II) were induced with streptozotocin-nicotinamide and treated with 0.9% NaCl. Groups II-V comprised diabetic rats administered glimepiride (10 mg/kg), Palmyrah tuber flour (200 mg/kg) or palmyrah tuber-based puttu mix (200 mg/kg), respectively, all given orally as suspensions in warm 0.9% NaCl for 28 days (Table 1).

2.6 Biochemical estimations

Fasting blood glucose levels were measured at baseline, 14 days and 28 days using a commercially available enzymatic assay kit. Body weight and plasma insulin were recorded at baseline and after 28 days using an enzyme-linked immunosorbent assay (ELISA). After 28 days of treatment rats were fasted for 16 h, sacrificed by cervical decapitation and blood was collected in tubes containing potassium oxalate and sodium fluoride for the estimation of total hemoglobin (Deutschlander *et al.,* 2012), HbA1C (Drabkin and Austin, 1932) and total protein (Nayak and Pattabiraman, 1981). Hepatic function was assessed by measuring plasma alanine transaminase (ALT), aspartate transaminase (AST) and alkaline phosphatase (ALP), while renal function was evaluated through urine urea and creatinine analysis. Lipid profile parameters including total cholesterol, triglycerides and high-density lipoprotein (HDL) were also determined. Oxidative stress was assessed by enzymatic antioxidants superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx) and non-enzymatic antioxidants, reduced glutathione (GSH), alongside lipid peroxidation levels measured as malondialdehyde (MDA), providing an overview of oxidative stress in diabetic rats.

Table 1: Experimental groups and treatments

Group I	Control group, 0.9 per cent sodium chloride (NaCl).
Group II	Streptozotocin with nicotinamide mediated diabetes control treated with 0.9% NaCl.
Group III	Diabetic rats applied to glimepiride (10 mg/kg^{-1} body weight) in 28 days.
Group IV	Diabetic rats fed with palmyrah tuber flour (200 mg/kg^{-1}) in 28 days.
Group V	Palmyrah tuber based puttu mix (200 mg/kg^{-1} 28 days) on diabetic rats.

2.7 Histopathological study

A fragment of pancreatic parenchyma was excised, fixed in 10% neutral buffered formalin, and processed using standard histological procedures. The tissues were embedded in paraffin, sectioned at / µm thickness and stained with hematoxylin and eosin. The sections were examined under a light microscope and photomicrographs were captured for analysis (Lowry *et al.,* 1951).

2.8 Statistical analysis

All quantitative data including body weight, fasting plasma glucose and selected biochemical parameters are presented as mea ± SEM. Data were analyzed using one way analysis of variance (ANOVA), followed by post hoc comparisons with the Newman Keuls multiple range test using Graph Pad In Stat Version 3.0. Statistical significance was considered at $p<0.05$.

3. Results

The antidiabetic effect of palmyrah tuber products in Streptozotocin-Nicotinamide induced diabetic rats was evaluated by assessing changes in body weight, plasma insulin, total hemoglobin, and HbA1C levels in normal and experimental animals. Diabetic control rats exhibited a significant increase in HbA1C levels along with a marked reduction in body weight, plasma insulin and total hemoglobin when compared to the normal control group ($p<0.01$). Administration of palmyrah tuber based products at a dose of 200 mg/kg as well as glibenclamide to diabetic rats significantly ameliorated these alterations, restoring body weight, plasma insulin, total hemoglobin and HbA1C levels toward near-normal values.

Table 2: Effect of value added products from palmyrah tuber on body weight in Streptozotocin and Nicotinamide induced type 2 diabetes in rats

Groups	Treatment	Body weight (g)		
		0 Day	14 Day	28 Day
Group-I	Normal control	213 ± 5.9	223 ± 6.8	248 ± 6.8
Group-II	Diabetic control	217 ± 6.4	196[a] ± 4.5[*]	177[a] ± 4.0[*]
Group-III	STD control	220 ± 6.8	238 ± 7.1	250 ± 6.7
Group-IV	Treatment control - palmyrah flour	222 ± 6.3	234 ± 6.7	245 ± 7.1
Group-V	Treatment control - puttu mix	215 ± 6.0	230 ± 6.8	248 ± 7.2

Note: Values are expressed as Mean ± SED; *[a]-Values are significantly different from normal control

3.1 Effect of palmyrah tuber value-added products on body weight in diabetic rats.

Normal control rats (Group I) exhibited a progressive increase in body weight from 213 ± 5.9 g at day 0 to 248 ± 6.8 g at day 28 (table 2). In contrast, diabetic control rats (Group II) showed a significant reduction in body weight at day 14 (196 ± 4.5 g) and day 28 (177 ± 4.0 g) compared to the normal control (p<0.01). Treatment with the standard drug (Group III) restored body weight to near normal levels (250 ± 6.7 g at day 28). Similarly, diabetic rats treated with palmyrah tuber flour (Group IV) and palmyrah tuber based puttu mix (Group V) showed significant improvement in body weight, reaching 245 ± 7.1 g and 248 ± 7.2 g, respectively, at the end of 28 days indicating the protective effect of palmyrah tuber value added products against diabetes induced weight loss.

Table 3: Effect of palmyrah tuber products on blood glucose and plasma insulin levels in Streptozotocin-Nicotinamide induced diabetic rats

Groups	Treatment	Blood glucose (mg/dl)			Plasma insulin (Micro litter per ml) 28[th] day
		0 Day	14 Day	28 Day	
Group-I	Normal control	93.4 ± 2.8	96.8 ± 3.4	97.5 ± 3.9	19.75 ± 0.97
Group-II	Diabetic control	272.2 ± 7.5	288.5[a] ± 7.3[*]	297.4[a] ± 7.8[*]	5.90[a] ± 0.45[*]
Group-III	STD control	260.4 ± 6.7	153.7[b] ± 4.4[*]	134.6[b] ± 3.7[*]	17.25[b] ± 0.85[*]
Group-IV	Treatment control-palmyrah flour	250.4 ± 5.8	168.7[b] ± 5.3[*]	144.2[b] ± 3.6[*]	15.5[b] ± 0.80[*]
Group-V	Treatment control-puttu mix	258.8 ± 6.4	170.8[b] ± 5.6[*]	149.7[b] ± 4.0[*]	14.5[b] ± 0.76[*]

Note: Values are expressed as Mean ± SED;[*a] Values are significantly different from normal control; [*b] Values are significantly different from diabetic control

3.2 Effect of palmyrah tuber products on blood glucose and plasma insulin levels

As shown in Table 3, diabetic control rats exhibited a marked elevation in fasting blood glucose levels from 272.2 ± 7.5 mg/dl at day 0 to 297.4 ± 7.8 mg/dl at day 28 along with a significant reduction in plasma insulin levels (5.90 ± 0.45 µl/ml) compared to normal control rats (p<0.01). Treatment with the standard drug significantly reduced blood glucose levels to 134.6 ± 3.7 mg/dl and restored plasma insulin to 17.25 ± 0.85 µl/ml by day 28. Similarly, administration of palmyrah tuber flour and palmyrah tuber based puttu mix (200 mg/kg) significantly lowered blood glucose levels to 144.2 ± 3.6 and 149.7 ± 4.0 mg/dl, respectively and improved plasma insulin levels (15.5 ± 0.80 and 14.5 ± 0.76 µl/ml) indicating notable antidiabetic activity comparable to the standard treatment.

3.3 Effect of palmyrah tuber on hematological and biochemical parameters in diabetic rats

Table 3 records the concentrations of plasma proteins, serum urea and creatinine between control and experiment rat cohorts. Statistically significant increase in urea and creatinine and relative decrease in the level of present protein were observed in the diabetic rats and when glibenclamide as well as palmyrah tuber extracts (200 mg/kg) were used the deviations were reduced to near normal levels.

Table 4: Effect of value added products from palmyrah tuber on different biochemical parameters in Streptozotocin and Nicotinamide induced type 2 diabetes in rats

Groups	Hb (mg/dl)	HBA1c (%)	Total proteins (g/dl)	Total cholesterol (mg/dl)	Triglycerides (mg/dl)	HDL (mg/dl)
Group-I	13.9 ± 1.5	6.10 ± 0.72	8.18 ± 0.80	140 ± 5.15	80.4 ± 2.50	45.5 ± 2.35
Group-II	8.4[a] ± 0.7*	12.90[a] ± 1.8*	4.72[a] ± 0.44*	265[a] ± 8.20*	172.4[a] ± 5.25*	24.[a] ± 1.68*
Group-III	13.2[b] ± 1.2*	7.33[b] ± 0.82*	7.47[b] ± 0.70*	174[b] ± 6.55*	88.4[b] ± 2.75*	38.45[b] ± 1.94*
Group-IV	12.9[b] ± 1.3*	7.48[b] ± 0.85*	7.12[b] ± 0.75*	177[b] ± 6.60*	92.8[b] ± 2.85*	36.20[b] ± 1.85*
Group-V	12.6[b] ± 1.2*	7.85[b] ± 0.90*	6.88[b] ± 0.64*	180[b] ± 6.62*	98.4[b] ± 3.55*	33.50[b] ± 1.84[*]

Note: Values are expressed as Mean ± SEM;[*a] Values are significantly different from normal control; [*b] Values are significantly different from diabetic control

Diabetic control rats (Group-II) showed significant reductions in hemoglobin (8.4 ± 0.7 mg/dl), total protein (4.72 ± 0.44 g/dl) and HDL cholesterol (24.5 ± 1.68 mg/dl) along with marked increases in HbA1c (12.90 ± 1.8%), total cholesterol (265 ± 8.20 mg/dl), and triglycerides (172.4 ± 5.25 mg/dl) compared to normal controls (Group-I: Hb 13.9 ± 1.5 mg/dl, total protein 8.18 ± 0.80 g/dl, HDL 45.5 ± 2.35 mg/dl, HbA1c 6.10 ± 0.72%, cholesterol 140 ± 5.15 mg/dl, triglycerides 80.4 ± 2.50 mg/dl). Treatment with glibenclamide (Group-III) and palmyrah tuber extracts at 200 mg/kg (Groups-IV and V) significantly improved these parameters, restoring hemoglobin (12.6 to 13.2 mg/dl), total protein (6.88 to 7.47 g/dl), HDL (33.50 to 38.45 mg/dl), and reducing HbA1c (7.33 to 7.85%), total cholesterol (174 to 180 mg/dl), and triglycerides (88.4 to 98.4 mg/dl) toward normal levels ($p<0.05$), indicating their antihyperglycemic and hypolipidemic potential (Table 4).

3.4 Effect of palmyrah tuber on liver and kidney function markers in diabetic rats

Consecutive oral intake of value added products from palmyrah tuber at a dose of 200 mg/kg for 28 days resulted in a significant ($p<0.05$) reduction in ALT, AST, ALP and triglycerides as well as a significant increase in HDL compared to the diabetic groups (Table 4).

Table 5: Effect of value added products from palmyrah tuber on different biochemical parameters in Streptozotocin and Nicotinamide induced type 2 diabetes in rats

Groups	ALT (U/L)	AST (U/L)	ALP (U/L)	Urea (mg/dl)	Creatinine (mg/dl)
Group-I	28.3 ± 2.12	47.3 ± 2.45	122.3 ± 4.75	30.2 ± 1.80	1.05 ± 0.26
Group-II	57.5^{a} ± 3.40*	95.2^{a} ± 3.20*	240.5^{a} ± 5.45*	83.75^{a} ± 3.30*	3.15^{a} ± 0.73*
Group-III	31.4^{b} ± 2.25*	53.4^{b} ± 2.60*	143.5^{b} ± 5.10*	41.5^{b} ± 1.98*	1.18^{b} ± 0.34*
Group-IV	33.8^{b} ± 2.73*	55.2^{b} ± 2.88*	150.7^{b} ± 5.05*	44.3^{b} ± 2.45*	1.28^{b} ± 0.44*
Group-V	35.3^{b} ± 2.60*	57.8^{b} ± 2.95*	158.6^{b} ± 5.40*	47.5^{b} ± 2.23*	1.37^{b} ± 0.40*

Note: Alanine aminotransferase (ALT); Aspartate Aminotransferase (AST); Alkaline phosphatase (ALP); Unit per litter (U/L); Values are expressed as Mean ± SEM; ^{*a} Values are significantly different from normal control; ^{*b} Values are significantly different from diabetic control.

Diabetic control rats (Group-II) exhibited significant elevations in liver enzymes ALT (57.5 ± 3.40 U/L), AST (95.2 ± 3.20 U/L) and ALP (240.5 ± 5.45 U/L) and renal markers urea (83.75 ± 3.30 mg/dl) and creatinine (3.15 ± 0.73 mg/dl) compared to normal controls (Group-I: ALT 28.3 ± 2.12 U/L, AST 47.3 ± 2.45 U/L, ALP 122.3 ± 4.75 U/L, urea 30.2 ± 1.80 mg/dl, creatinine 1.05 ± 0.26 mg/dl) ($p<0.05$). Oral administration of glibenclamide (Group-III) and palmyrah tuber products at 200 mg/kg (Groups-IV and V) for 28 days significantly restored these parameters toward normal levels, with ALT (31.4 to 35.3 U/L), AST (53.4 to 57.8 U/L), ALP (143.5 to 158.6 U/L), urea (41.5 to 47.5 mg/dl) and creatinine (1.18 to 1.37 mg/dl) ($p<0.05$) indicating hepatoprotective and renoprotective effects of palmyrah tuber in diabetic rats.

3.5 Effect of value-added palmyrah tuber products on hepatic antioxidant status in diabetic rats

The significantly high increase ($p<0.05$) in malondialdehyde (MDA) was accompanied by a significant decrease in the antioxidant parameters in hepatic parenchymal cells as shown in Table 5. On the other hand, Palmyrah products administered orally at a dose of 200 mg/kg^{-1} greatly reduced these perturbations in a dose dependent manner, which ultimately restored the values at the highest concentration used. Palmyrah tuber flour possesses significant high hypoglycaemic index value when compared to puttu mix.

Table 6: Effect of value added products from palmyrah tuber on hepatic oxidant antioxidant parameters in Streptozotocin and Nicotinamide induced type 2 diabetes in rats

Groups	SOD Unit/ (mg protein)	CAT mmol/min/ (mg protein)	GPx mmol/min/ (mg protein)	GSH mmol/100 (mg tissue)	MDA mmol/100 (mg tissue)
Group-I	9.22 ± 0.88	93.2 ± 3.38	11.1 ± 0.95	58.40 ± 3.25	1.21 ± 0.30
Group-II	4.75^{a} ± 0.44*	40.8^{a} ± 2.85*	5.22^{a} ± 0.45*	24.20^{a} ± 1.35*	2.35^{a} ± 0.56*
Group-III	8.65^{b} ± 0.77*	82.7^{b} ± 3.13*	10.50^{b} ± 0.88*	53.45^{b} ± 3.08*	1.45^{b} ± 0.38*
Group-IV	7.87^{b} ± 0.74*	74.4^{b} ± 3.18*	9.40^{b} ± 0.84*	49.80^{b} ± 2.96*	1.50^{b} ± 0.39*
Group-V	7.44^{b} ± 0.69*	69.8^{b} ± 3.03*	8.80^{b} ± 0.75*	45.75^{b} ± 2.83*	1.55^{b} ± 0.42*

Note: Superoxide dismutase (SOD); Catalase (CAT); Glutathione peroxidase (GPx); Glutathione (GSH); Malondialdehyde (MDA); Values are expressed as Mean ± SEM; ^{*a} Values are significantly different from normal control; ^{*b} Values are significantly different from diabetic control

In Streptozotocin and Nicotinamide induced diabetic rats (Group-II) hepatic antioxidant enzymes were significantly decreased compared to normal control (Group-I) with SOD (4.75 ± 0.44 *vs.* 9.22 ± 0.88 Unit/mg protein), CAT (40.8 ± 2.85 *vs.* 93.2 ± 3.38 µmol/min/mg protein), GPx (5.22 ± 0.45 *vs.* 11.1 ± 0.95 µmol/min/mg protein) and GSH (24.20 ± 1.35 *vs.* 58.40 ± 3.25 µmol/100 mg tissue), while lipid peroxidation marker MDA was elevated (2.35 ± 0.56 *vs.* 1.21 ± 0.30 µmol/100 mg tissue, $p<0.05$). Treatment with value-added palmyrah tuber products (Groups-III-V) significantly improved antioxidant status in a dose dependent manner with SOD (8.65 to 7.44), CAT (82.7 to 69.8), GPx (10.5 to 8.8) and GSH (53.45 to 45.75) approaching normal values and MDA reduced to

1.45 to 1.55 µmol/100 mg tissue (p<0.05 *vs.* diabetic control). These findings indicate the hepatoprotective and antioxidative potential of palmyrah tuber in type 2 diabetes (Table 6).

3.6 Effect of palmyrah tuber products on pancreatic histology

The histological analysis of the pancreatic tissue in healthy rats showed the normal structure of the islets of Langerhans that were homogenously distributed throughout the pancreatic parenchyma with varing sizes in the same lobule. Each of the islets was organized in a system of anastomosing cellular plates and a reticular membrane separating it and the neighboring acini. On the other hand, peripheral widening of the pancreatic islets between the islet of Langerhans and the acinar units around it was observed in the pancreatic of the diabetic control group. Glibenclamide group, Langerhans cells were packed tightly and there was little intercellular space and no inflammatory infiltration. Some form of architectural chaos was evident in the pancreas in comparison with the animals with diabetic control that had been injected with insulin. Nevertheless, diabetic rats fed on palmyrh tuber products at the dose of 200 mg/kg^{-1} exhibited a relatively less pronounced architectural changes and peripheral expansion between the acinar and Langerhans cells compared to diabetic rats. In general, the highest recovery was obtained using the value-added palmyrah tuber product at a dose of 200 mg/kg^{-1} min the hyperplasia in all of the histopathological alterations with a partial proliferation of beta cells (Figure 4).

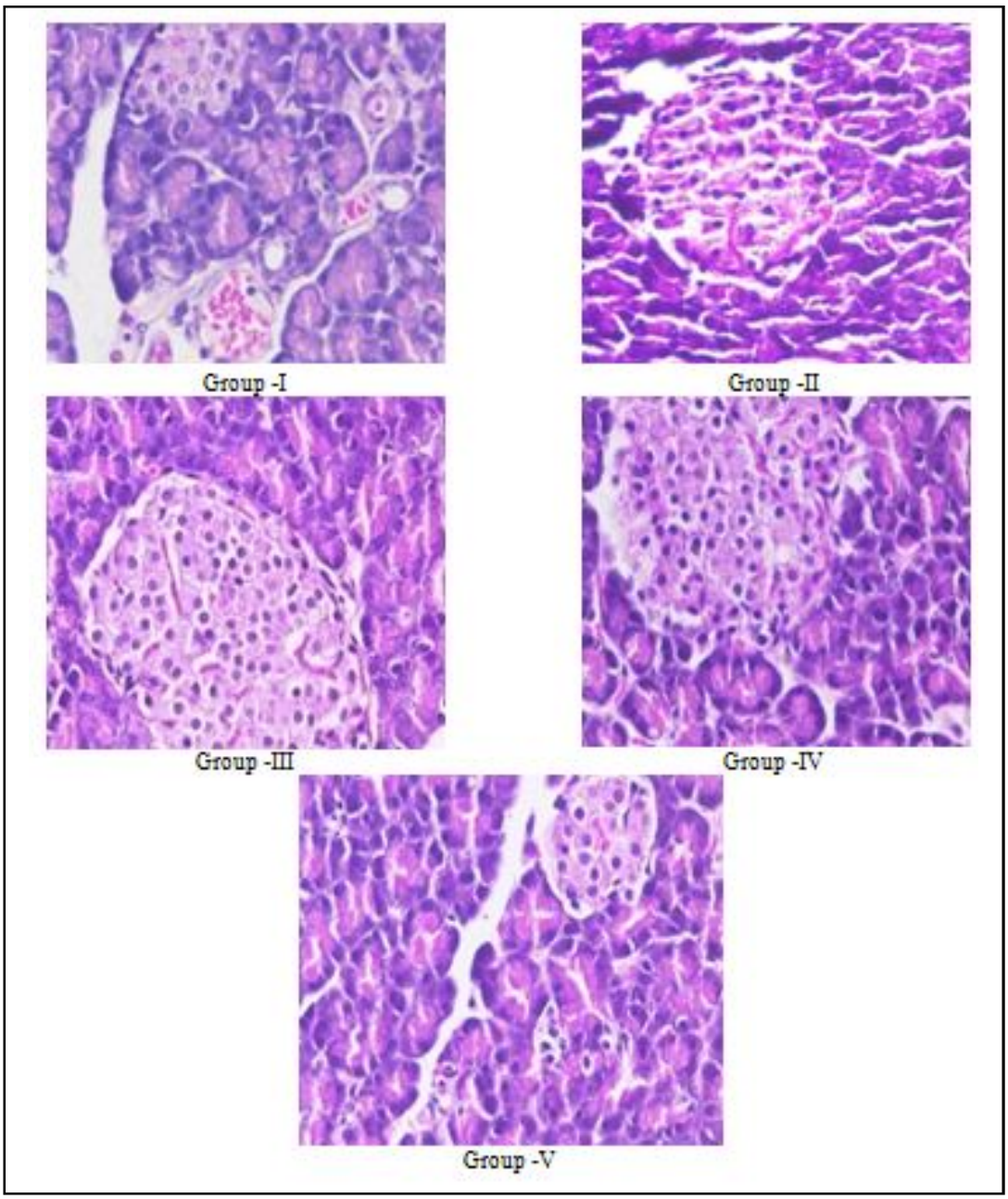

Figure 4: Histopathological studies of palmyrah tuber products treated type 2 diabetes rats.

4. Discussion

The use of herbal medicine by human beings dates back to millenniums. Indigenous Indian systems like Ayurveda and Unani are classical systems that carefully list numerous crude pharma copoeia that had been developed to treat a variety of pathological conditions. These treatment preparations represent an immense botanical extracts tradition. There are many plant species exploited in vernacular medicine in various cultures and these could be used as a therapeutic agent in diabetes mellitus across the world. However, there is a broad array of oral and systemic antidiabetic drugs that are presently available in the pharmaceutical market. Need for the natural antidiabetic products are increasing as a complementary remedy (Pan *et al.,* 2014).

Studies indicated that the rats that are treated with Nicotinamide or Streptozotocin produced type 2 diabetes mellitus, but rats treated with streptozotocin alone induced type 1 diabetes mellitus (Ahangarpour *et al.,* 2016). Streptozotocin selectively damages the insulin-secreting β-cells of the pancreas and thereby produces a diabetic condition. Insufficient insulin level further results in the disability of cells to use glucose and subsequently results in the production of reactive oxygen species (Rani *et al.,* 2019).

However, nicotinamide partially protects pancreatic β-cells against streptozotocin by inhibition of (adenosine diphosphate ribose-ribose) polymerase activity and serves as a precursor of nicotinamide adenine dinucleotide (Wu and Yan, 2015). Furthermore, these experimental rats demonstrate various diabetic complications such as cardiomyopathy, retinopathy, nephropathy and neuropathy which particularly develop through oxidative stress induced mechanisms (Giacco and Brownlee, 2010).

The current study demonstrated a significant drop in the levels of insulin in the fasting plasma of the diabetic rats. These findings agree with the known phenotypic profile of Nicotinamide or Streptozotocin induced type 2 diabetes mellitus in rodent models (Gharib and Montasser Kouhsari, 2019). Further, our research captures an intense increase in the insulin level accompanied by a considerable decrease in the level of fasting blood glucose in rats subjected to a polyherbal preparation.

Hypoglycaemic effect of palmyrah tuber extracts at 200 mg/kg^{-1} could be attributed plausibly to their ability to reinstate the integrity of the cellular plasma membrane as well as to regulate related functions, which are essentially regulated by insulin signaling. Moreover, the antioxidant property of palmyrah tuber food products at a dose of 200 mg/kg^{-1} might have a role in increasing insulin levels by protecting the β-cells of the pancreas against oxidative stress induced cellular injury (Pareek *et al.,* 2009).

In this research, it was established that palmyrah tuber products used at a dose of 200 mg/kg resulted in a significant reduction in the levels of blood glucose in diabetic rats after 28 days of treatment. The antidiabetic effect palmyrah tuber and puttu mix base at 200 mg/kg could be on the basis of higher secretion of insulin by the available beta-cells of the pancreas. The findings of the study are in agreement with studies reported by authors (Pareek *et al.,* 2009; Yang *et al.,* 2016). Similar study was reported by Peter *et al.* (2023) in ethanolic extracts of palmrah palm sprouts and it was considerably improved the glycemic state of rats with STZ induced diabetes. The study backs up palm sprout extracts' medicinal promise in the treatment of diabetes.

In addition, the antihyperglycemic effect that was realized when using palmyrah flour and puttu mixture at a dosage of 200 mg/kg^{-1} of body weight went hand in hand with a quantifiable increase in plasma insulin levels. This observation implies that the value-added products of the palmyrah tuber have insulin secretagogue functions. The observed increase in plasma insulin level suggests that the palmyrah tuber preparations (*Borassus flabellifer*) at the indicated dose induce the release of insulin not only of the remaining pancreatic β-cells but also of newly regenerated ones.

Body weight reduction due to the imbalance of metabolic pathways is commonly associated with diabetes mellitus. In the current study, diabetic rats treated with value added products from palmyrah tuber at a dose of 200 mg/kg^{-1} significantly gained weight, most likely due to reversing the glycogenolysis and gluconeogenesis and thereby helping the restoration of normal metabolic pathways (Rines *et al.,* 2016). Increased non enzymatic glycosylation is one of the possible mechanisms linking hyperglycaemia and vascular complications of diabetes. During diabetes, the excess glucose present in the blood reacts with haemoglobin to form HbA1C (Kondeti *et al.,* 2010).

The diabetic rats in the current study showed substantially high levels of HbA1c compared to their non-diabetic counterparts which highlights ineffective glycaemic control. Value-added palmyrah tuber products at a dose of 200 mg/kg was significantly associated with the reduction of HbA1c levels and the simultaneous increase of total haemoglobin, which is a predictor of increased glucose homeostasis. High hepatic enzyme activity is usually associated with hyperglycaemic in type 2 diabetes patients (Teshome *et al.,* 2019). In line with this, the present study indicated that serum ALT, AST and ALP have gone up significantly in the diabetic cohort. According to Qian *et al.* (2015), the increase in transaminases can enhance the development of diabetic ketogenesis and gluconeogenesis.

Alternatively, when diabetic rats were administered with palmyrah tuber extract at 200 mg/kg, there was a significant decrease in the hepatic levels of transaminase. These results confirm the hepatoprotective effect of the purified product in the diabetic environment. Moreover, hyperglycaemic nephropathy presented itself in the form of significantly increased serum urea and creatinine (Yu and Bonventre, 2018). The current experiment found that diabetic animals were characterized with severe renal dysfunction, in terms of elevated urea and creatinine levels, thus, demonstrating a lack of excretory capacity. Interestingly, rats that were treated with the 200 mg kg^{-1} palmyrah tuber preparation showed significant reduction in the serum creatinine and urea levels, a phenomenon that indicates a renoprotective effect.

In diabetes mellitus, autoxidation of glucose results in the production of ROS that further enhance lipid peroxidation and produce lipoxidation end products and more free radicals (Bajaj and Khan, 2012). Lipid peroxidation is accountable for protein aggregation, which causes liver damage and vascular complications of diabetes mellitus (Moldogaziev *et al.,* 2019). Hyperglycemia is coupled with a rise in plasma malondialdehyde a lipid peroxidation product and a marker of free radical production. In experimental models of diabetes mellitus, high levels of lipid peroxidation in both hepatic and renal parenchymal cells were noticed which are in line with the findings of the present study (Ananthan *et al.,* 2003). In the current study, it is determined that the use of value-added products of palmyrah tuber 200 mg per kilogram of body weight is effective in reducing lipid

peroxidation to a significant degree and that the likelihood of tissue damage is reduced significantly. Analysis of photomicrograph of Streptozotocin-Nicotinamide damaged pancreatic tissue illustrates the damage that Streptozotocin-Nicotinamide causes to the exocrine and endocrine compartments. Furthermore, glibenclamide encourages recovery of the pancreatic islets, a fact that seems to explain the recorded increase in plasma insulin levels recorded in the biochemical tests and supported by histological studies (Oche *et al.,* 2014).

The findings of the current study showed that the value-added products of palmyrah tuber, when used at a concentration of 200 mg/kg^{-1} protect against injury caused by reactive oxygen species in the Langerhans islets of the pancreas. These preparations are capable of antidiabetic and antioxidant activity and can be presumably attributed to a synergistic effect of their phytochemical components.

5. Conclusion

The present study confirms the antidiabetic efficacy of palmyrah tuber value-added food products in Streptozotocin-Nicotinamide induced type 2 diabetic male Wistar rats. Diabetic control rats exhibited severe hyperglycaemia (297.4 ± 7.8 mg/dl), reduced plasma insulin (5.90 ± 0.45 µl/ml), elevated HbA1c (12.90 ± 1.8%) and significant weight loss (177 ± 4.0 g). Oral administration of palmyrah tuber flour and palmyrah tuber based puttu mix at 200 mg/kg body weight for 28 days significantly reduced fasting blood glucose to 144.2 ± 3.6 mg/dl and 149.7 ± 4.0 mg/dl, respectively, while increasing plasma insulin levels to 15.5 ± 0.80 and 14.5 ± 0.76 µl/ml, comparable to the standard drug group. Treatment also markedly improved HbA1c (7.48 to 7.85%), total hemoglobin (12.6 to 12.9 mg/dl), lipid profile by reducing total cholesterol (177 to 180 mg/dl) and triglycerides (92.8 to 98.4 mg/dl) and increasing HDL levels (33.5 to 36.2 mg/dl). Hepatic enzymes (ALT, AST, ALP) and renal markers (urea and creatinine) were significantly restored toward normal levels indicating hepatoprotective and renoprotective effects. Furthermore, palmyrah tuber products significantly enhanced antioxidant enzymes (SOD, CAT, GPx, GSH) and reduced lipid peroxidation (MDA: 1.50 to 1.55 µmol/100 mg tissue). Histopathological findings supported these biochemical improvements by demonstrating partial regeneration and protection of pancreatic β-cells. Overall, the results demonstrate that palmyrah tuber based food products possess significant hypoglycemic, hypolipidemic and antioxidant properties and may serve as a promising functional food or complementary dietary strategy for the management of type 2 diabetes mellitus.

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Conflict of interest

The authors declare no conflicts of interest relevant to this article.

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