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Pancreatic Enzyme Replacement Therapy in hyperglycemic Male Rats

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Abstract

The study was designed to evaluated exocrine pancreatic enzyme replacement therapy in hyperglycemic male rats induced by alloxan. A total of forty five adult male rats were use in this study, hyperglycemia was induce in thirty rats by single intraperitoneal (i.p) injection of Alloxan 100mg/kg/B.W. and the experimental (45) distributed into three groups: the first group (15 animals) administered distilled water orally service as control group. The second group hyperglycemic groups (15 animals) was treated with exocrine pancreatic enzyme(EPE).The third group hyperglycemic group non treated with exocrine pancreatic enzyme(NEPE). Blood samples were take in days 20,40,60 of experiment for measurement the following parameters: Serum some biochemical parameters, tissue pancreatic activity.

The results showed that tissue orally gavages exocrine pancreatic enzyme(EPE) caused a significant elevation in tissue amylase activity in all treated group as compared to the control group. and significant increase in tissue amylase and decrease lipase and protease in concentration in EPE treated group as compared to the non-treated (NEPE) group. Also serum insulin concentration significant decrease and glucose showed a significant increase in EPE and NEPE groups when compared with the control group in after 20,40 and 60 day of experiment. The serum albumin showed significant decrease in all EPE groups when contrasted with the control group. And also The results illustrated that significant decrease in albumin concentration in all NEPE as compared to the control group. The Serum protein concentration was significant decrease in EPE and NEPE groups as compared to the control group in all experimental day. The results showed significant increase in serum cholesterol and triglyceride in EPE as compared to the NEPE . The results showed that PEP rats caused a significant decrease in serum high density lipoprotein (HDL)concentration as compared to the control group and significant decrease in HDL concentration in NEPE as compared to the control group. results showed that doses rats caused a significant($P<0.05$) increase in serum VLDL and LDL concentration in EPE as compared to the control group and significant decrease in NEPE as compared to the control group. On conclusions, the present study confirmed that the exocrine pancreatic function are very frequently and severely altered in diabetes mellitus rats also can enhancements of some metabolic of diabetes mellitus rats when it orally gavages with replacement therapy of pancreatic exocrine enzyme .

Key word: Pancrease , Enzyme, Hyperglycemic, Rats.

العلاج باستبدال إنزيم البنكرياس في ذكور الجرذان المصابة بفرط سكر الدم

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الخلاصة :

صممت الدراسة لتقييم العلاج ببدائل انزيمات البنكرياس الخارجية في الجرذان المصابة بداء السكري المستحدث بالالوكسان. حيث تم استخدام خمسة وأربعون جرذًا ذكراً بالغًا في هذه الدراسة ، تراوحت أوزان جسدهم بين (200-300) غرام

تم احداث ارتفاع السكر في الدم في ثلاثين جرذ بعد صيام الحيوانات لمدة 24 ساعة ثم حقنها داخل التجويف البريتوني بتركيز 100 ملغ/ كغم من وزن الحيوان بعد ثلاث أيام من الحقن تم قياس نسبة الكلوكوز في الدم بعد تصويمها طول الليل عيث اعتبرت التراكيز الأعلى 150 ملغ / ديسي لتر بانها جرذان مصابة بالسكري. قسمت الحيوانات التجريبية (45) إلى ثلاث مجموعات: المجموعة الأولى : (15 حيوانًا) كانت تجرع بالماء المقطر فمويا كمجموعة سيطرة. المجموعة الثانية : (15 حيوانًا) حيوانات مصابة بالسكري وتجرع فمويا بإنزيم البنكرياس الإفرازي .المجموعة الثالثة : (15 حيوانا) حيوانات مصابة بالسكري ولا تجرع فمويا بإنزيم البنكرياس الإفرازي .

تم أخذ عينات من الدم في أيام 60،40،20 من التجربة من خلال ثقب القلب من كل جرذ في كل مجموعة من أيام نهاية المجموعة ، ثم تم جمع المصل لقياس المعلمات التالية: الجلوكوز في الدم ، الأنسولين ، البروتين الكلي ، الزلال ، الكولسترول الكلي ، الدهون الثلاثية ، البروتينات الدهنية واطئة الكثافة ، البروتينات الدهنية عالية الكثافة وتركيز. تم أخذ نسيج البنكرياس لقياس نشاط البنكرياس الأميليز والتريبسين واللايبيز.

أظهرت النتائج أن الأنزيمات التي تعطى عن طريق الفم كبدائل لإنزيم البنكرياس الإفرازي تسبب في زيادة كبيرة في تركيز الأميليز في الأنسجة في جميع المجموعات المعالجة مقارنةً بمجموعة التحكم. وزيادة كبيرة في تركيز الأميليز الأنسجة في المجموعة المعالجة بالمقارنة مع مجموعة غير المعالجة. وأظهرت النتائج أن العلاج تسبب في انخفاض كبير في تركيز اللايبيز في الأنسجة في مجموعة المجرعة بالمقارنة مع مجموعة المراقبة. وكذلك أظهرت النتائج وجود زيادة ملحوظة في تركيز اللايبيز في كل مجموعة الغير مجرعة مقارنة مع مجموعة التحكم في جميع الأيام التجريبية. وأوضحت النتائج انخفاضًا كبيرًا في تركيز اللايبيز في كل ايام التجربة في مجموعة المجرعة مقارنةً بمجموعة غير المجرعة.

أظهرت النتائج أن الانزيمات المجرعة تسبب في زيادة كبيرة في تركيز البروتياز في الأنسجة في كل مجموعة المجرعة مقارنة مع مجموعة التحكم في كل يوم تجريبي. وانخفاض كبير في تركيز البروتييز في كل مجموعة الغير مجرعة مقارنة مع المجموعة السيطرة.

وأظهرت النتائج أن تركيز الأنسولين في مصل الدم في مجموعات المجرعة و الغير مجرعة ينقص بشكل ملحوظ بالمقارنة مع مجموعة السيطرة بعد 40،20 و 60 يومًا من التجربة. وأظهرت النتائج انخفاضًا كبيرًا في تركيز الأنسولين في المصل في كل يوم تجريبي في المجموعة المجرعة مقارنةً بمجموعة غير المجرعة. بينما أظهر تركيز الجلوكوز في الدم زيادة كبيرة في تركيز الجلوكوز في الدم في جميع المجموعات المعالجة مقارنةً بمجموعة السيطرة. وكذلك أظهرت النتائج وجود زيادة ملحوظة ($P < 0.05$) في تركيز الجلوكوز في مصل الدم في جميع المجموعات غير المجرعة مقارنة بـ المجموعات المجرعة ومجموعة السيطرة.

وأظهرت نتائج المصل انخفاض كبير في جميع المجموعات المجرعة بالمقارنة مع المجموعة السيطرة. وكذلك أظهرت النتائج وجود انخفاض كبير في تركيز المصل في الغير مجرعة مقارنةً بمجموعة السيطرة. كان تركيز البروتين في المصل انخفاضًا كبيرًا في مجموعات المجرعة و غير المجرعة مقارنةً بمجموعة السيطرة في كل يوم تجريبي.

أظهرت النتائج انخفاضًا كبيرًا في تركيز الكوليسترول في الدم في مجموعات المجرعة مقارنةً بمجموعة السيطرة . وزيادة كبيرة في الكوليسترول في الدم في المجرعة بالمقارنة مع غير المجرعة. وأظهرت النتائج أن مجموعة المجرعة زيادة كبيرة في تركيز الدهون الثلاثية في الدم بالمقارنة مع مجموعة السيطرة. وأظهرت النتائج وجود انخفاض كبير ($P < 0.05$) في تركيز الدهون الثلاثية في مصل الدم في غير المجرعة مقارنةً بمجموعة السيطرة. أظهرت النتائج أن الجرذان المجرعة تسببت في انخفاض كبير في تركيز البروتين الدهني عالي الكثافة في المصل بالمقارنة مع مجموعة السيطرة وانخفاض كبير في تركيز البروتين الدهني عالي الكثافة في غير المجرعة مقارنة مع مجموعة السيطرة. أظهرت النتائج أن جرعات الجرذان تسببت في زيادة معنوية ($P < 0.05$) في تركيز مصل الدم البروتين الدهني ,واطئ الكثافة في المجرعة مقارنةً بمجموعة السيطرة وانخفاض كبير في غير المجرعة مقارنةً بمجموعة السيطرة.

نستنتج من الدراسة ان انزيمات البنكرياس الهضمية تتغير تباعا للحالة المرضية للاصابة بالسكري وبذلك تتغير بعض تراكيز بعض المواد الايضية بالجسم وبالامكان تقليل هذا التغير باعطاء بدائل انزيمات البنكرياس عن طريق الفم .

الكلمات الافتتاحية : انزيمات، البنكرياس .داء السكري ، الجرذان

1. Introduction

The pancreas of rat located just slantwise in the retroperitoneum of the superior abdomen as it's enveloped with stomach, transverse colon, as so as transverse mesocolon consecutively (1). The rat pancreas includes two parts in single texture: the exocrine gland which inclusing of pancreatic acinar cells beside the duct cells that secrete digestive enzymes with sodium bicarbonate, the endocrine gland involves four cells, termed (α , β , δ , PP and ε) cells which secrete glucagon, insulin, somatostatin, pancreatic polypeptide, with ghrelin in sequent (2). Whereas the physiological function of exocrine pancreas which is synthesis digestive enzymes that in charge of chemical digestion of heaped up chyme, the endocrine pancreas function is to produce peptide hormones that play a huge role in symmetric conservation of glucose homeostatic system. The pancreatic

total functions are finely controlled and regulated by neurocrine, endocrine, paracrine and/or intracrine varietal mechanisms. therefore, dysregulation of these pathways must have significant influences on ours health (3). The phrase diabetes originated from Latin and archaic Greek and verbally translation "siphon". The phrase "diabetes". The word mellitus derived from Latin and translated to "honey sweet". It was Thomas Wills in the year 1675 who presented the suffix word "mellitus" to diabetes in reference to the excreted urine of diabetic patients who had a relative sweet taste (4) . An inquisition of serum immunoreactive trypsin level with pancreatic isoamylase efficiency in diabetes mellitus patients of has clarified that exocrine pancreatic deficit in utmost maximal in insulin dependent diabetics, middle average in those regulated with sulphonylureas, and lost in patients under biguanides influence or diet or both (5). A significant links among the serum concentration amount of both of these enzymes of pancrease and C peptide that were established. the blood concentrations of pancreatic enzyme revealed to relation to glycosylated haemoglobin level, the given the trearment of insulin, or the actual age of starting of diabetes. The influx levels of immunoreactive trypsin was shown to be decrease in generality of cases of insulin dependent diabetics in this enzyme in which measurement was taken at the time of the clinical start of diabetes (6). Therefore exocrine pancreatic defects in diabetes .correlatley counterpart and resembles the endocrine ,B cell disorder and happens concordantly with the fundamental clinical aspect of type I diabetes. It is thusly potential that in type I diabetes equivalent mechanisms are required in the complete pathogenesis of both endocrine and exocrine pancreatic action (7).

The study will be modeling and design to study the effect of exocrine pancreatic enzymes therapy in diabetic rats animal induced by alloxan.

2. Materials And Methods

2.1.Induce Hyperglycemia

Hyperglycemia was caused by a single intraperitoneal (i.p) injection of Alloxan 100mg / kg (B.W) in thirty rats fasting for 24 hours. Alloxan potent of produced lethal hypoglycemia as a result of huge pancreatic liberation of insulin to avoid hypoglycemia, the rats must be treated with 5% of glucose solution for the subsequent 24hs, aftert hree days of alloxan trated, the fasting blood glucose levels were estimation and when greater than 150 mg / dL were deemed hyperglycemia / diabetes. (8).

2.2. Experimental design

Forty five adult male rats were divided randomly and equally into three groups: the first group (15 animals) administered distilled water orally service as control group. The second group diabetic groups(15 animals) was treated with exocrine pancreatic enzyme (EPE). The third group diabetic group non treated with exocrine pancreatic enzyme (NEPE). Blood samples were take in days 20,40,60 of experiment through cardiac puncture from each rat in each groups end days and then serum was collected for measurement the following parameters: Serum glucose, insulin, total protein, albumin, total cholesterol, triglyceride, HDL , LDL and VLDL concentration. using semi-automatic chemistry analyzer Belgium using kit Cyan com./Belgium),.While, insulin concentration measured use Immunoenzymometric assay. The rats pancreatic tissue were be taken for measured tissue pancreatic lipase , amylase and trypsin concentration. According to rats enzymatic ELISA kit (Rat PRSS1 /Protease, Serine, 1/ Catalog No : E-EL-R0799) ; Rat PL/ Pancreatic Lipase/ Catalog No : E-EL-R2441) and Rat AMY2/ Amylase Alpha 2, Pancreatic/ Catalog No : E-EL-R2545) respectively.

2.3. Tissue homogenates:

According to company kit procedure the tissue should be minced in to small pieces and rinsed in ice cold PBS (0.01M,PH=7.4) to remove excess blood thoroughly. Tissue pieces should be weighed and then homogenized in PBS (tissue weight (g): PBS (ML) volume=1:9) with a glass homogenizer on ice. To further break down the cell, you can sonicate the suspension with an ultrasonic cell disrupter or subject it to freeze-thaw cycles. The homogenates are then centrifuged

for 5 min at 5000×g so get the supernatant. Cell culture supernatant or other biological fluids: Centrifuge samples for 20 minutes at 1000×g at 2-8C°.Collected the supernatant to carry out the assay.

2.4. Statistic Analysis

Statistical analysis of the experimental results was performed in accordance with SPSS version 13.00 where one and two methods (ANOVA) were used to determine the importance of differences between groups and periods. Results were represented as mean ± Standard Errors (SE) and P value<0.05 was deemed statistically significant LSD to be used to check the significant level between treatment methods. (SPSS., 2002).

2.5. Ethical consideration

The consent was taken from the Central Committee for Bioethics University of Kufa, with approval number 15229 data June/23/2019. Informed and written consents were obtained from all participants.

3. Results

3.1. Rat tissue pancreatic amylase detection.

The concentration of tissue amylase (TA) (ng / dL) in control and amylase EPE and NEPE is present in table(1). The results showed that oral gavages of exocrine pancreatic enzyme (EPE) caused a significant (P<0.05) elevation in the concentration of tissue amylase in all EPE groups (G2) compared to NEPE(G1) and control(C) community. And also The findings showed a significant (P<0.05) decrease in the concentration of tissue amylase in NEPE groups (G1) compared to the control group .

3.2. Rat tissue pancreatic lipase detection.

The concentration of tissue lipase (TL) (pg / mL) in control and EPE and NEPE treated male rats shown in table (2). The results showed that EPE therapy resulted in a significant (P<0.05) increase in the concentration of tissue lipase(TL) in the EPE group (G2) compared to the NEPE (G1). Furthermore, the findings showed a significant (P<0.05) decrease in the concentration of tissue lipase(TL) in all NEPE groups (G1) compared to the control group(C) in all experimental days.

3.3. Rat tissue pancreatic protease detection.

The concentration of tissue protease (TP) in control and EPE and NEPE in induced diabetic male rats are shown in table (3). The results showed that EPE caused a significant increase in the concentration of tissue protease (TP) in all groups of EPE (G2) compared to the control group (C) and NEPE (G1) during all experimental days. And also The results showed a significant (P<0.05) decrease in the concentration of tissue protease in all NEPE groups (G1) compared to the control group (C).

3.4. Rat serum pancreatic insulin detection.

Table (4) shows the concentration of Serum insulin (SI) (ng / mL) in control , EPE and NEPE in induced diabetic male rats. The results showed that the serum insulin concentration in the EPE and NEPE groups decreased significantly (P<0.05) compared to the control group after 20,40 and 60 days of the experiment.

3.5. Determination of the serum glucose concentration (mg/dl).

Table (5) demonstrated the concentration of serum glucose (SG) (mg / dL) in experimental male rats. The findings showed a significant (P<0.05) rise in serum glucose concentrations in EPE (G2) compared to the control group (C). And also The findings showed a significant (P<0.05) rise in serum glucose concentrations in NEPE (G1) relative to the EPE groups (G2) and control groups (C).

4.6. Determination of serum total albumin concentration(mg/dL).

Table (6) explained the concentration of serum albumin (SA) (ng / mL) in male control rats, EPE and NEPE. The findings showed a significant (P<0.05) rise in serum albumin concentration in all EPE groups (G2) compared to NEPE and control group (C). And also The findings showed a non significant (P<0.05) in serum albumin concentration in all EPE groups (G2) compared to the control group (C).

3.7. determination of serum total protein concentration(mg/dL).

The concentrations of serum protein (SP)mg / dL in EPE and NEPE treated are shown in table (7). The results showed that SP decreased significantly (P<0.05) in the EPE and NEPE groups compared to the control group in all experimental days. Meanwhile, there was no significant increase between EPE and NEPE in 20 and 40 days, but there was a significant increase in SP in the EPE group after 60 days of the experimental day compared to each other.

3.8. Determination of serum total lipid profile concentration

Table(8) clarified the total serum cholesterol concentration (mg / dL) in EPE and NEPE of experimental animals. The results showed a significant (P<0.05) decrease in serum cholesterol concentration in all NEPE groups compared to the EPE and control group (C). And also showed non significant (P<0.05) in serum cholesterol concentration between EPE and control group in all experimental days.

The concentration of serum triglyceride(ST)(mg / dL)in control , EPE and NEPE in diabetic male rats was shown in table(9). The results showed that EPE caused a significant increase in serum triglyceride concentration (P<0.05) compared to NEPE in day 20,40,60 of experiment. The table also showed a non significant (P<0.05) in serum triglyceride concentrations in EPE groups compared to control group in all experimental days.

Serum high density lipoprotein (HDL-C) (SH) concentration (mg/mL) in experimental male rats is present in table(10).The results showed that orally gavage of EPE to the rats resulted in a significant (P<0.05) increase in serum (HDL-C) concentration in all EPE groups (G2) compared to NEPE(G1) after20m40,60 of treatment. Also the results showen that non significant (P 0.05) between EPE and control group after 60 days pf experiment when compared with each other.

Serum very low density lipoprotein (VLDL-C),concentration(ng / mL)in control and EPE and NEPE in diabetic male rats is present in table(11).The results showed that doses of rats caused a significant (P<0.05) decrease in serum (VLDL-C) concentration in NEPE groups compared to the control group(C) in all experimental days. And also The findings showed a non significant (P<0.05) in serum (VLDL-C) concentration in NEPE groups compared to the control group (C).

Serum low density lipoprotein (LDL-C), concentration(mg / dL)in expermental male rats is present in table(12). The results showed non significant (P<0.05) in serum (LDL-C) concentration EPE,NEPE and control groups when compared with each other in day40 of experment. And also showed non significant (P<0.05) between EPE and control in day 60 of experment.

4. Discussion

The results of the present study related to biochemical and microscopic results the diabetic was induced by intraperitoneal injection of alloxan which direct effect on pancreas cells and causes diabetes. The mechanism of alloxan to causes diabetic was the attachenent of alloxan with GLUT2 carrier on the β- cells which leads to fast cellular destruction (9). when alloxan become in side the β- cell, the alloxan converted to dialuric acid, and then generate free radicals , like superoxide its can format hydrogen peroxidase (H2O2), and deduction the Fe3 and formation of hydroxyl radicals, both of them can injury the DNA of the β- cell leading to cell necrosis. As well as ,The H2O2 have been roled to be implicated a rise of intracellular calcium which leaded to the increase initial peak in insulin concentration followed alloxan injection (10) . From the observation of results of experimental diabetic rats throughout the experimental period showed improved in the concentration of enzymes in all groups and the enhancement increase when the period of given was increase.

The results of present study showed that alloxan causes significant decrease in tissue amylase level in all treated and non treated groups because the alloxan causes destruction and necrosis of pancreatic beta cell (10). And the results showed that orally gavages exocrine pancreatic enzyme (EPE) caused an increase in tissuea mylase concentration in all

days of experment (6). Panchbhai and his colleagues (5) reported that a lower tissue amylase levels in diabetics than in non-diabetic group. In their report, As well as, Yadav and his coauthers., (11) and Udia and his colleagues., (12) clarified that low decrease serum amylase activity in diabetics group as compared with control health group. And the results showed that orally gavages exocrine pancreatic enzyme (EPE) caused an increase in tissue amylase concentration in all days of treated group. That were given play us replacement therapy for pancreatic enzyme that not secreted due to destruction of cells of its secretion by alloxan. And the results of present study showed that alloxan causes significant increase in tissue lipase level in non treated groups because the alloxan causes destruction and necrosis of pancreatic cell (10). The high activity of lipase in diabetes groups are associated with an impaired insulin action due to insulin resistance and inadequate insulin secretion (13). And the results showed that orally gavages exocrine pancreatic enzyme (EPE) caused an increase in tissue lipase concentration in all days of treated group with EPE .

Aloulou *et al.*, (14) reported that replacing all functions of these lipases with a single enzyme may be best choice in diabetic rats that suffering from malabsorption of fat.

Chen and Innis.,(15) the authors of the article in gastroenterology suggest that replacement lipase may be suitable as stand alone therapy for PEI (Pancreatic exocrine insufficiency). And the results showed that diabetic caused an increase in tissue lipase concentration. Sikkens and his coauthers., (16) reported the pancreatic exocrine insufficiency (PEI) in relationship to the food consumption happen when the level of pancreatic exocrine enzymes like amylases ,proteases and lipases were deficient to digest food adequately due to diabetes millets,and the results of present study showed that alloxan causes significant decrease in tissue protease level in NEPE groups because the alloxan causes destruction and necrosis of pancreatic cell. The decrease of protease caused by destruction the site of synthesis and secretory vesicles of protease that lead to un secrete of an activated enzyme so diabetes millets influenced on level of secretion of protease (17). And the results showed that orally gavages exocrine pancreatic enzyme (EPE) caused an increase in tissue protease concentration in all days of treated group. As well us, the replacement therapy for pancreatic enzyme play important role in enhancement of digestion of protein (18). The results of present study showed that alloxan causes significant decrease in tissue insulin level in EPE and NEPE groups because the alloxan causes destruction and necrosis of beta cell that responsible to insulin production and secretion (10). The hyperglycemia or diabetes millets occur when the beta cells are destruction that lead to fall in secretion of insulin to the body (19). The insufficiency of insulin or the insensitivity of its receptors which a main causative agent for formation diabetic disease. as known the beta cell on of the pancreatic cell that responsible for insulin that regulate body glucose and destruction of this cells lead to hyper glycemia and (20). If the amount of insulin is insufficient, or if the insulin itself is deficient, then glucose is cannot absorbed normally by the cells that require it, and is not storage in appropriately in the liver and muscles (21). The net effect is persistently high serum glucose concentration. And the results showed that an increase in serum glucose concentration in all days of treated group (G1). because the drug that were given play us replacement therapy for exocrine pancreatic enzyme and not treated the destruction and necrosis of beta cells that mean remaining the lack of insulin production and remain diseased of diabetes mellitus.(14).

Proteins are the main important accessoires of whole cells in the body , so the measures of serum total protein concentration its mean the sum of the amount two kined of proteins present in the blood fluid . These are albumin and globulin. The patients with diabetic disease sufferd from low levels of blood total protein concentration as a result of chronical increase in blood glucose concentration leads to the formation of glycosylated matrials with subsequently prefiltration (a potential elevation in GFR 5%-10% and glomerular

hypertrophy (3). Evans and Capell., (22) clarified that pathoiolgical and physiologic components of diabetic nephropathy are not entirely seen yet incorporate glycosylation of circulating and intrarenal proteins, hypertension, and anomalous intrarenal hemodynamics. The results of present study showed that significant decrease in serum albumin concentration in diabetic NEPE As, well as, Jefferson *et al*., (23) reported in diabetes animal, the concentration of albumin in blood is decreased in diabetes groups., and administration of insulin is required to prevent hypoalbuminemia. Also, Kimball et al., (24) reported in our biochemical studies that insulin stimulates albumin production in the liver by activating gene transcription. And the results showed that orally gavages exocrine pancreatic enzyme (EPE) caused an increase in serum albumin concentration in day 20 of treated group (G2). Because the replacement drug that were given play us replacement therapy for pancreatic enzyme that not secreted because destruction of cells of its secretion by alloxan.

The hyperglycemic subjects including both type 1 and type 2 diabetes indicators of poorly cholesterol absorption efficiency (25). And the results showed that an decrease in serum total cholestrol concentration in all days of NEPE group. because the drug that were given play us replacement therapy for exocrine pancreatic enzyme and not replace the lack of insulin production. Simonen et al., (26). Illustrated in their report that low cholesterol absorption is associated with insulin resistance and metabolic syndrome. Also, demonstrated that high serum triglyceride has a well-established association and impact on increasing cases of high glucose levels in blood (27). Insulin resistance happens if the body does not respond to the insulin you produce, resulting in sugar being unable to reach the cells and staying in the blood stream instead, insulin resistance contributes to high insulin and glucose levels and can lead to uncontrolled diabetes, uncontrolled diabetes may of course, result in high triglycerides (3). Diabetes generally promotes not only quantitative changes in the amount of circulating lipids – particularly an decrease in HDL serum concentration,and increased HDL catabolism are the main changes occurring in diabetes(9). In insulin-resistant case are primarily due to an elevation in hepatic cells production of VLDL particles, postprandial hyperlipidemia (28). The abrormales in lipid contents in diabetes patients, often termed "diabetic dyslipidemia", are typically regognized by elevation of LDL particles. Low density lipoprotein cholesterol (LDL-C) levels may be moderately increased or normal (29).On conclusions, the present study confirmed that the exocrine pancreatic function are very frequently and severely altered in diabetes mellitus rats also can enhancements of some metabolic of diabetes mellitus rats when it orally gavages with replacement therapy of pancreatic exocrine enzyme .

5. Conflict of interest

Authors declare that they do not have any conflict of interest.

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Table 1: tissue amylase concentration (ng/mL) in adult diabetic male rats treated with exocrine panceratic enzyme(EPE) replacement therapy for 60d days. **LSD=1.601845**

Days Groups	20	40	60
G1	745.89±0.24 aC	742.60±0.42 b C	740.99±0.31 cC
G2	749.08±0.23 bB	751.09±0.28 aB	751.45±0.36 aB
C	753.56±1.25 aA	753.47±0.55 aA	754.05±0.56 aA

Table 4-2: tissue lipase concentration (pg/mL) in adult diabetic male rats treated with exocrine pancreatic enzyme(EPE) replacement therapy for 60d days. **LSD=0.4554**

Days Groups	20	40	60
G1	236.15±0.34 aB	235.89±0.25 aC	234.88±0.16 bB
G2	236.92±0.16 aA	236.97±0.16 aB	235.95±0.09 bA
C	237.17±0.03 bA	237.63±0.25 aA	236.2±0.12 cA

Table 3: Serum protease concentration (ng/mL) in adult diabetic male rats treated with exocrine panceratic enzyme(EPE) replacement therapy for 60d days.**LSD=0.01**

Days Groups	20	40	60
G1	239.29±0.04 aC	234.47±0.28 aC	226.61±0.30 aC
G2	292±0.09 aA	284.45±0.47 aA	304.4±0.62 aA
C	264.45±0.29 bB	247.23±0.1 bA	270.01±0.68 aB

Table 4: Serum insulin concentration (ng/mL) in adult diabetic male rats treated with exocrine panceratic enzyme(EPE) replacement therapy for 60d days**.LSD=1.5701**

Days Groups	20	40	60
G1	4.108±0.199 aB	4.103±0.373 aB	4.507±0.374 aB
G2	4.505±0.372 aB	4.704±0.401 aB	5.304±0.318 aB
C	14.502±0.798 aA	14.296±0.948 aA	15.507±0.662 aA

Table 5: Serum glucose concentration (ng/mL) in adult diabetic male rats treated with exocrine pancreatic enzyme(PEP) replacement therapy for 60d days.**LSD=42.74825**

Days Groups	20	40	60
G1	396.06±29.02 aA	431.26±4.94 aA	422.2±13.8 aA
G2	228.91±19.81 aB	237.36±9.00 aB	271.37±14.45 aB
C	159.21±8.95 aC	172.23±6.58 aC	151.95±11.65 aC

Table 6: Serum albumin concentration (mg/dL) in adult diabetic male rats treated with exocrine pancreatic enzyme(PEP) replacement therapy for 60d days.**LSD=0.886663**

Days Groups	20	40	60
G1	2.830±0.273 aA	2.578±0.183 aA	2.874±0.281 aA
G2	3.340±0.145 aB	2.299±0.135 bA	3.018±0.425 aA
C	3.245±0.287 aB	2.944±0.212 bA	3.570±0.569 bB

Table 7: Serum protein concentration (mg/dL) in adult diabetic male rats treated with exocrine pancreatic enzyme(PEP) replacement therapy for 60 days.**LSD=2.100349**

Days Groups	20	40	60
G1	9.332±1.081 aB	8.300±0.272 aB	7.518±0.733 bC
G2	9.462±0.867 aB	8.098±0.545 bB	10.432±0.369 aB
C	12.836±0.424 aA	11.892±0.771 aA	12.858±1.040 aA

Table 8: Serum cholesterol concentration (mg/dL) in adult diabetic male rats treated with exocrine pancreatic enzyme(EPE) replacement therapy for 60 days.**LSD=8.472606**

Days Groups	20	40	60
G1	48.770±2.174 aB	61.374±1.941 bB	53.934±1.644 bB
G2	84.352±3.067 aA	73.324±1.704 bA	73.696±3.268 bA
C	75.870±3.924 aA	77.812±2.422 aA	79.360±4.802 aA

Table 9: Serum triglyceride concentration (mg/dL) in adult diabetic male rats treated with exocrine pancreatic enzyme(EPE) replacement therapy for 60 days. **LSD=12.37191**

Days Groups	20	40	60
G1	33.964±0.942 aA	32.452±1.371 aB	30.372±2.352 bB
G2	40.464±2.596 aA	43.490±1.635 aB	152.718±11.799 bA
C	36.684±1.354 aa	46.300±1.972 aA	46.278±2.187 aA

Table 10: Serum high density lipoprotein (HDL-C)concentration (mg/dL) in adult diabetic male rats treated with exocrine pancreatic enzyme(EPE) replacement therapy for 60 days. **LSD=2.180119**

Days Groups	20	40	60
G1	14.630±0.445 aB	11.416±0.472 bC	10.170±0.323 bB
G2	14.891±1.545 aB	15.237±1.317 aB	17.559±0.388 bA
C	17.752±0.428 aA	18.660±0.426 aA	18.315±0.487 aA

Table 11: Serum very low density lipoprotein (VLDL-C)concentration (mg/dL) in adult diabetic male rats treated with exocrine pancreatic enzyme(EPE) replacement therapy for 60 days.**LSD=2.474383**

Days Groups	20	40	60
G1	6.792±0.188 aA	6.490±0.274 aB	6.074±0.470 aB
G2	8.092±0.519 aA	8.698±0.327 aB	30.543±2.359 bA
C	7.336±0.270 aA	9.260±0.394 aA	9.255±0.437 aA

Table 12: Serum low density lipoprotein (LDL-C)concentration (mg/dL) in adult diabetic male rats treated with exocrine pancreatic enzyme(PEP) replacement therapy for 60 days.**LSD=9.088984**

Days Groups	20	40	60
G1	27.347±1.702 aC	43.465±2.313 bA	37.639±2.283 bB
G2	61.367±4.053 aA	49.388±2.790 bA	25.592±2.905 cA
C	50.781±4.048 aB	49.890±2.267 aA	51.788±4.766 aA

µ±SE -C= control -G2= Animals intraperitoneally injected with alloxan and treated with exocrine pancreatic enzyme(EPE) replacement therapy

-G1= Animals intraperitoneally injected with alloxan and nontreated with exocrine pancreatic enzyme(NEPE) replacement therapy

- Capital letters denote differences between groups, P>0.05.

- Small letters denote differences within group, P>0.05.