ISSN: 2320 - 4230

Journal of Drug Discovery and Therapeutics

Available Online at www.jddt.in

CODEN: - JDDTBP (Source: - American Chemical Society)

Volume 3, Issue 28, May, 2015, 45-52

Research Article

Study of Possible Correlation between Pancreatitis and Daily Consumption of Catha edulis in Experimental Animals

Mohamed M. Rezq Battah[1], Doa'a Anwar Ibrahim[2] and Amani M. Shamsher[3]

[1,2]Department of Pharmacology, Faculty of Pharmacy, University of Science and Technology, Sana'a, Yemen

[3] Histopathology Department , UST hospital Lab., Sana'a, Yemen

Received 01 May 2015; Accepted 21 May 2015

ABSTRACT

Khat (*Catha edulis*) is widely used throughout the world especially in Yemen and East Africa**;** It becomes a serious public health phenomena in Yemen that affects the health features of life. The present study evaluates the possible correlation between pancreatitis and daily consumption of khat leaves or extract in rabbits. Thirty adult male rabbits were divided randomly into five groups, 6 each: (I)control, (II)L-arginine, (III)pioglitazone, (IV)khat leaves and (V)khat extract. From group (II) to (V) were given L-arginine (450mg/100g b.wt) i.p to induce acute pancreatitis. Treatments were given via oral gavage for 4 weeks. FBS, p.amylase, lipase, insulin, TAS were measured as well as histopathology examination of pancreas. FBS was significantly reduced, whereas insulin level was increased in both groups of khat (leaves and extract). Additionally, p.amylase and lipase levels were decreased, and increased in TAS level. These results similar to that in pioglitazone group compared to L-arginine group, which found that significantly increased in p.amylase, lipase and FBS, while decreased in insulin and TAS levels. All these were accompanied with pancreatic histopathological improvements seen in khat leaves, khat extract and pioglitazone groups when compared to L-arginine group. The improvement of acute pancreatitis induced by L-arginine reflected remarkably on FBS and insulin levels in khat treated groups, this indicating that there is no correlation between acute pancreatitis and daily consumption of khat.

Keywords: Acute pancreatitis, L-arginine, *Catha edulis***,** Pioglitazone

INTRODUCTION

The most common name of *Catha edulis* frosk is khat, and the family is *Celastraceae* [1]. Khat is widely used throughout the world especially in Yemen and East Africa [2], during the last decade(s) it is became more common in the so-called western or developed countries which are prohibited by law in several European and American countries [3]. The WHO described that khat chewing has come to be a public problem that has effects on the health features of life[4]. Khat plant contains a variety active ingredients in the leaves and young branch, the most important of these active ingredients are: alkaloids, glycosides, terpenoids, flavonoids, ascorbic acid, tannins, saponins and more than 40 other types of alkaloids. Cathinone considerd the most important of them. It has d-(+)- amphetamine like action and structure. Cathinone is originating mostly in the fresh leaves and shoots; it is comparatively unstable and quickly metabolized during maturation to norephedrine and cathine (norepseudophedrine). It is released during chewing within 15-45 min and detected in plasma within 24 hrs after khat chewing and stimulating euphoria, analgesia and psycho-stimulant effects[5,6]. Also there are other alkaloids as: cathine, phenylpentenylamines, cathedulines. The central nervous activity and mechanism of action of cathinone are similar to that of amphetamine, which affecting by releasing catecholamine from presynaptic storage sites. The consumption of khat leaves has been associated with many clinical and adverse effects [2] such as, anorexia, mydriasis, insomnia, endocrinological disturbances, hyperthermia and acute autonomic responses (Hassan *et al*., 2000)[7]. In addition, prolonged chewing of these leaves may result in psychoneurological disturbances such as neurosis [8], increasing diastolic blood pressure [9], and vasoconstriction of coronary vasculature [10]. Khat extract consumption may affect liver and kidney tissues through induction of lipid peroxidation and oxidative stress [11]. Cathinone has sympathomimetic actions that would be probable to increase levels of catecholamine in plasma, which increased the level of blood glucose. Acute pancreatitis is a rapid-onset inflammation non-pancreas; it is a reversible inflammatory disorder that causes focal edema, fat necrosis and widespread hemorrhagic parenchymal necrosis. Acute abdominal pain and increasing the concentration of serum lipase and amylase

*Corresponding author: Mohamed M. Rezq Battah |E-mail: pri.priyankaph@gmail.com

are the most common symptoms of it [12]. The experimental models of acute pancreatitis that similar to the human state are an essential tool to increase the knowledge of the complex mechanisms and in developing therapeutic strategies for this disease. In addition, these models may be divided according to the method induction of acute pancreatitis into: invasive models, and non-invasive models. L-arginine is non-invasive model [13] that can produce acute necrotizing pancreatitis when administrated with large dose [14].

Pioglitazone belongs to thiazolidinediones group (TZDs) that used for treatment of type 2 diabetes [15]. It may offers a new therapeutic options with treatment of acute pancreatitis [16], and increases function of β cell with protection of islet cell changes [17].

MATERIALS AND METHODS

Fresh khat leaves were brought from local markets in Sana'a, Yemen. The plant was identified and authenticated at Pharmacognosy Department, UST. The leaves were washing with distilled water to remove dust and debris.

Drugs and chemicals:

L-arginine monohydrochloride and ethanol were purchased from HiMedia Company, p.amylase and lipase estimation kits from Roche Company, Glucose and insulin estimation kits from Sigma Aldrich, Total Anti-oxidant status kits from Randox Laboratories Ltd, and Pioglitazone (Actazone^{R}) from Asia Pharmaceutical Industries.

Experimental Animals:

Thirty adult male New Zealand White Rabbits with weight range from 1.5-2 kg and aged 9 months ±1 week were supplied by the Faculty animal house. Animals were acclimatized at laboratory condition for five days before commencement of experiment. They were kept in a clean cage with free feed and water under observation. Human therapeutic oral dose was extrapolated to rabbit according to method of Paget's and Barnes, 1964[18].

Plant extract's preparation:

1kg of *Catha edulis* (stem tips and leaves) were extracted with 5 L of Water/Ethanol (HiMedia) mixture (70/30%) by using a Soxhlet apparatus-Buchi-type. Then, the final extract was evaporated and dried by using a Rotary evaporator- Buchi-type under vacuum at 40◦C to eliminate all traces of mixture. The resulting (100 g dried extract) was saved in a desiccator until the time of use [19, 20].

Induction of Acute Pancreatitis:

L-arginine monohydrochloride was prepared in 20% solution with saline and the pH was adjusted to 7.0.The volume of L-arginine monohydrochloride solution was calculated and administrated to animals according to their body weight by intraperitoneal route [21].

Study Design:

30 rabbits were divided randomly into five groups, (each containing 6 animals), Group I was kept as (control), given free access of food and water, from group II to V were given 20% L-arginine (450 mg/100 g body weight) [21] i.p at the last 3 days of experiment to induce pancreatitis, additionally, group III was given pioglitazone (2.7 mg/kg body weight)[22], group IV was given khat leaves (2 g/kg body weight) [23], and group V was given khat extract (10 mg/kg body weight) [24] suspended in distilled water according to the body weight of each rabbit. The rout of administration of testing drugs in all groups was through oral gavage feeding, and the duration of this study was 4 weeks according to method of Sakr *et al.*, 2011[11].

Biochemical Estimations:

At the end of this experiment, animals were kept fasting for at least 8 hours, blood samples were collected to measure FBS, Insulin levels, TAS, Serum of P.amylase and lipase.

Histopathological study of pancreas:

The last day of experiment, rabbits were anesthetized and sacrificed; pancreas was removed and kept in 10% formaldehyde solution till tissues were prepared for histological examination. Tissue processed and then stained with hematoxylin & eosin (H&E)[25]in University of Science and Technology hospital lab. The study protocol was approved by the University Ethics committee before any experiment can be conducted.

Statistical Analysis:

Data were summarized as means ± SEM. One way analysis of variance (ANOVA) followed by Dunnet's multiple comparison test was used to conduct the significance of association using SPSS program version 21. Differences were considered significant at P values of less than 0.05

RESULTS

Measurement of pancreatic function tests:

Rabbits treated with L-arginine were shown a significant increase in serum p. amylase and lipase levels as well as a reduction in TAS compared with control group. While treated groups either with pioglitazone or khat (leaves or extract) were ameliorated the toxic effect of L-arginine as they reduced the levels of serum p. amylase and lipase and increased the TAS levels significantly (table 1).

Table 1: Effect of L-arginine, pioglitazone, khat leaves and khat extract on serum of p. amylase (U/L), lipase (U/L) and TAS (µmol/l)

Treatment	Lipase	P. amylase	TAS
Control	150.2±20.53	298.0±28.56	332.8±25.68
L-Arginine	268.3±2.86*	372.2±10.51*	210.3±4.59*
Pioglitazone	99.7±9.54**	227.3±7.90**	393.4±0.63**
Khat leave	63.7±2.01**	179.3±9.94**	290.2±26.13**
Khat extract	101.5±6.68**	230.6±9.28**	390.1±1.84**

Data expressed as mean ± SEM, *p-value is <0.05, which is statistically significant compared with control,**p-value is <0.05, which is statistically significant compared with standard drug (L-arginine). TAS; Total antioxidant status, p. amylase; pancreatic amylase.

Measurement of insulin levels and FBS:

Rabbits treated with L-arginine resulted in a significant increase in FBS as well as a reduction in insulin levels compared with control group. In contrast, treated groups either with pioglitazone or khat (leaves or extract) were ameliorated the toxic effect of L-arginine as they reduced the levels of FBS and increased the insulin levels significantly (table 2).

Table 2: Effect of L-arginine, pioglitazone, khat leaves and khat extract on FBS (mg/dL) and insulin levels (µIU/mL)

Treatment	Insulin	FBS
Control	11.3±0.50	131.4±3.45
L-Arginine	8.1±0.69*	183.4±5.22*
Pioglitazone	14.1±0.39**	89.4±1.23**
Khat leave	12.1±0.44**	82.6±2.40**
Khat extract	12.7±0.41**	80.5±3.19**

*Data expressed as mean ± SEM, *p-value is <0.05, which is statistically significant compared with control,**p-value is <0.05, which is statistically significant compared with standard drug (L-arginine). FBS: Fasting blood sugar.*

However, khat extract group showed a significant reduction in the body weight when compared to control group. In addition, pioglitazone group showed a significant increase in the body weight compared to L-arginine group (figure 1).

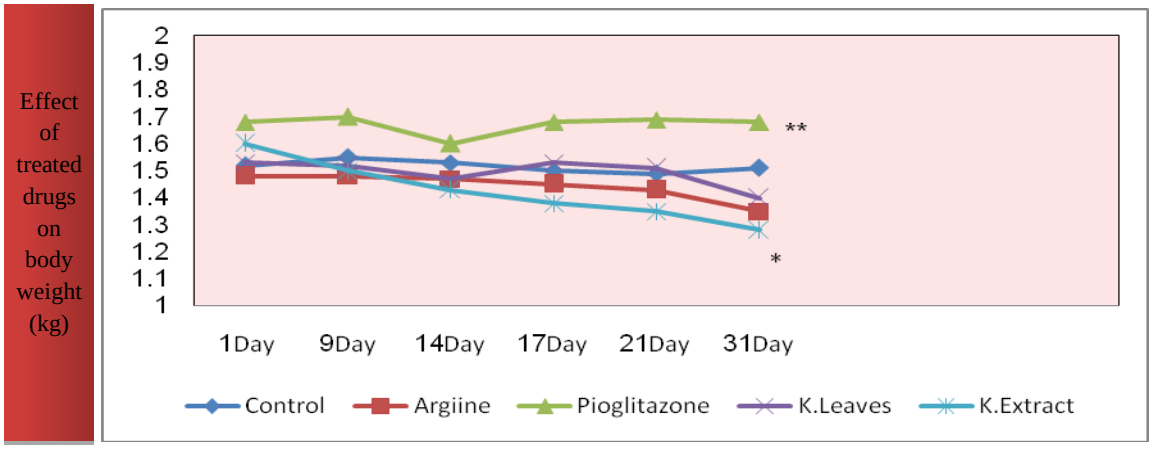

Figure 1: Body weight differences according to type of treatment

**Significant as compared with control at (P< 0.05), ** Significant as compared with L-arginine at (P< 0.05)*

Histopathological findings:

In corresponding with current results of biochemical tests of p.amylase, lipase, TAS, FBS and insulin levels in present study, the histopathological changes of the pancreatic section of rabbits were significantly observed in L-arginine group, showed that ductal proliferations of pancreatic tissue with sever reactive atropia at the peripheral lobules compared to control group. However, in the other groups, it was significantly observed that these changes improved when treated with either pioglitazone or khat (leaves or extract), and the toxic effect of L-arginine was ameliorated, the pancreatic sections of rabbits in these groups showed that a normal pancreatic tissue with islet of Langerhans, pre pancreatic connective tissue without abnormal inflammatory cells, and improved all histopathological changes compared to L- Arginine group (standard drug). As shown in figure 2.

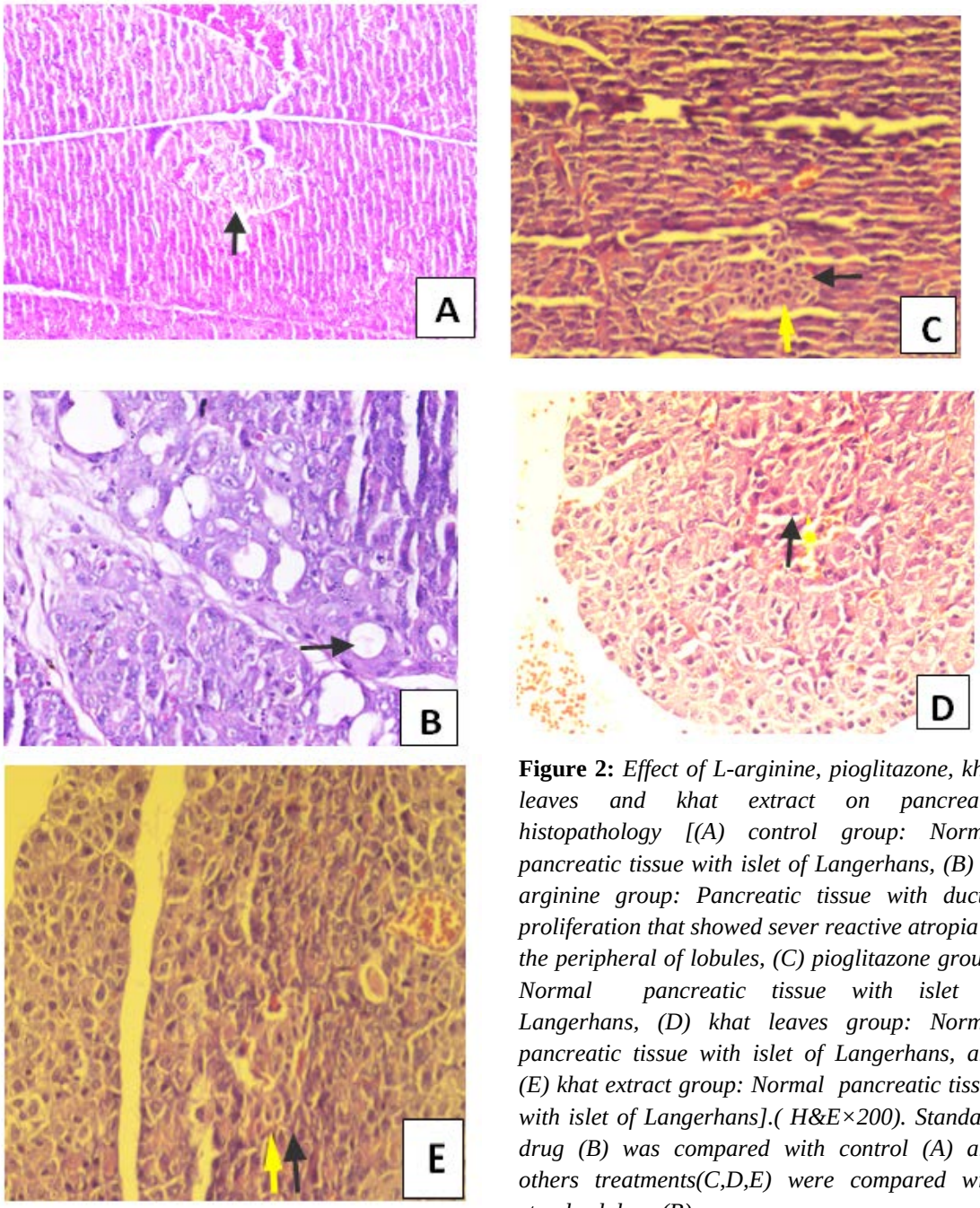

Figure 2: *Effect of L-arginine, pioglitazone, khat leaves and khat extract on pancreatic histopathology [(A) control group: Normal pancreatic tissue with islet of Langerhans, (B) L-arginine group: Pancreatic tissue with ductal proliferation that showed sever reactive atropia at the peripheral of lobules, (C) pioglitazone group: Normal pancreatic tissue with islet of Langerhans, (D) khat leaves group: Normal pancreatic tissue with islet of Langerhans, and (E) khat extract group: Normal pancreatic tissue with islet of Langerhans].(H&E×200). Standard drug (B) was compared with control (A) and others treatments(C,D,E) were compared with standard drug (B).*

DISCUSSION

Khat plant (*Catha edulis*) chewing are widely used by group of peoples as a social habit in Yemen, it's biological effects are incompetently examined and debatable [26,27]. In this study, the non-invasive experimental model was based on the previous reports [21,28] that have used 450 mg/100 g body weight of L-arginine intraperitoneal injection for induction of acute necrotizing pancreatitis in animal's model.

In present study, administration of 450mg/100g L-arginine IP for rabbits led to acute pancreatitis, which significantly increased levels of serum p.amylase, lipase and FBS, in contrast, significantly decreased insulin level and TAS. This effect probably due to production of reactive oxygen species that increase inflammatory mediators like: oxygen free radicals and nitric oxide. In addition, L-arginine may increase oxidative stress and nitric oxide syntheses that contributed to the pancreatic injury in this model. These effects reflected remarkably on the histopathological findings of this group which significantly showed changes on the pancreatic section of rabbits, these changes were revealed that ductal proliferations of pancreatic tissue with sever reactive atropia at the peripheral lobules. Our results were similar to the previous results of Hegyi *et al.*, 2004 [14,29]. However, the present study demonstrated that treated group with pioglitazone (2.7 g/kg) efficiently reduced the severity of L-arginine induced acute pancreatitis in rabbits; it significantly decreased levels of serum p.amylase and lipase as well as increased TAS levels, moreover, it was significantly increased insulin level and decreased FBS. All these findings indicated that pioglitazone has protective effects from pancreatitis and may improve the functions of pancreas. The reasons for that may be attributed to decrease the severity of Sodium Taurocholate (STC)-induced sever acute pancreatitis, and enhance β cell functions with preservation of islet cell changes. Moreover, pioglitazone is a PPAR-γ agonist and insulin sensitizer; it decreases production of some mediators that may cause resistance of insulin, like resistin and tumor necrosis factor α (TNF α), so it decreases insulin resistance. In addition, Pioglitazone enhances peripheral and hepatic sensitivity of insulin [30,31]. Additionally, Pioglitazone thereby increases inhibiting of gluconeogenesis and increases peripheral and splanchnic glucose uptake [32]. These findings were in agreement with the histopathological changes in this group that showed a significant improvement of the pancreatic section of rabbits, and improved the pancreas functions with reoccurrence it to normal without abnormal inflammatory cells, similarly to control group. In consistent with previous reports [17,16] pioglitazone may offers a new therapeutic options with treatment of acute pancreatitis, it significantly decreased levels of serum p.amylase, lipase, and increased TAS levels.

From the results obtained in present study, both khat groups (leaves and extract) showed a significant decrease in the serum of p.amylase and lipase, and increase in TAS levels -resembling with pioglitazone group- indicating that both khat leaves and extract have pancreatic protective effects, and possibly they have anti-inflammatory effects by flavonoid constituents that existing with large amounts in khat, which reduced the inflammatory mediators. In addition, khat leaves and extract probably have improved pancreas functions, and reduced the severity of L-arginine induced acute pancreatitis in rabbits through antioxidant activity as they contain flavonoid constituents that inhibit the activities of group of enzymes such as; monooxygenase, cyclooxygenase, lipoxygenase, xanthine oxidase, and protein kinases. Moreover, antioxidant properties of the flavonoids include their protection against iron-induced free radical reactions that generated at the active site of these enzymes [33]. Additionally, khat leaves and extract contain some essential components of metallo-enzymes, for example; zinc, copper, selenium, manganese, iron and ascorbic acid, are essential in intra- and extra-cellular antioxidant protection [34]. This finding was contradictory with the study of Al-Zubairi *et al.* 2003 and Sakr, *et al.,* 2011 [11,35].

In the current work, the results showed a significant decrease of FBS and increase of insulin levels in both khat groups (leaves and extract) feeding rabbits. This may be referred to either the presence of flavonoid as free radical scavenger or tannins and inorganic ions existing in both khat (leaves and extract) probably induced the delay of glucose absorption, that may contributed to reduce the levels of blood sugar [1]. Moreover, both khat (leaves and extract) may contributed to the delay of gastric emptying which in turns decreased the levels of blood sugar after food intake[36]. Moreover, Ahmed and Al-Qirbi, 1993 [37]; Ramadan *et al.,* 1979[38] were in agreement with current study, as they reported that khat have a hypoglycemic effect and significantly decreased the levels of blood sugar in chewers. This effect probably attributed either to induce peripheral glucose uptake or to interact with β-cells of the pancreas directly. Additionally, Al Habori and Al Mamary, 2004 [39];Taleb and Bechyn, 2009[40] were in consistence with our results.

Moreover, these findings supported by the histopathological findings in both khat groups (leaves and extract), that were in agreement with current results of biochemical test, and showed a significant improvement of the pancreatic section of rabbits, with improved the pancreatic functions and reoccurrence it to normal pancreatic tissue and islet of Langerhans without abnormal inflammatory cells.

On the other hand, our findings were contradictory with previous studies [41,42] that showed a significant increase in the levels of serum glucose [2,41].

In this work, khat extract group showed a significant reduction of the body weight, though khat leaves group showed no significant, and was slightly reduction. These findings probably attributed to anorexigenic effect of khat and its constituents that may be found in extract with higher concentration than leaves. The anorexigenic effects of khat might be secondary to central mechanisms mediated via cathinone, which existing as a main active ingredient of khat, and it has a structure related to amphetamine, that affect appetite centrally via affecting the hypothalamus and peripherally by delaying gastric emptying [43,44]. In addition, the presence of tannins and inorganic ions in khat extract possibly contributed to delay the absorption of food that causes malnutrition and loss of body weight [45]. In contrast, pioglitazone group showed a significant increase of the body weight, may be due to water retention [30], and that was in agreement with Choi *et al*., 2007[17].

Indeed, the improvement of acute pancreatitis reflected remarkably on insulin and FBS levels in this experimental animal. Meanwhile, there were a good correlation between acute pancreatitis and diabetes mellitus especially type 2, this relationship may be obtainable in an incidence of diabetic patients in acute pancreatitis sequences and in opposite higher risk for acute pancreatitis in diabetic patients [46-48]. Moreover, acute pancreatitis occurrence in diabetic patients is 2-3 times greater than in non-diabetic patients [49]. Indeed, acute pancreatitis risk can be measured higher in diabetic patients [50], and acute pancreatitis advancement may be negatively influenced by diabetes. This correlation between acute pancreatitis and diabetes mellitus reflected contradictory on the decreasing of FBS and increasing of insulin levels in both khat leaves and extract groups of this study, indicating that no correlation between acute pancreatitis and khat consumption.

CONCLUSION

The present study suggested that, there is no correlation between acute pancreatitis and daily consumption of khat. Both khat leaves and extract consumption have pancreatic protective effect as well as improving its functions. They significantly contradict the severity of L-arginine induced pancreatitis by reducing the levels of serum p.amylase and lipase through increasing TAS levels. On the other hand, both khat leaves and extract significantly decreased fasting blood sugar and increased insulin levels. All these results were supported by histopatholological findings.

ACKNOWLEDGEMENT

This study was funded by University of Science and Technology. The author would like to thank Faculty of Pharmacy and UST hospital lab. team as well as Dr. Farouk Al-qadasi for statistics analysis

CONFLICT OF INTERESTS

There is no conflict of interests regarding the publication of this paper.

REFERENCES:

1. Mahmood S A and Lindequist U, A pilot study on the effect of cathaedulisfrosk., (Celastraceae) on metabolic syndrome in wokwrats. Afr J. Trad. CAM. 2008;5(3):271 – 277.
2. Al-Motarreb A, Al-Habori M, and K. Broadley J. Khat chewing, cardiovascular diseases and other internal medical problems: the current situation and directions for future research. *Journal of Ethnopharmacology*. 2010; 132, (3):540–548.
3. Balint, E. E, Khat *(Catha Edulis)* A controversial plant: blessing or curse?.*Cumulative impact factor*, *3.608.* 2012.
4. Kassim S, Islam S, and Croucher R. Validity and reliability of a Severity of Dependence Scale for khat (SDS-khat). *Journal of Ethnopharmacology*. 2010;132 (3): 570–577.
5. Graziani M, Milella MS, and Nencini P. Khat chewing from the pharmacological point of view: An update. Sub. Use Misuse. 2008;43: 762-783.
6. Al-Motarreb A, Baker K, and Broadley KJ. Khat: Pharmacological and medical aspects and its social use in Yemen. *Phytotherapy Res*. 2002;16: 403- 413 .
7. Hassan NA, *et al*. The effect of Qat chewing on blood pressure and heart rate in healthy volunteers. Tropical Doctor, Apr 2000;30(2):107-8
8. Hoffman R, and Al'Absi M. Khat use and neurobehavioral functions: suggestions for future studies. *Journal of Ethnopharmacology*. 2010; 132 (3): 554–563.
9. Getahun W, Gedif T, and Tesfaye F. Regular Khat (*Catha edulis*) chewing is associated with elevated diastolic blood pressure among adults in Butajira, Ethiopia: a comparative study," *BMC Public Health*. 2010; 10 (390).

10. Ali WM, Zubaid M, Al-Motarreb A, *et al*. Association of Khat chewing with increased risk of stroke and death in patients presenting with acute coronary syndrome. *Mayo Clinic Proceedings*. 2010; 85(11): 974–980.

11. Sakr H F *et al*. Khat (*Catha edulis*) Extract Increases Oxidative Stress Parameters and Impairs Renal and Hepatic Functions in Rats, *Bahrain Medical Bulletin, March 2011; 33(1).*

12. Matalka I I, Mhaidat N M, &Fatlawi L A. Antioxidant activity of simvastatin prevents L-arginine-induced acute toxicity of pancreas. International journal of physiology, pathophysiology and pharmacology, 2013; *5*(2): 102.

13. Su KH, Cuthbertson CH &Christophi CH. Review of experimental animal models of acute pancreatitis. HPB, 2006; 8: 264_286.

14. Hegyi P, *et al.* L-arginine-induced experimental pancreatitis. World J Gastroenterol. 2004 July 15; 10 (14): 2003-2009.

15. Katzung BG; Masters SB, Trevor A J. Pancreatic Hormones &Antidiabetic Drugs. In: (eds.) Basic& clinical pharmacology. 11th ed. China: The McGraw-Hill Companies, Inc.; 2009. p928 – 952

16. Xu P, *et al*. Pioglitazone attenuates the severity of sodium taurocholate-induced severe acute pancreatitis. *World J Gastroenterol*2007 April 7; 13(13): 1983-1988.

17. Choi SH, Zhao ZS, Lee YJ, Kim SK, Kim DJ, Ahn CW, Lim SK, Lee HC and Cha BS. The different mechanisms of insulin sensitizers to prevent type 2 diabetes in OLETF rats. *Diabetes Metab. Res. Rev.*, 2007; 23(5): 411-418.

18. Paget GE, and Barnes JM. From toxicity tests. In evaluation of drug activities: pharmacometrics. Edited by: Laurence DR, Bacharach AL.1964; 1: 50-161.

19. DallakMA , *et al*., In vivo acute effect of orally administrated hydro ethanol extract of Catha Edulis on blood glucose levels in normal, glucose- fed hyperglycemic , and Alloxan induced diabetic rats. Saudi Med J 2010;31(6)

20. Alsalahi A, *et al.* Toxicological Features of Catha edulis (Khat) on Livers and Kidneys of Male and Female Sprague-DawleyRats:ASubchronic Study, Evidence-Based Complementary and Alternative Medicine. 2012; (829401): 11.

21. Dawra R and Saluja AK. L-arginine-induced experimental acute pancreatitis. March 2012; 1.0.

22. Gad MZ, Ehssan NA, Ghiet MH, Wahman LF.. Pioglitazone versus metformin in two rat models of glucose intolerance and diabetes.. *Pak. J. Pharm. Sci*.2010;23(3):305-312

23. AL-Rajhi WI and Yousef OM. Effects of Catha Edulis Abuse on Glucose, Lipid Profiles and Liver Histopathology in Rabbit. *Journal of Life Sciences and Technologies*. March 2013;1(1).

24. Abderrahman S M & ModallalN. Genotoxic effects of Catha edulis (Khat) extract on mice bone marrow cells. *Jordan Journal of Biological Sciences*,2008;*4*: 165-172.

25. Jelodar G, Mohsen M &Shahram S. Effect of walnut leaf, coriander and pomegranate on blood glucose and histopathology of pancreas of alloxan induced diabetic rats. *African Journal of Traditional, Complementary and Alternative Medicines*, 2008; *4* (3): 299-305.

26. Carvalh oF. The toxicological potential of khat. *Journal of ethnopharmacology*, 2003;*87*(1): 1-2.

27. Dimba E A O, Gjertsen B T, Bredholt T, Fossan K O, Costea D E, Francis G W, ...&Vintermyr O K. Khat (Catha edulis)-induced apoptosis is inhibited by antagonists of caspase-1 and-8 in human leukaemia cells. *British Journal of Cancer*, 2004;*91*(9): 1726-1734.

28. Tani S, Itoh H, Okabayashi Y, Nakamura T, Fujii M, Fujisawa T,&Otsuki M. New model of acute necrotizing pancreatitis induced by excessive doses of arginine in rats.*Digestive diseases and sciences*, 1990; *35*(3): 367-374.

29. BiradarS,&Veeresh B. Protective effect of lawsone on L-Arginine induced acute pancreatitis in rats. 2013

30. Cheng AY and Fantus IG. Oral antihyperglycemic therapy for type 2 diabetes mellitus. *CMAJ*, 2005;172(2):213-226.

31. Tjokroprawiro A. New approach in the treatment of T2DM and metabolic syndrome (focus on a novel insulin sensitizer). *Acta Med. Indones*, 2006;38(3): 160-166.

32. Waugh J, Keating GM, Plosker GL, Easthope S and Robinson DM. Pioglitazone: a review of its use in type 2 diabetes mellitus. *Drugs*, 2006;66(1): 85-109.

33. Cao G, Sofic E and Priorr, L. Antioxidant and prooxidant behavior of flavonoids: structure-activity relationships. Free Radical Biol. & Med. 1997;22 (5): 749-760.

34. Zheng Y, Li X K, Wang Y &Cai L. The Role of Zinc, Copper and Iron in the Pathogenesis of Diabetes and Diabetic Complications: Therapeutic Effects by Chelators*. *Hemoglobin*, 2008*;32*(1-2): 135-145.

35. Al-Zubairi A, Al-Habori M & Al-Geiry A. Effect of Catha edulis (khat) chewing on plasma lipid

peroxidation. *Journal of ethnopharmacology*, 2003;*87*(1): 3-9.

36. Heymann T D, Bhupulan A, Zureikat N E K, Bomanji J, Drinkwater C, Giles P & MURRAY-LYON I M. Khat chewing delays gastric emptying of a semi-solid meal. *Alimentary pharmacology & therapeutics*, 1995; *9*(1): 81-83

37. Ahmed M B & El-QirbiA B. Biochemical effects of Catha edulis, cathine and cathinone on adrenocortical functions. *Journal of ethnopharmacology*, 1993*;39*(3): 213-216.

38. Ramadan M A, Tash F M, Fahmi M &Abul-kheir F A. Metabolism changes caused by khat consumption in Yemen.*Journal of Yemen Centre for Studies and Research*, 1979; *3*: 35-44.

39. Al-Habori M & Al-Mamary M. Long-term feeding effects of Catha edulis leaves on blood constituents in animals. *Phytomedicine*, 2004*;11*(7): 639-644.

40. Taleb M, Bechyn M. Effect Of*CathaEdulis*LeavesOn Plasma Glucose. Agricultura Tropica EtSubtropica. 2009; 42 (1)

41. Broadley K J. Autonomic pharmacology, in *Autonomic Nervous System*, Taylor & Francis, London, CRC Press, 1996.

42. Bajubair M A. *Effect of khat on the functions of the liver, the kidneys and on the blood glucose level* (Doctoral dissertation, Thesis]. Sudan: Faculty of Pharmacy, University of Khartoum), 1997.

43. Doong ML, Lu CC, Kau MM, Tsai SC, Chiao YC, Chen JJ, et al. Inhibition of gastric emptying and intestinal transit by amphetamine through a mechanism involving an increased secretion of CCK in male rats. Br J Pharmacol, 1998; 124: 1123 – 1130

44. Tucci SA. Phytochemicals in the control of human appetite and body weight. Pharmaceuticals, 2010; 3:748-763.

45. Murray CD, Le Roux CW, Emmanuel AV, Halket JM, Przyborowsk AM, Kamm MA, Murray-Lyon IM. The effect of khat (Catha edulis) as an appetite suppressant is independent of ghrelin and PYY secretion. *Appetite,* 2008;51(3):747–750.

46. Yadav D. & Lowenfels AB. Trends in the epidemiology of the first attack of acute pancreatitis: a systematic review. Pancreas, 2006; 33: 323-330.

47. Frey CF, Zhou H, Harvey DJ & White RH. The incidence and case-fatality rates of acute biliary, alcoholic, and idiopathic pancreatitis in California, 2006;1994-2001.Pancreas ;33: 336-344.

48. Lankisch PG, Karimi M, Bruns A &Lowenfels AB. Temporal trends in incidence and severity of acute pancreatitis in Luneburg County, Germany : a population-based study. Pancreatology , 2009; 9: 420-426.

49. Girman CJ, Kou TD, Cai B & Katz L. Patients with type2 diabetes mellitus have higher risk for acute pancreatitis compared with those without diabetes. Diabetes, Obesity and Metabolism, 2010;12: 766-771.

50. Gonzalez-Perez A, Schlienger RG & Garcia Rodriguez LA. Acute pancreatitis in association with type 2 diabetes and antidiabetic drugs. A population-based cohort study. Diabetes Care, 2010; 33(13): 2580-2585.