EFFECT OF LONG TERM CONSUMPTION OF SELECTED GREEN LEAFY PORRIDGES ON LIVER ENZYME PROFILES OF WISTAR RATS

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Introduction

Searching dietary remedies to prevent occurrence and to overcome complications of type II diabetes is a prevailing effort among the world population as the number of diabetic cases in the worldhas predicted to be up surged to 70 million by 2025(Hoskoteand Joshi, 2008). Nearly one fourth of Sri Lankan population is suffering from disglycaemia (Wijesuriya, 2010) and 90% of these patients consume herbal plants to treat diabetes with or without prior advice from a physician (Ediriweera, 2009). *Scopariadulsic* (SD) (Sinhala – Walkoththamalli, Tamil – Sarkaraivembu)and*Hemidesmusindicus*(HI) (Sinhala – Iramusu, Tamil - Nannari)are herbal plants,commonly used by diabetic patients as dietary remedies. *Scopariadulcis* (SD) is a tropical plant, used as fresh or dried plant extract to treat diabetes and many other ailments(Das and Chakraborty, 2011). *Hemidesmusindicus*isalso a commonly consumed herb and the leaves elicit antiasthmatic, bronchodilatory, anti-inflammatory, antiallergic, and immune suppressive properties (http://www.naturalhealthcure.org/).As the hypoglycaemic effectsof water/ ethanolic extracts of these leavesis proven (Latha*et al*, 2004), porridge made with these leaf extracts could be used as a dietary remedy for diabetics.A previously carried out study revealed that SD and HI porridges elicit low Glycaemic Index values (39±8 and 40±8 respectively) and peak blood glucose reduction percentages of 39% and 40% respectively, when compared to a glucose control (Senadheera and Ekanayake, 2011). However, no data is available on the toxic effects that can arise with the long term consumption of the above mentioned porridges. The aim of the present study was to evaluate the hepato-toxic effects (in Wistar rats) of SD and HI leafy porridges with long term consumption.

Methodology

Porridge of the SD and HI leaves were prepared according to a standard recipe considering the palatability (in final porridge - leaves: coconut milk: rice = 13:90:25). All ingredients were same in both porridges except the leaf variety. After preparing the porridge, a single portion size for a human contained 35mg/Kg BW amount of solid leaf content. Ethical approval for the study was obtained from the ethics review committee of Faculty of Medical Sciences, University of Sri Jayewardenepura. The study was carried out with (Streptozotocin) STZ induced diabetic male albino Wistar rats (28 days old, weight 150 -200g) purchased from the Medical Research Institute, Sri Lanka. Rats with fasting blood glucose level above 126mg/dL or random blood glucose above 300 mg/dL were divided in to six groups with 7 rats in each [2 green leafy porridge groups, coconut milk porridge group, diabetic control (DM) and normal control groups] without any significant difference ($p \leq 0.05$) in the mean blood glucose levels. Coconut milk porridge (CM) was included to determine whether the effects were due to coconut milk. Porridges were incorporated in to the normal WHO recommended diets of the rats in order to contain a 6 times high dose of porridge than human dose (Reagan-Shaw et al, 2008). Study

was continued for six months. Liver enzymes (AST, ALT, gamma GT) and creatinine levels were measured at the end of the third month. At the end of the study period rats were euthanized and the weights of the liver and pancreas were measured.

Discussion and Conclusion

Table 1 AST, ALT, Gamma GT and Creatinin levels (±standard deviation) in rats

Group	AST (U/L)	ALT (U/L)	Gamma GT (IU/L)	Creatinin (mg/ dL)	BW: liver wt	BW: pancreas wt
DM##	82.5±16.3[a]	23.7±2.1[a]	3.0±1.7[ab]	0.45±0.5[a]	23±1.5	199±12
	(75 – 81)	(22 – 26)	(1.7 – 5.9)			
SD#	88.9±15.1[a]	25.1±2.5[a]	2.5±1.9[ab]	0.5±0.1[a]	23±1.5	232±36
	(62 – 110)	(22 – 29)	(0.8 – 5.9)			
HI##	69.7±10.2[b]	27.6±3.6[a]	4.0±0.4[b]	0.51±0.1[a]	21±1.4	208±17
	(56 – 81)	(23 – 33)	(3.4 – 4.2)			
CM##	75.6±7.0[a]	25.3±2.4[a]	4.6±0.9[b]	0.41±0.1[a]	23±1.2	271±119 (n=3)
	(68 – 85)	(22 – 28)	(3.4 – 5.9)		(n=3)	
Normal#	64.1±9.7[b]	15.3±3.4[b]	2.2±1.6[a]	0.35±0.0[b]	23±2.0	229±29
	(52 – 78)	(9 – 18)	(0.8 – 4.2)			

(#n=7, ##n=6), BW= Body weight, Same superscript along a column indicate no significant difference ($p \geq 0.05$)

The body weight: liver weight (21.0 – 23.0) and body weight: pancreas weight ratios (199 - 271) were not significantly different among groups. ALT levels and creatinin levels were significantly higher ($P \leq 0.05$) in all rats with diabetes compared to normal control rats and not significantly different ($P \geq 0.05$) among diabetic groups. When compared to normal control, AST levels were significantly ($P \leq 0.05$) high in SD, CM and DM groups, but not in HI. There were significantly high ($P \leq 0.05$) gamma GT levels in HI and CM groups but not in DM or SD when compared to normal control. However, GGT levels among the groups with diabetic rats were not significantly different. Liver enzymes and creatinin levels and body weight: liver weight and body weight: pancreas weight ratios are given in table 1.

All rats of the SD fed and normal control group had gained weight while most of the rats in other diabetic groups lost weight (unpublished data). The weight loss may be mainly due to elevated catabolic actions in diabetes. However, within the 6monthperiod, a significant reduction ($P \leq 0.05$) in weight loss was seen in SD fed and the normal control group when compared to other diabetic groups. Compared to body weight, the weights of liver and pancreas were not significantly different among groups indicating no apparent liver or pancreatic damage. Alanine transaminase (ALT) is an enzyme prominent in hepatocytes and the incline of

ALT in all diabetic rats in the present study indicated that there was a liver cell damage which could be due to Streptozotocin(Zafar*et al*, 2009) and accumulation of thiobarbituric acid-reactive substances (TBARS) with the progression of diabetes (Rakesh*et al*, 1998).

A study carried out with Streptozotocin (STZ) induced (intra peritoneal) diabetic rats has elicited a decline of elevated ALT levels after 7 weeks from the diabetes induction, when 2-Hydroxy 4-methoxy benzoic acid (500µg/kg body weight), a compound isolated from HI root was administered (Gayathri and Kannabiran,2009). This was hypothesized to be due to the action of this compound which reversed STZ-induced hepato-cellular necrotic changes. However such an effect was not observed in the present study as the leaves may not contain the hepato protective active compound insufficient amount.

A spartate transaminase (AST) is an enzyme prominent in skeletal muscles, heart muscles, liver and red blood cells which could be significantly elevated mainly with haemolysis and muscle catabolism (Begum*et al*, 2000). Oxidative stress gradually increases with time in diabetic patients. Hence due to the tissue damage owing to oxidative stress and diabetes induced muscle catabolism, AST levels in rats with diabetes should be higher. However this significant elevation was not observed in HI group. Elevated (Gamma glutamyltransferase) GGT is observed only with the damage to liver and billiary tract epithelium as most of the other organs expel GGT in to their lumen (in pancreas) or in to urine (in kidneys) or to their secretions (in epididymis, mammary glands) when there is tissue damage. GGT levels in SD and DM groups were not significantly different ($p \geq 0.05$) from the normal control group and other diabetic groups. However, other diabetic groups (CM & HI) had significantly higher ($p \leq 0.05$) GGT compared to normal control group. These results could be due to some positive effect of SD though not significant on liver and billiary tract epithelium.

As diabetes increase the muscle catabolism and renal failure, an increase in creatinin levels were observed in all diabetic rats compared to the normal control rats. These levels were not significantly different among the diabetic groups.

From the present study it can be concluded that long term consumption of the leafy porridges made with SD or HI does not cause excessive damage to the liver or kidney comparative to untreated diabetics. However leaf doses given with porridges does not indicate any hepato-protective effect as seen with comparison with normal rats.

References

Begum T, Oliver MR, Kornberg AJ, (2000). Elevated aminotransferase as a presenting finding in a patient with occult muscle disease. J Paediatr Child Health;36:189–90

Das H, Chakraborty U. (2011) Anti-hyperglycemic effect of *Scopariadulcis*instreptozotocin induced diabetes. Research Journal of Pharmaceutical, Biological and Chemical Sciences.; 2(2): 334-342

Ediriweera ERHSS, Ratnasooriya WD. (2009). A review on herbs used in treatment of diabetes mellitus in Sri Lankan aurvedic and traditional physicians. J of Aur. 30(4): 373-391.

Gayathri M, Kannabiran K.(2009).Antidiabetic activity of 2-hydroxy 4-methoxy benzoic acid isolated from the roots of *Hemidesmusindicus*onstreptozotocin-induced diabetic rats. Int J Diabetes &Metabolism;17:53- 57.

Hoskote SS, Joshi SR. (2008). Are Indians destined to be diabetic? J Asso. Phys. India; 56: 225-226.

Latha M, Pari L, Sitasawad S, Bhonde R. (2004).*Scopariadulcis*, a traditional antidiabetic plant, protects against streptozotocin induced oxidative stress and apoptosis in vitro and in vivo.Journal of Biochemical and Molecular Toxicology;18(5):261-272.

Rakesh K, Subrahmanyam VM, Jasim R, Kailash P, Jawahar K. (1998). Increased oxidative stress in rat liver and pancreas during progression of streptozotocin-induced diabetes.Clinical Science; 94:623-632.

Reagan-Shaw S, Nihal M, Ahmad N. (2008). Dose translation from animal to human studies revisited. The FASEB Journal.;22:659-661.

Senadheera SPAS, Ekanayake S. (2011).Glycaemic responses and Glycaemic Indices (GI) of rice and some green leafy porridges, Annual Scientific Sessions of the Nutrition Society of Sri Lanka:14.

Wijesuriya M. (2010). Diabetes Association of Sri Lanka, http://ceycollphysicians.org/images/diabetes%20in%20the%20young%20col%20of%20physicians%20april%202011.pdf. Accessed on 14 October 2010.

Zafar, M, Naeem-Ul-Hassan Naqvi, S, Ahmed, M, Kaimkhani, Z A. (2009).Altered liver morphology and enzymes instreptozotocin induced diabetic rats. *Int. J. Morphol., 27(3)*:719-725.