

# Evaluation of Antidiabetic activity of the marine gastropod, *Turbinella pyrum* from Gulf of Mannar, India

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## Abstract

Diabetes mellitus is a chronic metabolic disorder and is becoming the world's third "killer" together with cancer, cerebro and cardiovascular diseases due to its wide spread prevalence, morbidity and mortality. Modern anti-diabetic agents produce undesirable adverse effects. As a result, alternative therapy is required. A shift towards marine natural products is desperately needed. Hence, the present study has been carried out to examine the antidiabetic activity of methanolic extract of *Turbinella pyrum*. Diabetes was induced in the experimental albino rats by Alloxan. A comparison was made between the action of methanolic extracts of *Turbinella pyrum* and a known antidiabetic drug glibenclamide (600 µg/kg body wt). An oral glucose tolerance test (OGTT) was also performed in experimental diabetic rats. The diabetic rats were randomly divided into four groups (n=6/group). Group I served as control and received distilled water. Group II served as standard (Glibenclamide 600µg/kg). Group III and IV received the methanolic extract of *Turbinella pyrum* at a dose of 200mg/kg and 400mg/kg respectively. Maximum effect was observed at 3 hours with the low (200 mg/kg) and high (400 mg/kg) doses of methanolic extract of *Turbinella pyrum* which exerted more pronounced antidiabetic effects (87.33mg/dl and 57.17mg/dl). The effects of methanolic extract of *Turbinella pyrum* were more than that of the standard drug glibenclamide. Treatment of diabetic rats with the methanolic extract of *Turbinella pyrum* improved the body weight that is 15.49g and 16.75g at 200 and 400mg/kg doses respectively. The glibenclamide treated diabetic rats also showed improved body weight (13.88). These results demonstrate significant antidiabetic potential of the methanolic extract of *Turbinella pyrum* (p<0.01) and can be used for drug production in pharmaceutical industries.

**Keywords:** Alloxan, Antidiabetic, Wistar albino rats, *Turbinella pyrum*, Pharmaceutical.

## INTRODUCTION

Diabetes mellitus is one of the common metabolic disorders of inadequate control of blood glucose levels. 2.8% of the population suffers from this disease throughout the world and it may cross 5.4% by the year 2025. Diabetes is caused by the improper carbohydrate metabolism which is associated with low blood insulin levels or insensitivity of target organs to insulin (Maiti et al.,2004) and results in inherited and/or acquired deficiency in the production of insulin by the pancreas or by the ineffectiveness of the insulin produced. It is caused by either from inadequate secretion of hormone insulin, an inadequate response of target cells to insulin or a combination of these factors. This disease necessitates diagnostic testing, treatment and lifestyle modification. It is associated with reduced quality of life and increased risk factor for mortality and morbidity (Rawat Mukesh and Namita, 2013). If not cured or controlled, diabetes may even lead to acute or chronic complications causing ketoacidosis, micro-angiopathy and other related infections. Therefore once diagnosed, it is well regulated by means of various therapeutically effective drugs. Eventhough many drugs targeting carbohydrate hydrolysing enzymes, insulin release from pancreatic β cells, glucose utilization, insulin sensitizers are in clinical trials, the diabetes cannot be cured. Several of these drugs are linked to liver toxicity along with a number of deaths from hepatic failure as well as increasing the symptoms and risk factors of the heart disease which can lead to heart failure (Aparna Lakshmi et al., 2011). As a result, the long term of risk and effect on diabetes complications associated with these drugs are unknown. Besides, the therapy based on chemotherapeutic agents, the present century has progressed towards naturopathy. Thus, marine natural products have an ever emerging role to play in the treatment or management of lifelong prolonging diseases like diabetes mellitus, especially in developing countries where resources are meager (Chauhan et al.,2010). Thus, the present study was undertaken to evaluate the antidiabetic effect of *Turbinella pyrum* in alloxan induced diabetic rats.

## MATERIALS AND METHODS

The specimens of *Turbinella pyrum* were collected from the Gulf of Mannar coastal region of Thoothukudi during low tides from the sea. In the present study, whole-body tissue extract of *T.pyrum* was used for the antidiabetic assay. The freshly collected samples were cleaned and washed with fresh seawater to remove all impurities. The shells were broken, and the tissues were taken, then dried in a hot air oven at 60°C for 48 hours. One hundred grams of powder was exhaustively extracted with methanol in a Soxhlet apparatus and concentrated in a rotary vacuum evaporator when 15g of brown sticky mass was obtained.

### Induction of experimental diabetes

Male Swiss albino mice were fasted overnight (12–14 hours) and their weight and fasting blood glucose levels were recorded with a glucometer and then diabetes was induced by a single intraperitoneal injection (a volume of 1 ml/kg) of freshly prepared alloxan monohydrate solution (20 mg/ kg body weight). Alloxan was prepared by weighing according to individual animal weight and solubilized with 0.5 mL sodium citrate at pH 4.5 before injection. Food and water were presented to the animals 30 min. after administration of alloxan. After 48 h of alloxan injection, plasma blood glucose level of each animal was determined by taking the blood from the tail and animals with a fasting blood glucose level above 200 mg/dl were included in the study.

### TREATMENT PROTOCOL

The animals were divided into four groups for the evaluation of fasting blood glucose level with six animals in each group. They were treated with the methanolic extract of *T.pyrum* two days after alloxan injection excluding the diabetic control groups. Blood samples were collected from the tail vein after overnight fast at the intervals of 0, 1, 3, 5 and 7 hrs. Changes in body weight were also recorded. Groups 1 served as diabetic control (received only distilled water). Group 2 received standard drug (glibenclamide, 10 mg/kg per day orally). Groups 3 and 4 received the methanolic extract at a dose of 200 & 400 mg/kg, respectively daily in one mL aqueous solution using oral gavage for two weeks.

### Statistical analysis

The results, expressed as the mean  $\pm$  SE, were evaluated using the t test. Values of  $P < 0.01$  were considered statistically significant.

## RESULTS

In the present study, there were observable changes in body weight of the treated and untreated diabetic rats. Treatment of diabetic rats with the methanolic extract of *Turbinella pyrum* improved the body weight i.e 15.49g and 16.75g at 200 and 400mg/kg doses respectively. The glibenclamide treated diabetic rats also showed improved body weight (13.88g) (Table 1).

A dose dependent reduction in blood glucose levels was observed in alloxan – induced diabetic rats treated with methanolic extract of *Turbinella pyrum*. After a single dose of the extract given alloxan- induced diabetic rats, there was a significant ( $P < 0.01$ ) reduction in blood glucose levels of the diabetic rats within the period of acute study compared to the control (Table 2). The maximum effect was observed at 3 hours with the low (200 mg/kg) and high (400 mg/kg) doses of methanolic extract of *Turbinella pyrum* which exerted more pronounced effects ( $87.33 \pm 2.18$  mg/dl and  $57.17 \pm 1.87$  mg/dl). The effects of methanolic extract of *Turbinella pyrum* on reduction in blood glucose levels were more than that of the standard drug glibenclamide (Table 2)

During prolonged study (14days), the methanolic extract of *Turbinella pyrum* produced a sustained significant ( $P < 0.01$ ) reduction in blood glucose levels of the diabetic rats compared to control. The 200mg/kg and 400mg/kg of methanolic extract of *Turbinella pyrum* indicated significant reduction of  $39.02 \pm 2.02$  and  $37.87 \pm 2.28$  in the blood glucose levels respectively compared to the diabetic control ( $226.48 \pm 2.23$ ). However, glibenclamide level was  $55.43 \pm 2.68$  which was slightly high when compared to methanolic extract of *Turbinella pyrum* (Table 3).

**Table – 1 Effect of methanolic extract of *Turbinella pyrum* on body weight of alloxan – induced diabetic rats**

Treatments	Dose (mg/kg)	Day 0 (gm)	Day 15 (gm)	% Increase/ Decrease in body weight (gm)
Control	10ml	156.44±1.88	132.48±1.62	14.60
Methanolic extract of <i>Turbinella pyrum</i> 200mg/kg	200	142.87±1.83	162.60±2.86	15.49
Methanolic extract of <i>Turbinella pyrum</i> 400mg/kg	400	140.11±2.68	158.38±1.18	16.75
Glibenclamide	10ml	143.10±1.88	156.22±3.28	13.88

**Table – 2 Antidiabetic effect of methanolic extract of *Turbinella pyrum* on blood glucose level of alloxan – induced diabetic rats (Mean ± SEM)**

Treatments	Dose mg/kg	Blood glucose level (mg/dl) in hours				
		0hr	1hr	3hrs	5hrs	7hrs
Control	10	230.20±1.17	250.11±3.12	253.1±1.62	256.00±1.66	249.17±2.22
Methanolic extract of <i>Turbinella pyrum</i> 200mg/kg	200	225.01±1.07	159.03±2.17	87.33±2.18*	95.00±2.11*	101.13±2.66
Methanolic extract of <i>Turbinella pyrum</i> 400mg/kg	400	211.14±1.04	132.01±2.16	57.17±1.87*	88.18±2.11*	99.17±2.26*
Glibenclamide	10	235.40±1.16	239.11±2.87	225.60±2.06	207.07±1.87	196.38±2.47

Values expressed as the mean ± SE, n = 6 animals in each group; \*highly significant (P < 0.01) compared to control and standard groups

**Table – 3 Effect of methanolic extract of *Turbinella pyrum* on blood glucose level of alloxan – induced diabetic rats during prolonged treatment (Mean ± SEM)**

Treatments	Dose mg/kg	Blood glucose level (mg/dl) in days						
		Day 0	Day 1	Day 3	Day 5	Day 6	Day 7	Day 15
Control	10	230.21±1.72	235.20±3.41	242.31±2.87	248.38±1.16	235.41±2.17	231.87±1.18	226.48±2.23
Methanolic extract of <i>Turbinella pyrum</i> 200mg/kg	200	226.32±1.88	87.45±2.01	210.28±1.13	150.12±2.18	62.08±1.22*	73.07±3.1*	39.02±2.02*
Methanolic extract of <i>Turbinella pyrum</i> 400mg/kg	400	221.27±2.86	79.33±1.93	201.22±1.08	146.38±1.11	58.35±2.21*	68.08±1.09*	37.87±2.28*
Glibenclamide	10	231.31±1.61	206.20±2.25	172.30±1.86	132.11±1.30	85.68±1.38*	76.42±2.88*	55.43±2.68*

Values expressed as the mean ± SE, n = 6 animals in each group; \*highly significant (P < 0.01) compared to control and standard groups

## DISCUSSION

Diabetes mellitus is a syndrome that is distinguished by a loss of glucose homeostasis due to defects in insulin secretion and insulin action, both of which are caused by defects in the metabolism of glucose and other energy-producing fuels such as lipids and proteins (Grijesh Kumar Mall et al., 2009). Diabetes is associated with a number of complications like neuropathy, retinopathy and peripheral vascular insufficiencies. Glucose homeostasis is the best approach for lowering blood glucose levels. The treatment of type II diabetes is exacerbated by numerous risk factors associated with the disease. Increased oxidative stress has been suggested as mechanism underlying diabetes and its related complications. Researchers pay increasing interest towards the potential of marine organisms as an alternate source for isolating novel metabolites. Natural products such as secondary metabolites, plant, animals and microbes are an excellent source for bioactive molecules that have been finally evolved into medication (Faten K. Abd El- Hady et al., 2014)

In the light of the results, the present study indicates that the methanolic extract of *Turbinella pyrum* has significant antidiabetic activities in alloxan induced hyperglycemic rats with little improvement in body weight. The result of the present study agrees well with the findings of Syed Mansoor ahmed et al., (2005) who studied the antidiabetic activity of *Terminalia catappa* and found out significant anti-hyperglycemic activities in alloxan induced hyperglycemic rats. In vitro antidiabetic activity of stem bark of *Bauhinia purpurea* showed a good anti-diabetic activity (Gupta Daksha et al., 2012). Chauhan et al., (2010)

In the present study, the groups treated with methanolic extract of *Turbinella pyrum*, Group II 200mg/kg and Group III 400mg/kg showed a significant reduction in blood glucose in a dose dependent manner from 60 minutes onwards. At the end of the experimental duration, the level of glucose in the group administered with highest dose of extract (400mg/kg) was less ( $37.87 \pm 2.28$ ) than that of the standard drug ( $55.43 \pm 2.68$ ) treated groups. Similar to the present findings Shanmugapriya et al., 2015 and Meenakshi et al., (2012) observed the decrease in the level blood glucose in the methanolic extract of *Phallusia nigra* and *M.exasperatus*. Kajal chakraborty and Manju joy (2016) observed the antidiabetic activities of ethyl acetate and methanolic extracts of cephalopods *Sepia pharonis*, *S.inermis*, *Cistopus indicus* and *Amphioctopus marginatus*. The present study showed that the methanol extracts of *T.pyrum* produced marked decrease in blood glucose in diabetic rats at 200mg/kg and 400mg/kg concentrations after 7 hours of treatment ( $101.13 \pm 2.66$  and  $99.17 \pm 2.26$ ). The antidiabetic effect of *T.pyrum* may be due to increased release of insulin from  $\beta$  cells of pancreas. 15.49 and 16.75 percentage of weight reduction was seen in the treated groups. The results of the present study agrees well with the findings of Meenakshi et al., 2013 from the ethanol extract *Microcosmus exasperatus*. According to Khan and Shechter (1991), a 25% reduction can be considered to have potential hypoglycemic effect. Maintenance of blood glucose level in the treated groups indicates the effectiveness of the extract.

It is suggested that the methanolic extract of *Turbinella pyrum* shows antidiabetic properties in animal model. Detailed studies on isolation, characterization and structural determination of the exact chemical compounds could lead to pharmacologically potent antidiabetic drug molecules.

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