



RESEARCH ARTICLE

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**EFFECTS OF BITTERGOURD ON GLUCOSE TOLERANCE,  
OXIDATIVE STRESS AND BLOOD PRESSURE IN FRUCTOSE  
INDUCED DIABETIC RATS**

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**Abstract:** Bitter melon (*Momordica charantia*) or bittergourd commonly known as karella, (family Cucurbitaceae), has been proved for hypoglycaemic effects. The objective of the present study was to evaluate effects of bittergourd (*momordica charantia*) on glucose tolerance, oxidative stress and blood pressure in fructose induced diabetic rat. Wister rats were made diabetic by fructose diet for 6 week. Animals were divided in to two groups. One as diabetic control and other one group was treated by 400mg/kg bitter gourd alcoholic extract for next 2 weeks. After 8 weeks treatment they were checked for glucose tolerance, oxidative stress and blood pressure. The significant improvement in bitter gourd alcoholic extract treated group compare to diabetic group was found. So, from present study it is concluded that bitter gourd fruit has beneficial effects on glucose tolerance, oxidative stress and blood pressure in fructose induced diabetic rats.

**Keywords:** Bittergourd; diabetes; fructose diet; glucose tolerance; blood pressure

## INTRODUCTION

Diabetes mellitus affects an estimated 285 million adults worldwide, of whom approximately 85% to 95% have type 2 diabetes (T2DM)<sup>1</sup>. The heart disease is a major cause of death in diabetic patients<sup>2,3</sup>, and coronary artery disease and atherosclerosis are mainly involved in the increased incidence of cardiovascular dysfunction<sup>4,5</sup>. Hypertension and diabetes are interrelated metabolic disorders<sup>6</sup> that strongly predispose an individual to atherosclerotic cardiovascular disease (CVD)<sup>7,8</sup>. The disease causes morbidity and long-term complications and an important risk factor for cardiovascular diseases<sup>9</sup>. Pharmacotherapy of diabetes without any side effects is still a challenge. So that need for complementary and alternative medicine with antidiabetic activity and less side effects.

The Bitter melon (*Momordica charantia*) or Bittergourd fruit (BF), commonly known as karela (L), family: Cucurbitaceae), is grown in tropical countries in South Asia, South America and Africa. The juice of bittergourd fruit has been proved for hypoglycaemic effects in experimental type 1 diabetes and in type 2 human diabetes<sup>10, 11</sup>. The juice of bittergourd can increase glucose uptake by

tissues *in vitro*<sup>12</sup>. Many species of the genus in the Cucurbitaceae family, like *Momordica Charantia*<sup>13, 14, 15</sup> and *Momordica Cymbalaria*<sup>16</sup> have been reported for significant antidiabetic effects. The active fractions from fruits of *M. charantia* such as saponins<sup>17, 18</sup> and peptides<sup>19, 20, 21</sup> had hypoglycemic effects. In view of various effects of bittergourd fruit juice, this study was designed to assess the effect of bitter gourd fruit juice on glucose tolerance, oxidative stress and blood pressure in fructose induced diabetic rats.

## MATERIALS AND METHODS

### Preparation of Bitter gourd alcoholic extract

Fresh green fruit of BF were purchased from local market and were identified and authenticated as *Momordica charantia* fruit by Dr. H.B. Singh, Head of Raw Materials Herbarium & Museum (RHMD), National institute of science communication and information resources (NISCAIR), New Delhi, India. Voucher specimen was deposited in the herbarium of the institute. Bitter gourd fruits were washed thoroughly, and this plant material was dried in shade. The dried fruit was ground to homogeneous powder. Bitter

gourd fruit powder was soaked in the 95% methanol solvent in glass jars for 2 days at room temperature and protected from light with stirring at regular intervals. Then the solvent was filtered through Whatman #1 filter paper and this was repeated three to four times until the extract gave no coloration. The extract was filtered and dried at 55°C using a rotovapor vacuum drier. The dried sample was crushed into powder and stored at -80°C until use. These extract was used for further studies. The yield of the extract was 6.3 % (w/w in terms of dried starting material).

### **Experimental animals**

The experimental protocol (No: IAEC/SKCOP/11-12/05) was approved by Institutional Animal Ethics Committee and animals were maintained under standard conditions in an animal house approved by Committee for the Purpose of Control and Supervision on Experiments on Animals (CPCSEA).

### **Induction of diabetes mellitus**

Wister rats, initially weighing 200–230 g, were obtained from the zydus research center (Ahmedabad, Gujarat, India) and rats were housed in air conditioned room at 22±3 °C with humidity (55 ± 5%) and 12h/12h light-dark cycle. and tap water *ad libitum*.

After a 1-week acclimation period, rats were divided randomly into two groups. The normal control (NC) group was fed control diet, whereas the experimental group was fed a 60% fructose diet for 8 weeks. The fructose-enriched diet was composed of 21% protein, 60% carbohydrate (as fructose), and 5% fat (of total energy, % kcal), sodium 0.49%, and potassium 0.49%. This composition was comparable to the control diet.

### **Treatment protocol**

After the first 6 weeks, the fructose-treated rats were further subdivided into two groups.

Group-1: Normal control (NC); Group-2: Diabetic control (DC); Group3: Diabetic treated with 400 mg/kg alcoholic extract of bitter gourd fruit (DAE)

During the last 2 weeks, the NC and DC rats were treated with vehicle only. The other group was received bitter gourd alcoholic extract for 2 weeks while rats were still on fructose diet. Body weight was measured weekly throughout the study. During the study daily food intake and water intake were recorded in all the groups. These dietary periods lasted for 8 weeks and rats were maintained in accordance with the Animal Experiment Committee guidelines.

At the end of 8 weeks, for glucose tolerance test all groups received glucose solution (1.5

g/kg, orally) 30 minutes after BFJ treatments. Blood glucose levels were determined at 0, 30, 90 and 120 min. after glucose administration. The blood pressure was recorded by invasive method (carotid artery cannulation). Then rats were sacrificed. The heart was collected from each rat.

Antioxidant parameters were measured from left ventricle heart tissue. The reported methods were used to measure Malondialdehyde (MDA) Ohkawa et al<sup>22</sup>, GSH level (Reduced Glutathione) Beutler et al<sup>23</sup>, Superoxide dismutase (SOD) Misra et al<sup>24</sup>, Catalase Aebi et al<sup>25</sup>, and tissue protein levels Lowry et al<sup>26</sup>.

### Statistical analysis

Results are presented as Mean + SEM. Statistical differences between the means of the various groups were evaluated using one-way analysis of variance (ANOVA) followed by Tuckey's test. Data were considered statistically significant at P value < 0.05.

## RESULTS AND DISCUSSION

### Results

#### General features of experimental rats

All group rats started with similar mean body weights (210±7.6 g). At week 8, the body

weight is increased in the DC group as compared with the NC group ( $P < 0.05$ ). After treatment with bitter gourd alcoholic extract the bodyweight was decreased as compared with DC group ( $P < 0.05$ ) (Figure 1). There were no significant differences in consumption of food and water among the all groups at week 6 and week 8 after treatment.

### OGTT

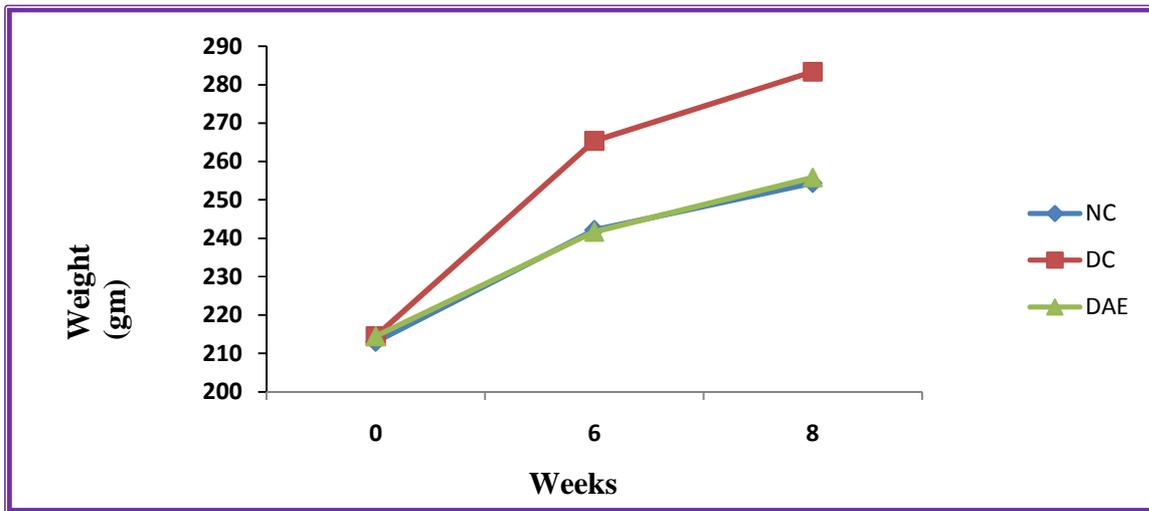
Glucose loaded rats showed significantly elevated glucose levels. Pretreatment with bitter gourd alcoholic extract reduced blood glucose significantly (Figure 2).

### Blood Pressure

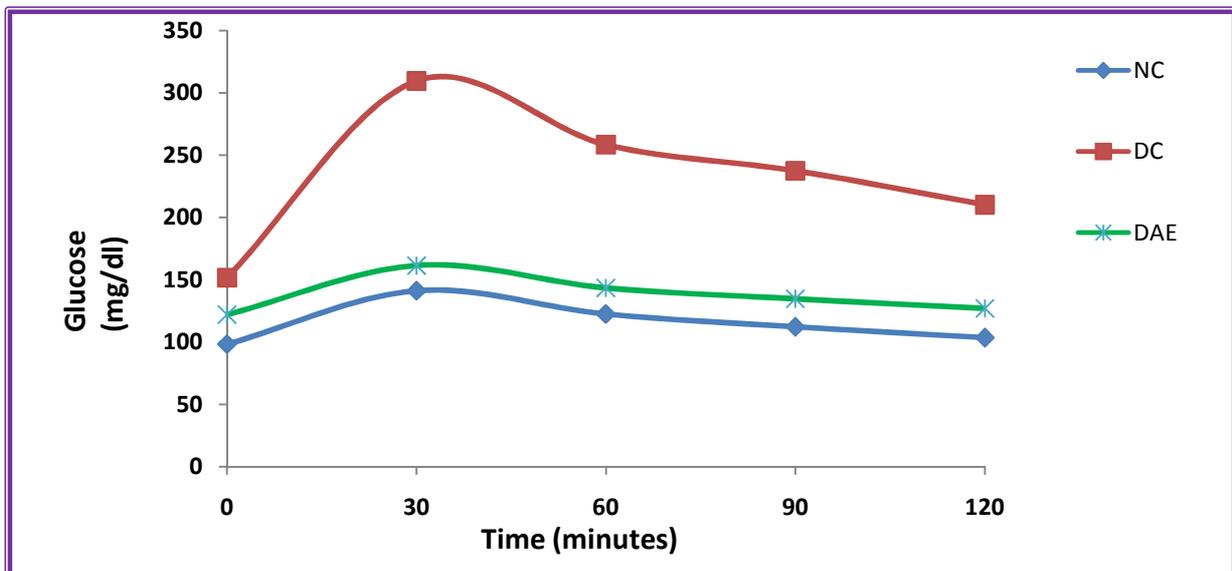
The mean blood pressure was significantly ( $P < 0.05$ ) increased after 8 weeks study in DC rats as compared to NC rats. Treatment with bitter gourd alcoholic extract showed significant ( $P < 0.05$ ) inhibition in rising of blood pressure in fructose induced diabetic animals (Figure 3).

### Effects on Antioxidant parameters

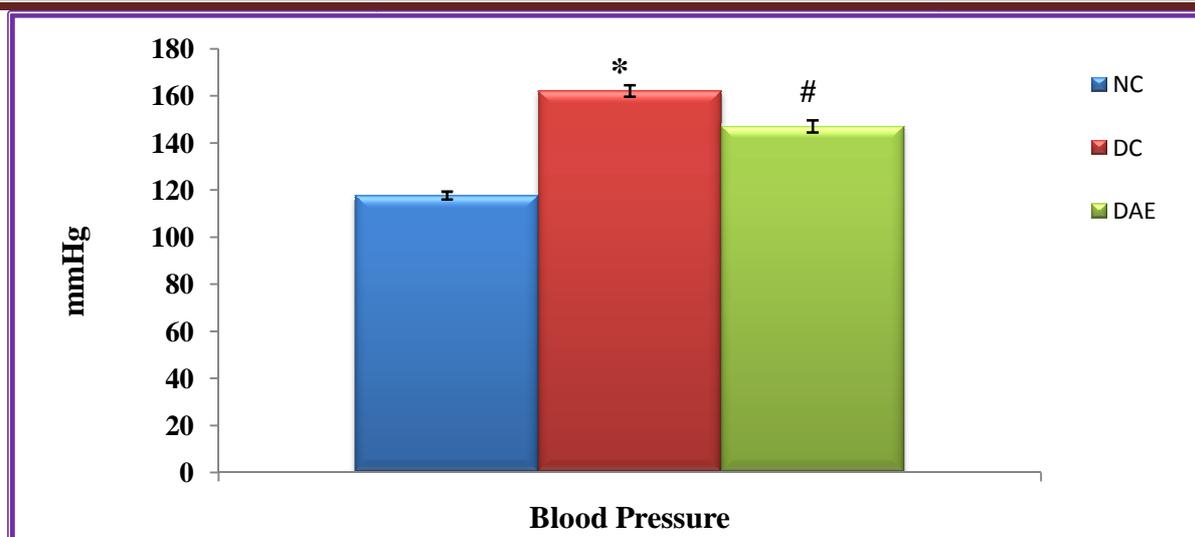
At 8 week diabetic animals showed oxidative stress due to lower levels of SOD, GSH and Catalase and higher level of MDA compare NC rats. Treatments with bitter gourd alcoholic extract in diabetic rats showed significant higher levels of SOD, GSH and Catalase and lower level of MDA (Table 1).



**Figure 1:** Effect on body weight of the diabetic rat by oral treatment of bittergourd alcoholic extract for 8 weeks: Bittergourd alcoholic extract significantly prevented in elevation of body weight of diabetic rats.



**Figure 2:** Effect on glucose tolerance of the diabetic rat by oral treatment of bitter gourd alcoholic extract for 8 weeks: Bitter gourd alcoholic extract produced significant improvement in glucose tolerance.



**Figure 3:** Effect on blood pressure of the diabetic rat by oral treatment of bitter gourd alcoholic extract for 8 weeks: Bitter gourd alcoholic extract prevented in rising of blood pressure of diabetic rats.

\* $P < 0.05$  compared with the normal control group that reveals significant difference between two groups;

#  $P < 0.05$  compared with the diabetic group that reveals significant difference between two groups.

## Discussion

It is reported that chronic feeding of fructose in experimental animals produces glucose intolerance associated with hyperglycemia and insulin resistance<sup>27, 28</sup>. In present study, it was found that fructose diet produced weight gain and no significant effects on food and water intake in diabetic rats compare to normal rats. Treatment with bittergourd alcoholic extract significantly prevented the elevation of weight in diabetic rats.

In the present study diabetic animals were also found to have impaired glucose tolerance with

high glucose level after glucose load compared to control animals. The results of this study have demonstrated that oral administration of bittergourd alcoholic extract daily over duration of 2 weeks diabetic rats can significantly improve the impaired glucose tolerance. This result is consistent with those reported earlier<sup>29</sup>. That may be through the regulation of PPARs-mediated pathway<sup>30</sup>. Momordica charantia fruit extracts act as insulin sensitizer and activate the glucose transport possibly by upregulation of Glut- 4, PPAR $\gamma$  and PI3K<sup>31</sup>. Therefore, we assumed

that bitter melon used in our study behaved similar to several PPARs ligands. Isolation, purification and standardization of these active constituents from plant extracts leads novel molecule, is worth pursuing and the same is in future.

Oxidative stress is associated with complications of diabetes<sup>32, 33</sup>, also links to insulin resistance *in vitro* and *in vivo*<sup>34</sup>. When glucose and free fatty acid (FFA) increase, they cause oxidative stress along with activation of stress sensitive signaling pathways<sup>34</sup>. In our study there was high of serum MDA level and low SOD, catalase, glutathione level in diabetic group compare to normal animals. The treatment of bittergourd alcoholic extract reduces elevated level of MDA and increase decreased level of SOD, catalase, glutathione. So they decrease the oxidative stress.

Fructose fed rats show a moderate hypertension and glucose intolerance, with high levels of plasma insulin, cholesterol and triglycerides cause cardiovascular dysfunction associated with obesity and metabolic disorders<sup>35, 36</sup>. In terms of the mechanisms behind the fructose-induced cardiovascular

changes, there is evidence for a role of the sympathetic nervous and renin angiotensin systems (RAS)<sup>36, 37</sup>. In our study, blood pressure of diabetic animals was found to be higher as compared to control animals. Treatment of diabetic rats with bitter gourd alcoholic extract over a period of 2 weeks normalized the elevated blood pressure. This result suggests that BFJ possesses anti-hypertensive properties.

The results of bitter gourd alcoholic extract highlight the topic for research and discussion for antihypertensive effects of bitter gourd.

### CONCLUSION

Our data suggest that bitter gourd prevents the fructose induced metabolic abnormalities as evident from the improvement of glucose intolerance, oxidative stress and high blood pressure.

### ACKNOWLEDGEMENT

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**Table 1**

Effect of bitter gourd alcoholic extract treatment on oxidative stress of diabetic rats.

Parameters	NC	DC	DAE
SOD (units/min/mg protein)	2.85± 0.25	0.67± 0.13*	2.14±0.21 <sup>#</sup>
Catalase (units/min/mg protein)	6.25± 0.17	2.18± 0.17*	5.12±0.57 <sup>#</sup>
MDA (nmoles/mg protein)	3.45± 0.33	9.14±0.41*	4.12±0.27 <sup>#</sup>
GSH (µgm/mg protein)	9.66± 1.02	1.77± 0.35 *	6.42±0.42 <sup>#</sup>

Values are expressed as Mean ± S.E.M

\*- significantly different from NC (p &lt; 0.05)

#- significantly different from DC (p &lt; 0.05)

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