Therapeutic Potential of Cinnamomum Zeylanicum Extract to Mitigate Hyperglycemia

Muhammad Ahmar Jahangir, Aamir Shehzad, Masood Sadiq Butt, Muhammad Shahid

Abstract

Background: Nutrition and health have become one of the most intriguing focuses in the world today. Technological advancement, nutritional imbalances and sedentary lifestyle have ascended numerous health issues worldwide. Scientific evidences have provided the chemopreventive and chemotherapeutic role of dietary phytochemicals to cure these ailments. Cinnamon (Cinnamomum zeylanicum) has been utilized as a potential therapeutic agent in various cultures for centuries.

Objective: Trans-cinnamaldehyde (3-phenyl-2-propanal) contributes as a major constituent of cinnamon bark oil approximately about 49.9-62.8% of the total amount and has hypoglycemic, hypocholesterolemic and anticancer potential.

Methods: The ethanolic and supercritical fluid extracts of cinnamon bark were subjected to in vivo modelling to evaluate the hypoglycemic potential of cinnamaldehyde. Purposely, efficacy trial was performed on normal and hyperglycemic Sprague dawley rats for 8 weeks. Three types of diets i.e. normal (D₀), nutraceutical containing 0.5% conventional extract (D₁) and nutraceutical containing 0.1% supercritical fluid extract (D₂) were used throughout the study.

Results: Feed & drink intake and body weight were increased during the trial. Serum analyses exhibited the maximum reduction 11.65% in glucose level in hyperglycemic rats as an effect of diet D₂ followed by D₁ which depicts 9.94% reduction. Nutraceutical diet D₂ also increased the insulin level up to 7.23% in hyperglycemic rats whereas this increment was 2.37% in normal rats.

Conclusion: The current research helps us to conclude that cinnamon extract is effective against hyperglycemia.

Keywords: Cinnamaldehyde, Hyperglycemia, Nutraceutical, Antioxidant.

Introduction

Across the globe, nutritional imbalances in the diet are causing a number of physiological dysfunctions which engrossed the adoption of diet based therapies as an intervention against various infirmities. Amongst principal therapeutic tool of apposite dietary guidelines, the use of functional and nutraceutical food provides an opportunity to alleviate these health problems within the population. Overwhelming evidences from epidemiological and biological studies have illuminated the extensive use of plant based products owing to their rich phytochemistry against numerous ailments. Hence, dietary approbations for the prevention and cure of chronic diseases have emphasized the con-
sumption of variety of plant foods.\textsuperscript{3} Spices having a virtuous recognition in cultural heritage and food appreciation possess pivotal health linkages and have also been used effectually in ethnic system of medicine. Being common dietary adjunct, spices contribute as a source of numerous bioactive compounds which influence various digestion and metabolic processes. Spices rich in polyphenols, inhibit oxidation processes in food products and exhibit health promoting effects by increasing antioxidative potential of the body on consumption.\textsuperscript{3} Amongst spices, cinnamon (\textit{Cinnamomum zeylanicum}) belonging to family \textit{Lauraceae} has been utilized as a potential therapeutic agent in various cultures for centuries. Cinnamon bark is one of the oldest known spices used against gastrointestinal complaints, chronic bronchitis and inflammation of eyes in ayurvedic medicine for over 6000 years.\textsuperscript{4} The major constituents of cinnamon bark oil includes 75% cinnamaldehyde, 5% cinnamyl acetate, 3.3% caryophyllene, 2.4% linalool and 2.2% eugenol.\textsuperscript{5}

Diabetes mellitus is a chronic metabolic disorder that affects about 4\% of the world’s population and is expected to upsurge by 5.4\% in 2025.\textsuperscript{6} According to W.H.O. prediction, diabetes will become the 7\textsuperscript{th} leading cause of mortality worldwide by the end of 2030.\textsuperscript{7} They projected the diabetes prevalence with total number of people affected will increase from 171 to 366 million between 2000 and 2030.\textsuperscript{8} It is also associated with abnormalities in body’s metabolism which ultimately leads to numerous other complications like an increased risk of cardiovascular disease, neuropathy, retinopathy and nephropathy in addition to damaging liver, kidney and beta-cells of pancreas. Improvements in glycemic control have proved to be beneficial in reducing the risk of these complications. Consumption of cinnamon has been found effective in improving the glycaemic control and also reduces the onset and progression of diabetes.\textsuperscript{9}

Cinnamaldehyde (3-phenyl-2-propanal) represents the main constituent of the cinnamon bark oil contributing about 49.9 to 62.8\% of the total amount.\textsuperscript{10} It provides protection against metabolic syndromes, cardiovascular complications and diabetes. Functionality of insulin receptors is improved by virtue of enzyme activation (insulin receptor kinase) which is responsible for insulin binding to the cells. Likewise, it is also responsible for hindrance in the enzyme (insulin receptor phosphatase) activity that impede this process, ultimately leading to the maximum phosphorylation of insulin receptor, which is associated with improved insulin sensitivity. Additionally, glucose tolerance is also increased by reducing effect of cinnamaldehyde on activity of hexokinase and glycogen content in the liver and skeletal muscles. Cinnamon polyphenols contribute to the regulation of various proteins like glucose transporter 4 (GLUT4), insulin receptor β and tristetraprolin involved in insulin signal transduction pathway.\textsuperscript{11} Recent research illustrates that cinnamaldehyde contributes positively towards consumers health by reducing glucose, cholesterol and LDL levels.

**Materials and Methods**

The dried cinnamon (\textit{Cinnamomum zeylanicum}) bark was procured from local market and ground to obtain particle sizes in the range of 300-500 µm for further analyses. The reagents (analytical and HPLC grade) and standards were purchased from Merck (Merck KGaA, Darmstadt, Germany) and Sigma-Aldrich (Sigma-Aldrich Tokyo, Japan). For efficacy trial, male Sprague Dawley rats were acquired from National Institute of Health (NIH) Islamabad. housed in the Animal Room of NIFSAT. For biological assay, diagnostic kits were purchased from Sigma-Aldrich, Bioassay (Bioassays Chemical Co. Germany) and Cayman Chemicals (Cayman Europe, Estonia).

The cinnamon extracts were prepared using ethanol and water (50\% v/v) for a period of 60 min with constant temperature of 50\textdegree C following the outlines of Mariod et al.\textsuperscript{12} Resultant solvent extract was filtered and recovered through Rotary Evaporator (Eyela, Japan). Cinnamon powder was subjected to supercritical fluid extraction system (SFT-150 Supercritical Fluid Technologies, Inc.) to obtain supercritical fluid extracts using 99.8\% pure CO$_2$. Accordingly, sample was placed in 100 mL extraction vessel followed by Optimization of CO$_2$ at 5000 psi while maintaining time and temperature conditions constant.\textsuperscript{13}

The bio evaluation trial was conducted to investigate the therapeutic potential of cinnamaldehyde against hyperglycemia. Purposely, seventy male Sprague Dawley rats were procured from National Institute of Health (NIH) Islamabad and housed in the Animal Room of National Institute of Food Science and Technology, University of Agriculture Faisalabad. Intentionally, rats were acclimatized by feeding on basal diet for a period of one week under environmentally controlled conditions of temperature (23 ± 2\textdegree C) and relative humidity (55 ± 5\%) along with 12 hrs light-dark period. At the initiation of study, some rats were
sacrificed to get baseline trend of selected biochemical parameters. During efficacy trial of sixty days, two types of studies were conducted separately in order to determine the therapeutic effect of conventional solvent extract (CSE) @ 0.5% and supercritical solvent extract (SFE) @ 0.1% on selected parameters such as lipid profile, glucose and insulin level. Study I comprised of rats fed on normal diet, whereas high sucrose diet was administrated to induce hyperglycemia in study II (Table 1). The composition of diets given to rats is presented in Table 3. On the basis of administrated diet, each study was further subdivided into three groups containing 10 rats in every group. During 60 days trials, simultaneous provision of nutraceutical CSE (D_1), nutraceutical SFE (D_2) diets along with control (D_0) were given to respective groups (Table 2).

### Table 1: Different Studies Conducted during Efficacy Trials.

<table>
<thead>
<tr>
<th>Study I</th>
<th>Normal rats</th>
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<tbody>
<tr>
<td>Study II</td>
<td>Hyperglycemic rats</td>
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</table>

In each study, the collected sera were evaluated for glucose concentration by GOD-PAP method as described by Sailesh and Padmanabha whereas, insulin level was assessed following the procedure described by Ahn et al. Serum lipid profile of rats including total cholesterol, low density lipoprotein (LDL), high density lipoprotein (HDL) and triglycerides were measured according to their respective protocols. Triglycerides and cholesterol level in sera samples were determined by liquid triglyceride (GP-O-PAP) and CHOD-PAP method respectively, outlined by Vafa et al. Likewise, HDL of serum samples using cholesterol precipitant method and LDL were analyzed as per guidelines of Alshatwi et al.

The resultant data were examined through completely randomized design (CRD) by employing Cohort version 6.1 (Costat-2003). Moreover, level of significance was measured using analysis of variance (ANOVA) following the principles outlined by Steel et al.

### Results

Efficacy studies were performed in vivo on male Sprague Dawley rats in order to determine the functional and nutraceutical importance of cinnamon extract against hyperglycemia. For this purpose, rodent experimental modeling was used instead of human subjects because of appropriate supervision, easy handling, feasibility of controlled environmental conditions and

<table>
<thead>
<tr>
<th>Ingredients (%)</th>
<th>Normal Diet</th>
<th>High Sucrose Diet</th>
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<tbody>
<tr>
<td>Corn starch</td>
<td>82</td>
<td>42</td>
</tr>
<tr>
<td>Corn oil</td>
<td>10</td>
<td>10</td>
</tr>
<tr>
<td>Casein</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>Minerals</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>Vitamin mixture</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Sucrose</td>
<td>-</td>
<td>40</td>
</tr>
</tbody>
</table>

14 Gain in body weight of experimental rats was measured weekly throughout the study period to monitor any suppressing effect of cinnamaldehyde enriched diets.

It is assured that all the bioefficacy trials were performed in compliance with the institutional guidelines and relevant laws of the National Institute of Food Science and Technology, University of Agriculture Faisalabad, Pakistan. Additionally, all the experimental modeling embraces dietary and safety plans were reviewed and approved by the institutional committee(s).

Feed intake of individual group of rats was measured daily by subtracting the remaining diet from the total diet during the whole trial. Likewise, water intake of each group was also recorded on daily basis. Gain
safety concerns relating to active ingredient (cinnamaldehyde) being used.

Mean values for (study I) explicated feed intake 13.92 ± 0.46, 14.45 ± 0.63 and 14.87 ± 0.52 g/rat/day at initiation that raised to 18.65 ± 0.71, 19.12 ± 0.67 and 19.43 ± 0.56 g/rat/day for D₀, D₁ and D₂, respectively at the completion of study. The Figure 1 illustrated maximum feed intake at initiation in D₀ (16.78 ± 0.62 g/rat/day) followed by D₁ (16.21 ± 0.65 g/rat/day) and D₂ (15.49 ± 0.58 g/rat/day) whereas similar trend was observed at the termination of study II i.e. D₀ (24.59 ± 0.47 g/rat/day) trailed by D₁ (22.34 ± 0.45 g/rat/day) and D₂ (21.16 ± 0.44 g/rat/day). Means relating to drink intake revealed a gradual rise as a function of treatments and time. In this context, minimum intake was noticed in D₀ followed by D₁ and D₂ i.e. 18.51 ± 0.37 & 23.37 ± 0.64, 18.69 ± 0.32 & 24.18 ± 0.56 and 18.74 ± 0.41 & 24.63 ± 0.59 mL/rat/day, respectively at 1st and 8th week of the study I. Conversely, in study II (hyperglycemic rats), D₀ expounded the highest drink intake 22.49 ± 1.03 mL/rat/day followed by D₁ (22.37 ± 0.95 mL/rat/day) and D₂ (22.31 ± 0.98 mL/g/rat) at initiation of the trial. Whereas, increase in drink intake was recorded due to the effect of time as 31.62 ± 0.78, 30.13 ± 0.89 and 29.20 ± 0.83 mL/rat/day in D₀, D₁ and D₂, respectively at termination (Figure 2).

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**Fig. 1:** Feed Intake in Study II (g/rat/day).

**Fig. 2:** Drink Intake in Study II (mL/rat/day).
Table 4: Effect of Diets on Glucose (mg/dL) of Rats.

<table>
<thead>
<tr>
<th>Studies</th>
<th>Treatments</th>
<th>F value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>D₀</td>
<td>D₁</td>
</tr>
<tr>
<td>Study I</td>
<td>90.98 ± 5.07</td>
<td>89.14 ± 4.72</td>
</tr>
<tr>
<td>Study II</td>
<td>138.45 ± 6.69a</td>
<td>124.69 ± 5.45b</td>
</tr>
</tbody>
</table>

\( p \leq 0.05 \)

NS = Non-significant; ** = Highly significant; Study I: Normal rats, Study II: Hyperglycemic rats, D₀: Control diet, D₁: Diet containing nutraceutical\text{CSE}, D₂: Diet containing nutraceutical\text{SFE}

Fig. 3: Body Weight in Study II (g/rat/week).

Fig. 4: Percent Decrease in Glucose as Compared to Control.
Table 5: Effect of Diets on Insulin (µU/mL) of Rats.

<table>
<thead>
<tr>
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</thead>
<tbody>
<tr>
<td></td>
<td>D₀</td>
<td>D₁</td>
</tr>
<tr>
<td>Study I</td>
<td>7.84±0.51</td>
<td>7.99±0.48</td>
</tr>
<tr>
<td>Study II</td>
<td>13.47±0.63b</td>
<td>14.41±0.65a</td>
</tr>
</tbody>
</table>

(p ≤ 0.05)  
NS = Non-significant; * = Significant; Study I: Normal rats; Study II: Hyperglycemic rats;  
D₀: Control diet; D₁: Diet containing nutraceutical CSE; D₂: Diet containing nutraceutical SFE

The recorded body weights of groups D₀, D₁ and D₂ (Study I; Normal rats) were 131 ± 4.83, 135 ± 5.62 and 137 ± 5.14 g/rat, respectively that raised to 235.67 ± 4.19, 244.81 ± 3.73 and 253 ± 4.68 g/rat till the end of trial. The study II (Hyperglycemic rats) depicted initial body weight of D₀, D₁ and D₂ groups as 145 ± 5.76, 142.15 ± 5.08 and 139 ± 4.25 g/rat, correspondingly (Figure 3). However, at the end of trial resultant body weights were 207 ± 6.63, 225.89 ± 7.14 and 238.12 ± 8.49 g/rat for D₀, D₁ and D₂, respectively. The recorded body weights of groups D₀, D₁ and D₂ (Study I; Normal rats) were 131 ± 4.83, 135 ± 5.62 and 137 ± 5.14 g/rat, respectively that raised to 235.67 ± 4.19, 244.81 ± 3.73 and 253 ± 4.68 g/rat till the end of trial. The study II (Hyperglycemic rats) depicted initial body weight of D₀, D₁ and D₂ groups as 145 ± 5.76, 142.15 ± 5.08 and 139 ± 4.25 g/rat, correspondingly (Figure 3). However, at the end of trial resultant body weights were 207 ± 6.63, 225.89 ± 7.14 and 238.12 ± 8.49 g/rat for D₀, D₁ and D₂, respectively.

Means for glucose in study I (normal rats) showed values 90.98 ± 5.07, 89.14 ± 4.72 and 87.75 ± 4.13 mg/dL for D₀, D₁ and D₂ groups, respectively. Mean glucose concentration for D₀ group in study II was 138.45 ± 6.69 mg/dL that declined substantially to 124.69 ± 5.45 mg/dL in D₁. Whereas the lowest glucose level was noticed for D₂ as 122.32 ± 5.18 mg/dL in hyperglycemic rats (Table 4). It is obvious from results that diets containing nutraceutical CSE (D₁) performed better against glucose related deformities than diets containing nutraceutical SFE (D₂) and control diet (D₀). The Figure 4 depicted highest percent decline in glucose for study II i.e. 9.94 and 11.65% for D₁ and D₂, respectively. However, study I showed a non-significant decline in glucose level in groups; D₁ (2.02%) and D₂ (3.55%) as compared to control.

Means related to study I (Table 5) elucidated that minimum insulin level was 7.84 ± 0.51 µU/mL (D₀) that increased to 7.99 ± 0.48 µU/mL (D₁) and 8.03 ±
0.60 µU/mL (D₂). The study II indicated a momentous rise in insulin level; maximum level was recorded for D₂ (14.52 ± 0.64 µU/mL) followed by D₁ (14.41 ± 0.65 µU/mL) and D₀ (13.47 ± 0.63 µU/mL). It is noticeable from the Figure 5 that D₂-group showed maximum rise for insulin (2.37%) trailed by D₁ (1.88%) as compared to control (D₀) in study I. Accordingly, the study II presented a significant increase for insulin in the groups D₁ and D₂ as 6.52 and 7.23%.

In current investigation, the effect of cinnamaldehyde was evaluated on lipoprotein diagnostic indicators as total cholesterol, LDL, HDL and triglycerides in normal and hyperglycemic rats. It is evident from the F value that treatments exhibited significant variations on cholesterol level of various rats groups in study II whereas non-significant differences were observed in study I. The means for normal rats (study I) presented diminishing trend for cholesterol in D₀ (82.30 ± 3.95 mg/dL) trailed by D₁ (79.84 ± 4.23 mg/dL) and D₂ (79.36 ± 4.12 mg/dL). The cholesterol level in study II (hyperglycemic rats) showed maximum decline in D₂ (91.90 ± 4.64 mg/dL), whilst D₁ and D₀ had values as 93.51 ± 3.87 and 99.77 ± 4.26 mg/dL, respectively. In study II, the hyperglycemic rats exhibited a decrease in cholesterol level from 7.89% (D₂) to 6.27% (D₁).

In present research project, D₂ (diet containing nutraceutical_SFE) caused maximum reduction in LDL followed by D₁ (diet containing nutraceutical_CSE) as compared to D₀ (control diet). In study I, maximum LDL level was observed in D₀ (31.21 ± 2.05 mg/dL) followed by D₁ (30.08 ± 1.77 mg/dL) group however, minimum level (29.96 ± 1.56 mg/dL) for this trait was in D₂. In study II higher reduction was exhibited by D₂ (11.37%) trailed by lower decrease in D₁ (10.86%) with mean LDL values for D₀ (45.57 ± 2.14 mg/dL) that momentously reduced to 40.62 ± 1.92 and 40.39 ± 1.33 mg/dL in D₁ and D₂, respectively.

Statistical analysis (F value) indicated that treatments imparted significant differences on high density lipoprotein (HDL) in hyperglycemic rats (study II). However, study I (normal rats) evidenced non-momentous variations due to the effect of diets. Means for HDL values in study I were 33.46 ± 1.93, 34.15 ± 2.08 and 34.37 ± 2.15 mg/dL in D₀, D₁ and D₂ groups, respectively. Nonetheless in study II, lowest HDL level was recorded in D₀ (42.68 ± 1.51 mg/dL) that significantly uplifted to 43.99 ± 1.27 and 44.20 ± 1.69 mg/dL in D₁ and D₂ groups.

Cinnamaldehyde can reduce triglyceride level in hyperglycemic rats. Means for triglyceride in study I indicated non-momentous declining trend for D₀, D₁ and D₂ as 68.32 ± 4.21, 66.63 ± 4.05 and 66.25 ± 3.94 mg/dL, respectively. Triglyceride level in study II revealed a diminishing tendency for all groups; maximum value in D₀ (75.69 ± 3.18 mg/dL) followed by D₁ (71.04 ± 2.66 mg/dL) and D₂ (70.50 ± 2.97 mg/dL).

Discussion

In a study, Shatwan et al. assessed the effect of cinnamon extract on feed intake of male Wistar rats. They were of the view that feed intake was significantly higher (35.99 ± 3.17 and 30.23 ± 3.58 g) in diabetic and cinnamon treated rats as compared to control (25.93 ± 1.33 g) group. This increment in feed intake was due to the increase of neuropeptides YmRNA and reduced leptin receptors activity with inadequate insulin supply ultimately reducing the weight gain in diabetic rats. The previous findings of Anand et al. further strengthened the present results. They deduced the significant increase of about 4.37 times (175 ± 16 mL/day) in fluid intake of untreated diabetic rats when compared with control (40 ± 3.2 mL/day). They also illuminated the inverse relation between the oral administration of cinnamon ® 20 mg/kg body weight and fluid intake, which resulted in decreased (74 ± 8 mL/day) fluid intake in diabetic rats. The results of present research regarding body weight in cinnamaldehyde (CND) administered male wistar rats are in agreement with the findings of Anand et al. They reported significant decrease 37.9% in body weight of STZ induced diabetic rats after 60 days induction. They also noticed the restoration in body weight 209 ± 10 g after oral intake of cinnamaldehyde (20 mg/kg) to diabetic rats near to control 224 ± 16 g.

The results relating to glucose reduction with cinnamon polyphenols are in harmony with the earlier work of Cheng et al. In a 12 weeks trial, they obse-
erved significant reduction in fasting blood glucose (FBG) level as 0.3, 14.6 and 18.9%, respectively in a dose dependent manner. In hyperglycemic condition, insulin resistance causes dysregulation of gluconeogenesis in hepatic tissues which results in elevated glucose level. Cinnamaldehyde helps to reduce blood glucose level by increasing insulin release and inhibiting the gene expression of phosphoenolpyruvatecarboxykinase & glucose-6-phosphatase. Later, Sailesh and Padmanabha, examined the hypoglycemic effects of cinnamon bark extract in alloxan induced diabetic rats and inferred that cinnamon polyphenols reduced blood glucose level in a 21 days trial. The role of superoxide dismutase (SOD) and glutathione peroxidase (GSH-Px) to scavenge oxygen free radicals and pancreatic tissue protection form oxidative damage is well known. In diabetic mice, cinnamon polyphenols enhances SOD and GSH-Px activities and decreases malondialdehyde (MDA) contents in pancreatic tissue thus provide beneficial effects against hyperglycemia. In a research study, Ismail evaluated the variations in insulin hormone level with provision of different levels i.e. 100 and 200 mg/kg body weight of cinnamon extracts. He expounded progressive increase in the serum insulin level in a dose dependent manner with minimum insulin level 0.89 ± 0.13 ng/mL in untreated diabetic rats which substantially increased to 2.43 ± 0.12 ng/mL in rats treated with cinnamon extract @ 200 mg/kg which further confirmed our results.

The present trend for serum cholesterol reduction is strengthened by the efficacy study of IM et al. Ethanolic extract of cinnamon polyphenols elucidated a marked reduction in hypertriglyceridemia and hypercholesterolemia with 42.79% decrease in total cholesterol level in group treated with 70% polyphenol contents as compared to diabetic control. According to another study, cinnamon provides protection against hypercholesterolemia by reducing the serum cholesterol level from 91.50 ± 9.26 to 76.75 ± 5.46 mg/dL in high cholesterol fed and cinnamon treated white male albino rats.

Cinnamon ability to reduce serum LDL cholesterol level is supported by the work of Askari et al. They concluded that cinnamon intervention results in substantial diminution 55.8 ± 40.8 mg/dL in LDL cholesterol as compared to placebo control 90.3 ± 17.7 mg/dL in patients suffering from non-alcoholic fatty liver disease. The outcomes of Hasanein et al. strengthened the results of recent study as they observed an elevated level of HDL as 34.06 ± 1.08 mg/dL in rats fed with high fat diet treated with aqueous cinnamon extracts. In a study researchers explicated the boosting effect of cinnamon polyphenols on antioxidant ability of diabetic mice against hyperlipidemia. They also recorded substantial reduction in serum triglyceride level of CPS (cinnamon polyphenols) treated mice at the end of study.

Conclusion

Prevention of various metabolic syndromes by using different dietary patterns has become a major health concern worldwide especially in developing countries like Pakistan. In this milieu, cinnamon (Cinnamomum zeylanicum) provides a wide range of nutraceuticals especially cinnamaldehyde. The current study was an effort to assess the therapeutic worth of cinnamaldehyde to cope with hyperglycemia. In present study, reduction in glucose level was more in rats fed with nutraceuticalcse diet (D2) as compared to nutraceuticalcse diet (D1) followed by control diet (D3). In the nutshell, cinnamaldehyde enrichment supplementation should be encouraged at mass level and consumer awareness regarding diet based therapies should be promoted.

Acknowledgements

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