Evaluation of Supplementation of Bittergourd Fermented Beverage to Diabetic Subjects

C.S. Devaki and K.S. Premavalli^{*}

Food Preservation and Sensory Science Division, Defence Food Research Laboratory, Siddarthanagar, Mysore – 570 011, Karnataka, India

Abstract: Diabetes mellitus is a chronic endocrine diseased condition reflected by higher level of blood glucose which is due to less insulin production, insulin action or both. Bittergourd juice consumption is being traditionally practiced for the treatment of diabetes mellitus in developing country such as India, but not supported through clinical data and highly bitter juice is difficult to drink. Therefore bittergourd fermented beverage with improved nutritional strength and taste was developed and supplemented to diabetic subjects and the evaluation was carried out. The evaluation of the bittergourd fermented beverage, in the first stage was carried out by supplementing the beverage to 30 diabetic subjects as an early morning drink in fasting condition and the control group was asked to drink water. The fasting and post prandial blood sugar levels were studied and diabetic symptoms were noted. The impact of supplementation of bittergourd fermented beverage on diabetic subjects showed that subjects had significant improvement in reducing the symptoms of diabetes, as well reduced the fasting and post prandial glucose levels by 31% and 25% respectively when compared with the control group. Further 16 diabetic subjects who expressed their consent were given the bittergourd fermented beverage for a period of 5 months and the results of the long duration supplementaion indicated that there was a reduction of fasting blood glucose by 43% and post prandial blood glucose by 41% reflecting advantage of continued consumption of the beverage. In order to understand the direct action of the beverage on blood lipid profile-serum cholesterol, triglycerides, low density lipoprotein cholesterol, high density lipoprotein cholesterol as measured before and after supplementation showed the changes by 4-7% which is not considerable. However the glycoslated haemoglobin indicated encouraging results after the supplementation showing good control from fair control. Therefore, the study clearly reflected the positive effect of bittergourd fermented beverage in reducing and controlling blood sugar levels.

Keywords: Bittergourd fermented beverage, diabetes, supplementation, biochemical parameters.

1. INTRODUCTION

Bittergourd (Momordica charantia L) also known as bittermelon, balsam pear or karela is widely cultivated as a vegetable and medicinal herb in many Asian countries. Bittergourd contains biologically active chemicals that include crude fat, crude protein, soluble dietary fiber, minerals, essential oil, flavonoids, phenolic acids, glycolsides, triterpenes and has been shown to exert hypoglycemic effects in animal models and humans [1-3]. It is frequently observed that many diabetic patients squeeze bitter gourd into juice and drink in the morning for natural remedy. The freshly squeezed bittergourd juice is extremely bitter thereby very difficult to drink. Therefore, fermentation process was utilized to improve the nutritional strength and to modify the taste of bittergourd juice using curd as starter culture [4] and the developed bittergourd fermented beverage was utilized for the supplementation study in the diabetic subjects. Besides, vegetable beverages are low caloric, nutritious and rich in antioxidants. The developed formulation as dietetic drink from bittergourd will be very convenient and cheap for consumers to take for health related problems such as diabetes. Diabetes mellitus is a chronic endocrine disease marked by higher level of blood glucose from defects in insulin production, insulin action or both. India has the maximum number of diabetic patients in the world and this had given the country the dubious distinction of being the "Diabetic Capital" of the world. Diabetes mellitus with its devastating consequences has assumed epidemic proportions as its prevalence is on a risk globally. By 2030 there would be 366 million diabetes throughout the world and 79.44 million diabetes in India alone [5]. WHO projects that deaths due to diabetes will double between 2005 and 2030. Diabetes are more prone to silent heart attack and it is one of the major risk factors for CHD. Therefore the developed bittergourd fermented beverage has an absolute requirement to supplement to diabetic subjects and see its efficacy on sugar profile of the supplemented subjects. A large number of research confirms the studies hypoglycemic effects of bittergourd in animal models, while only a few clinical trials have been done with diabetic patients. A trend in the animal research seems to imply the need for some insulin production. NIDDM animal models as responded to bittergourd treatment while IDDM models rarely did [6-11]. This mechanism seems to be confirmed by in vitro evidence that shows protection of and increased numbers of pancreatic β cells and in some cases an insulin-releasing activity [12-15]. Only

^{*}Address correspondence to this author at the Food Preservation Discipline, Defence Food Research Laboratory, Siddarthanagar, Mysore – 570 011, Karnataka, India; Tel: 0821 – 2473828; Fax: 0821 – 2473468; E-mail: premavalli52@yahoo.co.in

few papers are published on clinical trials using bittergourd in humans. In one case, 86% of the NIDDM patients experienced a hypoglycemic response to drinking an aqueous homogenized suspension of the vegetable pulp [16]. This was similar to a report that 73% of type II diabetics responded to 60ml of bittergourd juice [17]. In both studies, response was measured by reduction in glucose tolerance after glucose challenge. Though bittergourd is being traditionally practiced for the treatment of diabetes mellitus in developing country like India, there is not much support on clinical data, therefore the developed bittergourd fermented beverage was supplemented to diabetic subjects and the evaluation was carried out on the blood glucose levels.

2. MATERIALS AND METHODS

The evaluation of the bittergourd fermented beverage has been carried out by conducting the trials at Defence Food Research Laboratory, Mysore and Mounted Police Department, Mysore. Institutional Human Ethical Committee approval was obtained by University of Mysore, Mysore for the approval of the protocol and to undertake the study. A written consent was taken from the volunteering subjects. The information regarding their age, gender, education, anthropometric measurements such as height and weight, dietary pattern and physical activity was obtained before starting the supplementation programme.

The evaluation of the beverage was carried initially on 85 subjects and the subjects were grouped into 3 groups viz 30 subjects for the diabetic control group, 30 subjects in the diabetic experimental group and 25 for the non diabetic experimental group. Developed bittergourd fermented beverage [4] was utilized for the supplementation programme. The first stage was carried out by supplementing the fermented beverage (45ml) to the subjects as an early morning drink in fasting condition and the control group was asked to drink water. The information regarding their clinical symptoms, fasting blood sugar (FBS) and post prandial blood sugar (PPBS) were carried out in all the selected subjects. Subjects of both experimental and control group were examined in the duration of every 3 and 4 days for FBS and PPBS levels with Accu check of glucometer® for а period one month supplementation to observe the impact of the supplementation on sugar profile levels.

Further the study was carried out by supplementing the beverage with 16 diabetic subjects with their

consent for a period of 5 months The parameters which were considered for the second stage study were blood sugar profile, blood lipid profile – serum cholesterol, triglycerides (TGL), low density lipoprotein (LDL cholesterol), high density lipoprotein (HDL cholesterol) and glycoslated haemoglobin (HbA1c) were measured before and after the supplementation. Every week the blood sugar profile was analysed with Accu check glucometer[®].

2.1. Tools for Assessing Nutritional Status

2.1.1. Anthropometric

Anthropometric measurements such as height and weight were selected to assess the nutritional status using standard techniques. Height was measured using non-expandable fibre tape. Body Mass Index (BMI) was calculated as the ratio of [Weight (Kg) / Height ² (m)]. These were compared with the World Health Organizations (WHO) suggested Asian standards [18].

2.1.2. Dietary Pattern

Frequency of consumption of various food groups were elicited by the inmates. Diet survey using 24 hour dietary recall method was carried out for assessing dietary intake using standardized cups. The nutrient intake of the subjects was calculated using a ready reckoner [19].

2.1.3. Biochemical Analysis

The biochemical attributes namely fasting blood sugar (FBS), post prandial blood sugar (PPBS) were recorded with the help of Accu check glucometer[®]. Blood lipid profile – serum cholesterol, triglycerides (TGL), low density lipoprotein (LDL cholesterol), high density lipoprotein (HDL cholesterol) and glycoslated haemoglobin (HbA1c) were carried out on all the selected subjects in the second stage, prior and after supplementation were analysed by M/s Biochemical Diagnostic Laboratory, Mysore using standard methods.

2.1.4. Statistical Analysis

The data collected before and after supplementation of the study were tabulated in a hyper table using computed excel software programme. The data was analysed for test of significance using Student's 't' test in order to observe the impact of bittergourd fermented beverage supplementation before and after the study on biochemical parameters such as blood sugar and serum lipid profile and on the clinical symptoms o diabetes.

3. RESULTS AND DISCUSSION

Bittergourd's hypoglyecemic ingredients have been shown in animal and human studies. Polypeptide-p, a plant insulin, charantin, vicine, glycosides, and karavilosides improve blood sugar levels by increasing glucose uptake and glycogen synthesis in the liver, muscles, and fat cells. They also improve insulin release from pancreatic β cells, and repair or promote new growth of insulin-secreting ß cells. Bittergourd contains four very promising bioactive compounds. These compounds activate a protein called AMPK, which is well known for regulating fuel metabolism and enabling glucose uptake, processes which are impaired in diabetics [20-23]. In numerous studies, at least three different groups of constituents found in all parts of bittergourd have clinically demonstrated hypoglycemic properties (blood sugar lowering) or other actions of potential benefit against diabetes mellitus [11, 13, 16, 24-30]. These hypoglycemic chemicals include a mixture of steroidal saponins known as charantins, insulin-like peptides, and alkaloids. The hypoglycemic effect is more pronounced in the fruit of bittergourd where these chemicals are in greater abundance.

The developed bittergourd fermented beverage had thiamine 0.33mg/100ml, riboflavin 0.27mg/100ml, niacin 0.67mg/100ml, pyridoxine 0.27mg/100ml and vitamin C 33.5mg/100ml. Besides these, the total phenols 40mg/100ml and antioxidant activity (% inhibition) of 54.2% were present in the beverage [4]. The quinine content was 287mg/100ml, lesser than that of fresh juice and the beverage was highly acceptable when compared to fresh bittergourd juice. In the initial stage, out of total of 85 subjects, 55 subjects were taken for the experimental group, who had supplemented with the fermented beverage and 30 subjects to the control group, who were supplemented with drinking water. The experimental group constituted

30 subjects, who were diabetic and remaining 25 subjects were non-diabetic. In the control group there were 35% of subjects who were diagnosed with diabetes (Figure 1). Bittergourd fermented beverage (45ml) was provided in self standing PP bottles to subjects of experimental group with the instructions to drink in fasting condition as a first drink in the morning and control group received drinking water as supplementation. Subjects of both experimental and control group were re-examined after period of one month supplementation to observe the impact of the supplement on blood sugar profile changes.

3.1. Demographic Features

Age is one of the most important social and biological factors without which nutritional status cannot be assessed. Table 1 shows that among the total subjects of 85, 89% were males and 11% were females. The mean age of female and male was 50 years. Diabetic experimental group contained 10% female and 90% male, while non diabetic experimental group contained 84% male and 16% female, whereas control group had 7% female and 93% male (Table 1). The majority of the age group having diabetes was found in the age group of 40-60years. The high prevalence may be due to life style changes and rapid urbanization [31]. Around 72% of male and 22% of female were studied secondary level followed by 28% of male and 78% of female were degree/diploma qualified. It is more evident from Table 2 that no subjects were illiterate and had good qualification.

3.2. Anthropometric Data

The mean height, weight and body mass index of the subjects are given in Table **3**. The mean overall height in the experimental diabetic males was 169 ± 6.5 cms and that of the female subjects was



Figure 1: Grouping of subjects for bittergourd fermented beverage.

	Gender		Age range (years)				Moon ogo	
Number			Below 40		40-60		weat aye	
	Male	Female	Male	Female	Male	Female	Male	Female
85(Total)	76(89)	9(11)	7(9)	2(22)	69(91)	7(78)	50	50
30 (Diabetic Control)	28(93)	2(7)	1(4)	0(0)	27(96)	2(100)	52	49
30 (Diabetic Experimental)	27(90)	3(10)	0(0)	0(0)	27(100)	3(100)	52	57
25 (Non Diabetic Experimental)	21(84)	4(16)	6(29)	2(22)	15(71)	2(78)	45	45

Table 1: Age Distribution of the Subjects in Bittergourd Fermented Beverage

[#]Values in parenthesis indicates percentage.

Table 2: Educational Levels of the Subjects According to Gender Age Distribution of the Subjects in Bittergourd

 Fermented Beverage

Qualification	Gender			
quanication	Male	Female		
Total	85			
Total	76(89)	9(11)		
Illiterate	0(0)	0(0)		
Primary	0(0)	0(0)		
Secondary	55(72)	2(22)		
Degree/Diploma	21(28)	7(78)		

[#]Values in parenthesis indicates percentage.

151±1.5cms; in the experimental non diabetic males was 166±8.1cms and that of the female subjects was 155±7.2cms and in the control diabetic males was 168±7.5cms and that of the female subjects was 158±5.7cms. Weights of the subjects in the each group are given in Table 3. BMI was used as the indicator to determine the nutritional status. BMI gives important clues regarding the nutritional status of an individual and the type of life style led. The BMI and type of life style are interrelated. BMI \leq 18.5 kg/m² was observed as under nourished, $\geq 23 \text{ kg/m}^2$ and $\geq 27.5 \text{ kg/m}^2$ represent an increased risk and a high risk of suffering from Non Communicable Diseases (NCDs) respectively. It was evident that 31% had a high risk,

58% had an increased risk of suffering from NCDs, while about 11% of the subjects were observed as undernourished (Figure **2**).

3.3. Food and Nutrient Intake

Seventy percent of the subjects were non vegetarian and 30% of the subjects were vegetarian. The nutrient intake of the subjects and calculated percentage of adequacy of nutrients is given in Table 4. The nutritive value of the food consumed by the individual subjects was calculated using a ready reckoner [19]. Twenty four hour recall method was used to estimate the food intake by all the subjects. The nutrient intake of the subjects was compared with

Table 3:	Anthropometric Data of Control and Ex	perimental Group Sub	jects in Bittergour	rd Fermented Beverage
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	Gender						
Parameters	Male	Female	Male	Female	Male	Female	
	Control D		Experimental D		Experimental N D		
	28(93)	2(7)	27(90)	3(10)	21(84)	4(16)	
Height (cms)	168 ± 7.5	158 ± 5.7	166 ± 8.1	154 ± 7.2	169 ± 6.5	151 ± 1.5	
Weight (Kgs)	74 ± 10.5	63 ± 9.4	75 ± 8.8	69 ± 7.9	70 ± 10.3	64 ± 15.2	
BMI (Kg/m ²)	26.3 ± 3.2	25.1 ± 2.0	27.3 ± 2.5	29.1 ± 1.4	25.0 ± 4.4	28.0 ± 6.9	



Figure 2: BMI classification of subjects in bittergourd fermented beverage.

Table 4:	Percent Adequacy of	Nutrient Intake with Refere	ence to RDA in Bittergourd	Fermented Beverage
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Group		No	Energy	Protein	Fat	Calcium	Iron
			kcal	gm	gm	mg	mg
Control Diabetic	Male	28(93)	75	65	125	102	78
	Female	2(7)	81	66	112	112	86
Experimental Diabetic	Male	27(90)	79	62	114	73	59
	Female	3(10)	80	62	123	79	63
Experimental Non Diabetic	Male	21(84)	74	68	158	98	68
	Female	4(16)	78	69	156	107	78

[#]Values in parenthesis indicates percentage.

the RDA for Indians given by ICMR [32] and found to be alarming. Protein (62%-69%) intake was not adequate in subjects. The fat intake of the subjects was very high (112%-158%) compared to RDA and this was reflected in their high BMI values (Table 3). Calcium intake was meeting the ICMR recommendations in female group as they were consuming more amount of milk and its products every day. When the food frequency was recorded rice, ragi was used in the daily diet and wheat was used on alternate days. Redgram dhal was used daily to prepare sambar or rasam. Horsegram dhal, blackgram dhal, greengram dhal, bengalgram dhal and field bean were used occasionally. Other vegetables such as beans, cabbage, radish, capsicum, leafy vegetables were used more frequently than potatoes, carrot. Milk was used in beverages like coffee, tea and milk, but the frequency of consumption was more in female group.

3.4. Effect of Feeding on Clinical Signs and Symptoms

Figure **3** shows that the majority (61%) of the subjects had diagnosed for diabetes before two years.

The frequently seen symptoms in diabetes were recorded in the experimental diabetic group subjects before and after the supplementation of the beverage. Majority of the subjects had polyhagia (64%), fatigue (61%), polyurea (49%), general weakness (46%) and burning feet (39%) before the supplementation. When recorded after the supplementation of bittergourd beverage they showed very good response to reduce the above mentioned symptoms (Figure 4). It is noteworthy to mention that, Polyphagia was reduced by 38%, fatigue was reduced by 37%, polyurea reduced by 33%, general weakness by 31% and burning feet by 19%. Exercise is considered to be beneficial in the treatment and prevention of insulin sensitivity and much of the effect occurs in muscle. As the selected subjects were also from the Police department, the subjects were actively involved in the physical activities such as horse riding, yoga, therefore hose riding too was considered as one of the physical activity. It is evident from Figure 5 that subjects who were diabetic were actively involved in the exercises such as horse riding (40% to 33%), yoga and walking (13 to 17%) but majority of the non diabetic (48%) subjects were not involved in any physical activity.



Figure 3: Subjects diagnosed for diabetes in bittergourd fermented beverage.



Figure 4: Effect of feeding bittergourd fermented beverage on clinical symptoms in experimental diabetic group.



Figure 5: Percentage of physical activity in the subjects in bittergourd fermented beverage.

3.5. Effect of Feeding on Blood Glucose Levels of the Subjects

The fasting and post prandial blood glucose levels were analysed in the 85 subjects before and after the supplementation of the bittergourd fermented beverage by Accu check glucometer[®]. Fasting blood sugar was drawn after the subjects had fasted overnight and after one and half hour to two hours after breakfast the blood was again drawn for post prandial blood sugar levels. Results from Figures 6 & 7 shows the changes in



Figure 6: Effect of supplementation of bittergourd fermented beverage on fasting blood glucose levels.



Figure 7: Effect of supplementation of bittergourd fermented beverage on post prandial blood glucose levels. ***Note:** CMD: Control male diabetic; CFD: Control female diabetic; EMD: Experimental male diabetic; EFD: Experimental female diabetic; EMND: Experimental male non diabetic; EFND: Experimental female non diabetic.

fasting and post prandial blood sugar levels of the subjects in each group. Supplementation of bittergourd fermented beverage showed reduction of blood sugar levels in both diabetic and non diabetic subjects. In the experimental diabetic subjects FBS reduced from 213mg/dL to 146mg/dL in males (Figure 6) and from 176mg/dL to 134mg/dL in females (Figure 6), and PPBS reduced from 263mg/dL to 196mg/dL in males and from 276mg/dL to 227mg/dL in females respectively. Around 24 to 31% of FBS and 18-25% of PPBS was reduced in the experimental group reflecting the hypoglycemic effect of bittergourd fermented beverage, while in the control group it was observed that reduction of 4mg/dL and 15mg/dL in FBS and PPBS levels respectively. It is clearly seen from Figures 6 & 7 that, the blood glucose levels were not reduced and it was almost same in the control group of subjects. In the group of non diabetic subjects the beverage had more influence on PPBS than FBS.

Around 25% of reduction was observed in PPBS in female group (Figure 7) and 15% in male group (Figure 7), where as it has not much influence on FBS levels. Therefore, even non diabetic subjects too can consume the developed beverage as a health drink as it influenced more on PPBS by not affecting FBS. Bitter gourd contains a lectin that has insulin-like activity. The insulin-like bioactivity of this lectin is due to its linking together 2 insulin receptors. This lectin lowers blood glucose concentrations by acting on peripheral tissues and, similar to insulin's effects in the brain, suppressing appetite. This lectin is likely a major contributor to the hypoglycemic effect that develops after eating bittergourd and why it may be a way of managing adultonset diabetes. Lectin binding is non-protein specific, and this is likely why bittergourd has been credited with immunostimulatory activity - by linking receptors that modulate the immune system, thereby stimulating said receptors. Charantin extracted by alcohol, is a

hypoglycemic agent composed of mixed steroids that is more potent than the drug tolbutamide, which is sometimes used in the treatment of diabetes to lower the blood sugar levels. Momordica also contains an insulin like polypeptide, polypeptide-P, which lowers blood sugar levels when injected subcutaneously into type 1 diabetic patient. The oral administration of 50-60 ml of the juice has shown good results in clinical trials. The effect of Momordica charantia on glucose and insulin concentrations was studied in nine non-insulindependent diabetics and six non-diabetic laboratory rats. A water-soluble extract of the fruits significantly reduced blood glucose concentrations during a 50 g oral glucose tolerance test in the diabetics and after force-feeding in the rats. Fried fruits consumed as a daily supplement to the diet produced a small but significant improvement in alucose tolerance. Improvement in glucose tolerance was not associated with an increase in serum insulin responses. These results shown that improves glucose tolerance in diabetes. Bittergourd has been shown to increase the number of beta cells in the pancreas thereby improving the body's ability to produce insulin. The fruit has also shown the ability to enhance cells' uptake of glucose, to promote insulin release, and potentiate the effect of insulin [33]. With the above results bittergourd fermented beverage showed excellent results in reducing the diabetic symptoms and reduced FBS and PPBS in the experimental groups. Twenty eight percent reduction in fasting blood glucose level was observed in experimental group and 22 % reduction in postprandial blood glucose level was observed when compared to non significant changes in control group. This indicates that consumption of 45ml of the fermented bitter gourd beverage per day in empty stomach helps in reducing blood glucose levels in diabetic subjects. A scientific study at the Jawaharlal Institute of Postgraduate Medical Education and Research, India, has also proved that bitter melon increases insulin sensitivity [33]. Also, in the Philippine

Department of Health issued a circular stating that bittergourd, as a scientifically validated herbal medicinal plant, can lower elevated blood sugar levels. The study revealed that a 100 milligram per kilo dose per day is comparable to 2.5 milligrams of the antidiabetes drug Glibenclamide taken twice per day [34].

As there is evidence of research work carried out by the earlier researchers that bittergourd is having effect on lipid profile further studies were carried out on 16 Diabetic subjects to evaluate the effect of bittergourd fermented beverage on blood sugar profile, lipid profile and Glycoslated Haemoglobin (HbA1c). Baky et al. [35] reported that bitter gourd exert hypoglycemic, hypolipidaemic and antioxidative influence on both normoglycemic and hyperglycemic diabetic rats. bitter gourd extracts show triglyceride and cholesterol lowering activity in diabetic animals [36-38], as well as non-diabetic animals fed cholesterol-rich diets [39]. The subjects of the present were supplemented with the beverage for a period of 5 months. The above mentioned parameters were carried out before and after the fermentation. The supplementation of the beverage showed significant (p<0.001) decrease in estimated average glucose (mg/dL) and in Glycosalated haemoglobin (HaA1c-%) levels (Table 5). It is more evident from Figure 8 that the supplementation of fermented beverage for 5 months showed decrease in the levels of FBS and PPBS. FBS reduced (43%) from 210 to 120mg/dL and PPBS reduced (41%) from 283 to 167mg/dL. Estimated average glucose reduced from 161.05 ± 38.17 to 118.24 ± 25.39 mg/dL. HbA1c test tells us about the glucose level in blood over 3 months and the results indicated that the subjects had fair control of glucose levels before the supplementation, where as after the supplementation they dropped out to good control in the glucose levels. In order to understand the direct action of the beverage on blood lipid profile-serum cholesterol, triglycerides, low density lipoprotein cholesterol, high density lipoprotein cholesterol as

Table 5: Effect of Supplementation of Bittergourd Fermented Beverage on Biochemical Parameters

Blood Parameters	Before	After	
Estimated Average Glucose (mg/dl)	161.05 ± 38.17	118.24 ± 25.39 ^b	
HbA1c (%)	7.23 ± 1.33	5.74 ± 0.89 ^b	
Serum Cholesterol (mg/dL)	187.13 ± 28.08	180.21 ± 13.83	
Serum Triglycerides (mg/dL)	156.35 ± 82.70	160.94 ± 78.03	
Serum HDL Cholesterol (mg/dL)	47.45 ± 9.04	$40.85 \pm 4.39^{\circ}$	
Serum LDL Cholesterol (mg/dL)	122.35 ± 36.91	128.98 ± 27.37	

[#]Values with different superscripts are significant difference with initial period at the level; ^ap>0.0001, ^bp<0.001, ^cp<0.01, ^dp<0.05.



Figure 8: Effect of supplementation of bittergourd fermented beverage on blood glucose levels of second batch subjects.

measured before and after supplementation showed the changes by 4-7% which is not considerable. Therefore, the supplementation of bittergourd fermented beverage on diabetic subjects showed significant improvement in reducing the symptoms of diabetes, reduced the fasting and post prandial glucose levels and helped to control the glucose levels when compared with the control group.

4. CONCLUSION

The impact of supplementation of bittergourd fermented beverage on diabetic subjects, in the initial study showed that subjects showed significant improvement in reducing the symptoms of diabetes, reduced the fasting and post prandial glucose levels and helped to control the glucose levels when compared with the control group. In the further study of bittergourd fermented beverage on 16 diabetic subjects showed reduction in fasting and postprandial blood sugar. HbA1c results indicated that after the supplementation they have raised to good control from fair control. The effect of beverage supplementation showed no considerable changes in the lipid profile and had no influence on lipid profile. Therefore, the study clearly reflected the positive effect of bittergourd fermented beverage in reducing and controlling blood sugar levels.

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