



## RESEARCH ARTICLE

# Estimation of glycemic index of single and multiple blend nutraceutical biscuits

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## ABSTRACT

The present study was undertaken for developing biscuits using functional ingredient like foxtail millet flour, flaxseed flour, rice bran oil and carrot flour and to assess its Glycemic Index (GI). The GI of the biscuits was estimated through animal experimentation. The animals were divided into two groups, one group was given 0.15 mg of standard glucose and the other group was fed with equi-load of carbohydrate content present in the developed biscuit. The GI of the developed biscuit was calculated from the area under blood glucose response curve of the test food and reference food. The GI of the developed biscuit was below 47 indicating that the biscuits were low glycemic. It can therefore be concluded that the developed low glycemic index biscuits from foxtail millet, flaxseed and carrot were nutritionally rich with enhanced sensory attributes. Its hypoglycemic effect could provide potential health benefits from degenerative diseases like diabetes mellitus..

**Keywords:** Glycemic index, multigrain, nutraceutical potential, biscuit, health benefit

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## INTRODUCTION

In developing countries like India with the increasing urbanization, technological, industrial and economic advances, the demand of processed food is also increasing rapidly. Among the processed foods, bakery products, particularly biscuits command wide popularity in rural as well as urban areas among all the age groups. Biscuits are one of the most popular widely consumed processed food product in India. Foods can be fortified with nutraceuticals to enhance their nutrition as well as provide health benefits (Directorate of Health Canada, 2002) and biscuits can be excellent vehicle for fortification using nutraceuticals.

Nutraceuticals play a very important role in control of diabetes. According to the American Diabetes Association, (2000) if the symptoms of developing diabetes are seen, then appropriate change in diet will certainly reduce the outcome of diabetes. The increase in diabetic population of the world is among the top ten causes of death. The eating habits and patterns of the diabetic patients are the main causes for poor glycemic control. It is reported in epidemiological studies that high fibre dietary intake can significantly reduce the incidence of diabetes (Sharma et al., 2013). Hence the present study was planned to develop and standardise biscuits for diabetes patients and to estimate the nutrient content and glycemic index of developed biscuits.

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## **MATERIALS AND METHODS**

### **Formulation of biscuits**

The biscuits were developed from flour mix prepared by substituting wheat flour with foxtail millet flour, flaxseed flour and carrot flour at different level of incorporation. For development of single and multiple blend nutraceutical biscuits the flour mix was prepared in three different ratios viz., 80:20, 70:30, and 60:40 respectively and were used to prepare 100 gm flour mix for biscuits. For multiple blend biscuits 50% of shortening was replaced with rice bran oil. The biscuits were prepared following the guidelines outlined by AACC (Anon., 1990) American Association of Cereal Chemists. Nutrient composition of the formulated biscuits were analysed using standard methods.

### **Acceptability trials of formulated biscuits**

Acceptability of biscuits were assessed by sensory evaluation. Sensory evaluation has been defined as a scientific discipline used to evoke, measure, analyse and interpret those responses to products as perceived through the senses of sight, smell, touch, taste and hearing (Sidel and Stone, 1993).

### **Glucose tolerance test**

The animals used for this experiment were white albino rats obtained from College of Veterinary Sciences, Dept of Pharmacology and Toxicology, AAU Khanapara. The animals were weighed (between 180 and 220 gram) and were divided into two groups, namely group A and group B feeding on single and multiple blend biscuits( test food) respectively. Reference group was formed which was fed on standard glucose. Under group A three groups were formed namely experimental group1, experimental group 2, experimental group 3 feeding on SB1, SB2 and SB3 respectively and under group B four groups were formed namely experimental group1, experimental group 2, experimental group 3 and experimental group 4 feeding on MB1, MB2, MB3 and MB4 respectively with 5 animals in each group, notably, with no statistical differences. The rats kept in a controlled condition, 12 hours light and dark cycle. Deionized distilled water was offered ad libitum. In the use of animals for experimentation, the guidelines of the Institutional Animal Ethics Committee were followed. Formulated single and multiple blend biscuits were taken as test samples.

### **Glucose tolerance test**

GTT was conducted on overnight fasted animals. Fasting blood glucose was drawn from the reference group before giving 0.15 g standard glucose dissolved in distilled water. Blood glucose was determined after feeding at 15, 30, 45, 60, 90 and 120 minutes. The same method was performed with the experimental groups by taking their fasting blood glucose and then giving them amount of test food containing 0.15 g carbohydrates for each group according to method outlined by Thannoun et al, (2010).

### **Blood sampling**

Blood samples were taken from tail tipping of the experimental animals. Blood glucose was determined by using glucose tester Device Accu check Roche.

### Measurement of incremental area under curve and glycemic index

Glycemic Index (GI) for each diet was determined by calculation of Incremental Area Under two hours of blood glucose response or Curve (IAUC) for each diet and compared with the IAUC for glucose solution standard according to the method of Jenkins et al. (1981), Jenkins et al. (1987) and Wolever et al. (1992) using the following equation:

$$GI = \frac{\text{Incremental Area Under 2h blood glucose Curve for food}}{\text{Incremental Area Under 2h blood glucose Curve for glucose}} \times 100$$

The glycemic index calculated for the test samples were classified by using the GI scale given below:

Glycemic index scale (Foster-Powell et al., 2002)

Low: 55 or low  
 Medium: 56-69  
 High: 70 or more

### Formulation and standardisation of biscuits

The biscuits were developed using standard recipe and acceptable level of incorporation of foxtail millet flour, flaxseed flour and carrot flour in biscuits was assessed by incorporating 20%, 30% and 40% of each flour in wheat flour and standardized for sensory characteristics. Acceptability trials were conducted by a semi-trained panel consisting of 15 numbers of judges from the Department of Food science and Nutrition and Department of Horticulture, AAU respectively. Scoring was done on nine point hedonic scale.

## RESULTS AND DISCUSSION

### Moisture

The moisture contents of single and multiple blend nutraceutical biscuits are presented in Table 1. The data revealed that the moisture content of single blend nutraceutical biscuits were 3.80 g/100g, 4.20 g/100g, 3.70 g/100g in SB1A, SB2B and SB3A respectively. Similarly, the moisture content of multiple blend nutraceutical biscuits were 3.41 g/100g, 3.83 g/100g, 3.15 g/100g and 4.10 g/100g in MB1A, MB2B, MB3C, and MB4A respectively. The moisture content of single and multiple blend nutraceutical biscuits increased significantly ( $p < 0.05$ ) than control biscuits. Increase in moisture content of foods may be due to the increase in protein content because of the hydrophobic property of protein. High moisture content in biscuit may also be because of higher crude fibre content which attributes to higher water absorption and moisture binding capacity.

Similar, observation were also made by Cui and Mazza (1996) that high moisture content of flaxseed biscuits can be attributed to fibre present in flaxseed (30%) which has higher moisture retention property. The findings of the present study were similar with the work done by Baljeet et al. (2014), on a healthy carrot fortified biscuit containing 3.1% moisture. The findings of the present study are comparable with values reported by Sujitha and Devi, 2014 of 3.58 g/100g of fibre in a flaxseed flour and buckwheat flour incorporated biscuit. Baljeet et al. (2014) also reported similar results that biscuit incorporated with 10-20% of

carrot powder increases the crude fibre content from 2.2 to 3.2 g/100g which might be due to higher content of crude fibre in carrot than refined wheat flour.

**Table 1. Nutrient composition of single and multiple blend nutraceutical biscuits**

| Nutrient           | Products            |                     |                     |                     |                     |                     |                     |                     | CD<br>0.05 |
|--------------------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|------------|
|                    | Control             | SB1A                | SB2B                | SB3A                | MB1A                | MB2A                | MB3A                | MB4A                |            |
| Moisture (g)       | 3.56 <sup>d</sup>   | 3.80 <sup>b</sup>   | 4.20 <sup>a</sup>   | 3.70 <sup>c</sup>   | 3.41 <sup>e</sup>   | 3.83 <sup>b</sup>   | 3.15 <sup>f</sup>   | 4.10 <sup>a</sup>   | 0.11       |
| Protein (g)        | 6.00 <sup>g</sup>   | 9.36 <sup>a</sup>   | 8.53 <sup>c</sup>   | 5.0 <sup>h</sup>    | 9.06 <sup>b</sup>   | 7.25 <sup>f</sup>   | 8.04 <sup>d</sup>   | 7.74 <sup>e</sup>   | 0.10       |
| Fat (g)            | 14.60 <sup>b</sup>  | 14.00 <sup>c</sup>  | 18.60 <sup>a</sup>  | 14.23 <sup>d</sup>  | 15.41 <sup>b</sup>  | 13.06 <sup>d</sup>  | 14.03 <sup>c</sup>  | 14.33 <sup>c</sup>  | 0.94       |
| Crude fibre (g)    | 0.82 <sup>h</sup>   | 2.25 <sup>g</sup>   | 16 <sup>a</sup>     | 2.92 <sup>c</sup>   | 3.26 <sup>b</sup>   | 1.78 <sup>e</sup>   | 1.70 <sup>f</sup>   | 2.08 <sup>d</sup>   | 0.14       |
| Total minerals (g) | 0.73 <sup>f</sup>   | 1.78 <sup>c</sup>   | 2.55 <sup>a</sup>   | 1.12 <sup>e</sup>   | 2.22 <sup>b</sup>   | 1.40 <sup>d</sup>   | 2.08 <sup>b</sup>   | 1.35 <sup>d</sup>   | 0.14       |
| Carbohydrate (g)   | 73.90 <sup>a</sup>  | 68.7 <sup>e</sup>   | 61.06 <sup>g</sup>  | 73.30 <sup>b</sup>  | 66.60 <sup>f</sup>  | 72.21 <sup>b</sup>  | 70.95 <sup>c</sup>  | 70.25 <sup>d</sup>  | 0.14       |
| Iron (mg)          | 2.08 <sup>e</sup>   | 2.22 <sup>d</sup>   | 4.65 <sup>a</sup>   | 2.04 <sup>e</sup>   | 3.11 <sup>b</sup>   | 2.31 <sup>c</sup>   | 3.08 <sup>b</sup>   | 2.99 <sup>b</sup>   | 0.14       |
| Beta-Carotene (mg) | 0.024 <sup>e</sup>  | 0.033 <sup>e</sup>  | 0.029 <sup>e</sup>  | 3.41 <sup>a</sup>   | 0.033 <sup>e</sup>  | 2.32 <sup>b</sup>   | 2.04 <sup>d</sup>   | 2.21 <sup>c</sup>   | 0.17       |
| Energy (kcal)      | 457.00 <sup>a</sup> | 437.33 <sup>d</sup> | 445.00 <sup>b</sup> | 437.00 <sup>d</sup> | 440.66 <sup>c</sup> | 437.33 <sup>d</sup> | 441.66 <sup>c</sup> | 440.66 <sup>c</sup> | 1.33       |

Means within rows separated by Duncan's multiple range test  $P < 0.05$ . Means followed by the same letter shown in superscript(s) are not significantly different.

### Crude protein

The crude protein contents of multiple blend nutraceutical biscuits are presented in Table 1 SB1A contains highest amount of protein 9.36 g/100g, SB2B contain 8.53 g/100g and SB3A contains 5.61 g/100g. The crude protein content increased significantly ( $p < 0.05$ ) due to incorporation of foxtail millet, flaxseed flour and decreased due to incorporation of carrot flour. Similarly the crude protein contents of multiple blend nutraceutical biscuits also significantly increased ( $p < 0.05$ ). The data revealed that the crude protein content of multiple blend biscuits were 9.06 g/100g, 7.25 g/100g, 8.04 g/100g, and 7.74 g/100g in MB1A, MB2A, MB3A and MB4A respectively.

The protein content of all single and multiple blend biscuits were found to be higher than that of control biscuits except SB3A which was prepared from incorporation of carrot flour and protein content was lower as compared with control biscuits. Mridula in 2011 reported that addition of carrot powder lowered the total protein content of biscuit samples significantly, which might be due to lower protein content in carrot powder (0.9 g/100g) as compared to other ingredients of composite flour blends. But a significant increase in protein content was noticed in other blends which might be due to high protein content of foxtail millet flour and flaxseed flour. Protein content of foxtail millet is 11.5 g/100g and flaxseed flour is 20 g/100g as reported by Gopalan et al. 2000. Masoodi et al. in 2012 and Shiny and Sheila in 2014 fortified biscuit with flaxseed and millets respectively, and reported that protein content increased because of incorporation of flaxseed and millets in biscuits and ranged between 8.62 g/100g to 10.08 g/100g.

### Crude fat

The fat content of single and multiple blend nutraceutical biscuits are presented in Table 1. The data revealed that the fat content of single blend nutraceutical biscuits were 14 g/100g, 18.60 g/100g and 13.23 g/100g in SB1A, SB2B and SB3A respectively. Similarly the fat contents of multiple blend nutraceutical biscuits were 15.41 g/100g, 13.06 g/100g, 14.03 g/100g, 14.33 g/100g in MB1A, MB2A, MB3A and MB4A respectively. The fat content of the developed single and multiple blend nutraceutical biscuits significantly ( $p < 0.05$ ) decreased in all the blends than control biscuits but was at par with MB1A blend and increased significantly in SB2B. This might be due to incorporation of flaxseed in both the blend and flaxseed flour contains a high amount of fat i.e. 38 g/100g (Hussain, 2008).

The results of the present study were similar to the work of Ganokar and Jain in 2014 who reported that biscuits made with flaxseed had a high fat content.. Shiny and Sheila (2014) also found the fat content of millet based cookies to be 13 g/100g which is similar to the findings of the present investigation. Among the different single and multiple blend nutraceutical biscuits a significant decrease ( $p < 0.05$ ) in fat content was observed in SB3A biscuits incorporated with carrot flour when compared with control biscuits which might be due to low oil absorption capacity of carrot flour. Similar observation was also reported by Baljeet et al. (2014) that a decrease in fat content with increasing extent of substitution of carrot flour might be due to low oil absorption capacity of carrot powder in comparison with wheat flour.

### Ash

Form the results presented in Table 1 it can be observed that the incorporation of foxtail millet, flaxseed and carrot powder in wheat flour biscuits significantly increased ( $p < 0.05$ ) the ash content of biscuits. The data reveal that the ash content of single blend biscuits were 1.78 g/100g, 2.55 g/100g, and 1.12 g/100g in SB1A, SB2B and SB3A respectively. Similarly the ash contents of multiple blend nutraceutical biscuits were 2.22 g/100g, 1.40 g/100g, 2.08 g/100g and 1.35 g/100g in MB1A, MB2A, MB3A and MB4A respectively. The increase in ash content in the developed biscuits may be due to the presence of minerals in the composite flour blends.

The findings of the present study are similar to that of Shiny and Sheila (2014) and Mridula (2011) who reported an increase in ash content of biscuits incorporated with millet flour and carrot flour respectively and ranged between 1.82 g/100g to 1.19 g/100g. Similar values of ash content were also reported by Sujitha and Devi (2014) who had developed flaxseed incorporated cookies and found an ash content of 2.09 g/100 g.

### Crude fibre

The crude fibre contents of single and multiple blend nutraceutical biscuits are presented in Table 1 The data revealed that the crude fibre content of single and multiple blend nutraceutical biscuits significantly increased ( $p < 0.05$ ) than the control biscuits. The crude fibre content of single blend nutraceuticals biscuits were 2.25 g/100g, 16 g/100g, and 2.92 g/100g in SB1A, SB2B and SB3A respectively. Similarly the crude fibre contents of multiple blend nutraceutical biscuits were 3.26 g/100g, 1.78 g/100g, 1.70 g/100g, and 2.08 g/100g in MB1A, MB2A, MB3A and MB4A respectively. The increase in crude fibre content of the nutraceutical biscuits might be due to higher content of crude fibre in carrot, flaxseed and foxtail millet.

## Carbohydrate

The carbohydrate contents of single and multiple blend nutraceutical biscuits are presented in Table 1. The data reveal that the carbohydrate content of single blend nutraceutical was 68.70 g/100g, 61.06 g/100g and 73.30 g/100g in SB1A, SB2B and SB3A respectively. Similarly in multiple blend nutraceutical biscuits carbohydrate content was 66.6 g/100g in MB1A, 72.21 g/100g in MB2A, 70.95 g/100g in MB3A and 70.25 g/100g in MB4A. The carbohydrate contents of single and multiple blend nutraceutical biscuits decreased significantly ( $p < 0.05$ ) than control biscuits. The high carbohydrate content in control biscuit may be due to high carbohydrate in refined wheat flour which mainly consist of endosperm portion of wheat and endosperm portion is a good source of carbohydrate.

Jacobs and Steffen (2003) also reported that the carbohydrate content for control refined wheat flour was significantly higher than foxtail millet flour, probably because refined wheat flour contains mainly the endosperm portion which is a good source of carbohydrate. Masoodi and Bashir in 2012 and Shiny and Sheila in 2014 reported carbohydrate content ranged from 68-80 g/100g in a nutritious biscuit enriched with flaxseed and millets.

## Beta Carotene

Beta Carotene contents of the single and multiple blend nutraceutical biscuits are presented in Table 1. The data revealed that the Beta-Carotene content of single blend nutraceutical biscuits were 0.033 mg/100g in SB1A, 0.029 mg/100g in SB2B and in SB3A it was 3.41 mg/100g. Similarly the Beta Carotene content of multiple blend nutraceutical biscuits were 0.033 mg/100g, 2.32 mg/100g, 2.04 mg/100g, and 2.21 mg/100g in MB1A, MB2A, MB3A, and MB4A respectively.

Gayas in 2012 reported that enrichment of wheat flour with low concentrations of carrot powder increases the beta carotene content of product. The findings of the present study is similar to the work done by Mridula (2011) who reported an increase in beta carotene content of 2.39 to 3.72 mg/100g in a carrot and soy fortified biscuit with 10%- 20% level of incorporation of carrot flour which is in the same range as the present investigation.

## Iron content

The iron content of the single and multiple blend nutraceutical biscuits are presented in Table 1. The data reveal that the iron content of single blend nutraceutical biscuits were 2.22 mg/100g in SB1A, 4.65 mg/100g in SB2B and 2.04 mg/100g in SB3A. Similarly in multiple blend biscuits iron content was 3.11 mg/100g in MB1A, 2.31 mg/100g in MB2A, 3.08 mg/100g in MB3A and 2.99 mg/100g in MB4A. The iron content of the single and multiple blend nutraceutical biscuits increased significantly ( $p < 0.05$ ) than control biscuits.

The highest amount of iron was observed in SB2B which may be due to high content of iron in flaxseed flour i.e. 6.01 mg/100g (Hussain et al., 2008). And low iron content was found in control biscuits which can be contributed to low iron content of refined wheat flour. (Iron content of wheat flour 2.7 mg/g reported by Gopalan et al. 2000). In 2014, Florence et al., 2014 reported that replacement of refined wheat flour with semi refined flour from pearl millet in cookies significantly ( $p < 0.05$ ) increased the iron content in biscuits and had an iron content of 4.2 mg/100g which was comparable with iron content of present investigation.

## Energy

The energy contents of single and multiple blend nutraceutical biscuits are presented in Table 1. The data revealed that the energy content of single blend nutraceutical biscuits were 437.33 Kcal/100 g in SB1A, 445.00 Kcal/100 g in SB2B and 437.00 Kcal/100 g in SB3A respectively. Similarly the carbohydrate contents of multiple blend nutraceutical biscuits were 440.66 Kcal/100 g in MB1A, 437.33 Kcal/100 g in MB2A, 441.66 Kcal/100 g in MB3A and 440.66 Kcal/100 g in MB4A respectively. The energy content of the single and multiple blend nutraceutical biscuits were significantly ( $p < 0.05$ ) lower than the control biscuit. Many studies have reported the energy content of low glycemic millet and flaxseed based biscuit ranged from 424.6 Kcal/100g to 489 kcal/100gm (Shiny and Sheila, 2014; Anju and Sarita, 2010 and Hassan, 2012).

## Estimation of glycemic index of the developed single and multiple blend nutraceutical biscuits

GTT was conducted on overnight fasted animals. Fasting blood glucose was drawn from the reference group before giving 0.15 g standard glucose dissolved in distilled water. Blood glucose was determined after administration of glucose at 15, 30, 45, 60, 90 and 120 minutes. The same method was performed with the experimental groups by taking their fasting blood glucose and then giving them amount of test food containing 0.15g carbohydrates for each group and their blood glucose level was determined according to method outlined by Thannoun et al. in 2010. The blood glucose response for reference food and test foods are presented in Table 2 and 3.

**Table 2. Mean blood glucose response for different single blend nutraceutical biscuit fed to normal healthy rats**

| Experimental groups I | Samples          | Mean blood glucose level at different Time intervals (blood glucose mg/100ml) |          |          |          |          |          |         |
|-----------------------|------------------|---|----------|----------|----------|----------|----------|---------|
|                       |                  | 0 min   | 15 min   | 30 min   | 45 min   | 60 min   | 90 min   | 120 min |
| Control group         | Glucose standard | 84±2.34   | 124±0.59 | 156±2.65 | 140±2.31 | 115±0.15 | 98±1.65  | 85±3.55 |
| Group 1               | SB1A             | 81±1.02   | 85±0.45  | 89±1.54  | 95±2.09  | 103±0.39 | 97±2.44  | 91±0.89 |
| Group 2               | SB2B             | 78±1.11   | 81±0.88  | 86±1.88  | 90±1.05  | 93±3.10  | 91±0.76  | 88±2.76 |
| Group 3               | SB3A             | 93±3.06   | 102±0.20 | 110±3.21 | 116±0.98 | 116±3.22 | 100±0.33 | 96±2.85 |

Values are expressed in mean ± SD

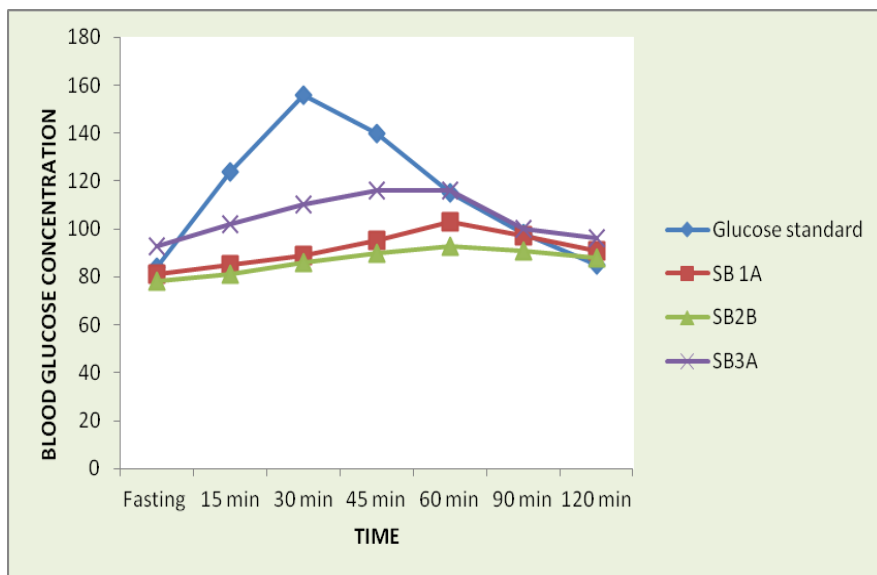
**Table 3. Mean blood glucose response for different multiple blend nutraceutical biscuit fed to normal healthy rats**

| Experimental groups II | Samples          | Mean blood glucose level at different Time intervals (blood glucose mg/100ml) |          |          |          |          |         |         |
|------------------------|------------------|---|----------|----------|----------|----------|---------|---------|
|                        |                  | 0 min   | 15 min   | 30 min   | 45 min   | 60 min   | 90 min  | 120 min |
| Control group          | Glucose standard | 84±2.34   | 124±0.59 | 156±2.65 | 140±2.31 | 115±0.15 | 98±1.65 | 85±3.55 |
| Group 1                | MB1A             | 82±3.65   | 84±2.69  | 89±1.11  | 93±2.89  | 96±1.23  | 92±1.12 | 91±1.04 |
| Group 2                | MB2A             | 80±2.43   | 85±0.88  | 89±1.09  | 93±3.74  | 101±2.87 | 95±1.87 | 88±0.66 |
| Group 3                | MB3A             | 77±0.58   | 79±3.21  | 82±2.78  | 89±3.48  | 92±3.09  | 89±2.09 | 86±0.98 |
| Group 4                | MB4A             | 79±3.44   | 83±2.16  | 88±2.86  | 93±0.98  | 95±4.08  | 93±3.44 | 89±1.55 |

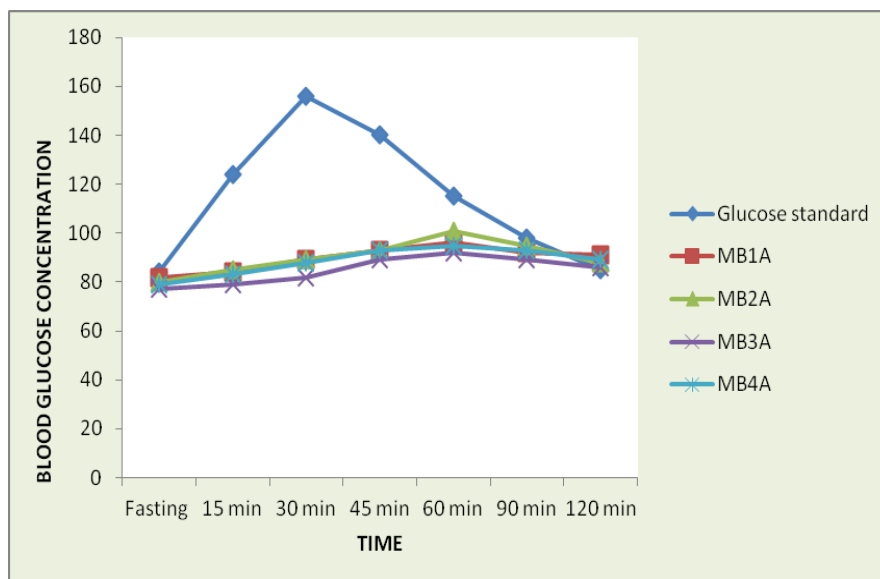
Values are expressed in mean ± SD

**Blood glucose response for single and multiple blend nutraceutical biscuits**

The blood glucose response of each test food were evaluated and presented in Table 2 and 3 for single and multiple blend nutraceutical biscuits respectively. The blood glucose level was monitored at fasting period and after feeding at an interval of 15 min, 30 min, 45 min, 60 min, 90 min and 120 min, respectively. The peak blood glucose level after consumption of the biscuits was obtained in 60 min which decreased till the observation period of 120 min (2 hrs) indicating slow digestion and absorption of the nutraceutical biscuits, whereas reference food showed peak blood glucose level in 30 min and then decreased progressively till the observation period of 120 min.(Fig. 1 and 2).



**Fig. 1. Mean blood glucose response curve for different single blend nutraceutical biscuit fed to normal healthy rats**



**Fig. 2. Mean blood glucose response curve for different multiple blend nutraceutical biscuit fed to normal healthy rats**



This result is attributed to the presence of dietary fibre content in the different biscuits which may have protected starch from enzymatic degradation (Wursch and PiSunyer, 1997). Also, during the baking of biscuits, 1, 6 anhydro D-glucose units may have been liberated from the starch and other polysaccharides to form enzyme resistant complexes, which are not readily digested (Garcia-Alonso and Goni, 2000; Leeman et al., 2005). Several authors have reported that during baking, a fraction of starch rendered itself inaccessible to amylases and total dietary fibre content increases due to retrogradation and thus slows down the digestion process (Rizkalla et al., 2007).

It was reported by Jalil et al. (2012) also showed that a high intake of millet based dietary fibre, improved the glycemic control, decreased the hyper insulinemia, and lowered the plasma lipid concentrations in patients with type 2 diabetes. Thus in the present investigation considerable level of positive change in glycemic parameters may be attributed to presence of fibre content in foxtail millet flour and flaxseed flour which slowly digested and absorbed in the intestine and hence plays a potential role in lowering glycemic index and thereby management of diabetes.

#### **Incremental area under blood glucose response curve (iAUC)**

From the above glucose response, incremental area under blood glucose response curve was calculated (Wolover et al., 1991). The (iAUC) value of reference food and test food are presented in Table 4. and 5. The mean (iAUC) of reference food was 3202 and single blend nutraceutical biscuits were 1196, 1005 and 1508 for SB1, SB2 and SB3 respectively. Similarly the mean (iAUC) of reference food was 3202 and multiple blend nutraceutical biscuits were 1085, 1380, 1222, and 1335 for MB1, MB2 MB3 and MB4 respectively. From the mean (iAUC) values it can be observed that the blood glucose levels for the test products were markedly lower than orally administered glucose.

#### **Estimation of Glycemic index of developed single and multiple blend nutraceutical biscuits**

Glycemic index of single and multiple blend were calculated from their iAUC and from iAUC of reference food. The GI of test samples, single blend nutraceutical biscuits and multiple blend nutraceutical biscuits are presented in Table 4. and 5. The glycemic index of single blend nutraceutical biscuits were 37 in SB1A, 31 in SB2B and 47 in SB3A. The lowest GI was observed in SB2B blend which might be due to high content of fibre in SB2B blend due to incorporation of flaxseed flour. Similarly the glycemic index of single blend nutraceutical biscuits were 33 in MB1A, 43 in MB2A 38 in MB3A and 41 in MB4.

**Table 4. Mean incremental area under curve (iAUC) and glycemic index for different single blend nutraceutical biscuit**

| Samples          | iAUC mg.min/100ml | GI |
|------------------|-------------------|----|
| Glucose standard | 3202              | -  |
| SB1A             | 1196              | 37 |
| SB2B             | 1005              | 31 |
| SB3A             | 1508              | 47 |

**Table 5. Mean incremental area under curve (iAUC) and glycemic index for different multiple blend nutraceutical biscuit**

| Samples          | iAUC mg.min/100ml | GI |
|------------------|-------------------|----|
| Glucose standard | 3202              | -  |
| MB1A             | 1085              | 33 |
| MB2A             | 1380              | 43 |
| MB3A             | 1222              | 38 |
| MB4A             | 1335              | 41 |

Findings of the present study were similar with the work done by Anju and Sarita (2010), that incorporation of foxtail millet in refine flour biscuits decreased the glycemic index of the biscuit which might be due to foxtail millet having higher soluble fibre and starch content. Many studies have also reported that incorporation of foxtail millet, flaxseed, buckwheat and soy flour in refine flour biscuits decreased the glycemic index of the biscuit which might be due to the high fibre content in the functional ingredients (Hassan, 2012 and Vujić et al., 2014). The high soluble dietary fibre content has been found to reduce gastric emptying and absorption of glucose after a meal, resulting in improved glucose tolerance. The soluble dietary fibre component has been reported to decrease the activity of digestive enzymes, thus resulting in incomplete hydrolysis of carbohydrates, protein and fats and delaying absorption (Wursch and Pi- Sunyer, 1997) Soluble dietary fibres have been shown to alter food texture, structure, and viscosity, and, therefore, the rate of starch degradation and digestion (Brennan and Samyue, 2004), which is related to the regulation of postprandial glucose levels. High fibre intake has been linked to a decreased risk of diabetes has been reported by (Wannamethee et al., 2009).

**Table 6. Pearson correlation coefficient between GI and carbohydrate and crude fibre content of developed single and multiple blend biscuits**

| Variable     | Correlation of GI with fibre and carbohydrate content of developed biscuits(r) |
|--------------|--|
| Carbohydrate | 0.922372 NS  |
| Crude fibre  | -0.5859 **   |

\*\*Correlation is significant at 0.01 level (2-tailed); NS : Non-significant

### Correlation between GI and crude fibre content of developed single and multiple blend biscuits

Pearson correlation showed that there was a significant negative correlation ( $r=0.5859$ ;  $p<0.01$ ) between the glycemic index and crude fibre content of developed single and multiple blend biscuits (Table 6). Similar results were reported by Meyer et al. (2000) that GI was inversely related to fibre content Barclay et al. (2007) also reported that fibre had a negative association with risk of diabetes in the age- and sex-adjusted analysis models in a prospective cohort study. Barclay et al. (2007) also conducted a

prospective cohort study, to examine the link between glycemic index (GI) and fibre and incidence of type two diabetes in an older Australian population and found that fibre had a negative association with risk of type two diabetes. It can be therefore concluded that low-GI foods developed should preferably be rich in dietary fibre, because increasing fibre in the diet may be protective against type two diabetes and other inflammatory diseases. High fibre intake may also decrease the risk of chronic diseases by improving the postprandial glycemic response and insulin concentration. Moreover they also help to prevent other complication that like hyperlipidemia, inflammatory bowel disease etc. (Lee et al., 2009).

### **Correlation between GI and carbohydrate content of developed single and multiple blend biscuits**

Pearson correlation showed that there was no significant correlation ( $r=0.5859$ ;  $p<0.01$ ) between the glycemic index and carbohydrate content of developed single and multiple blend biscuits Table 6. There is little evidence about the association between GI and carbohydrate intake. Dietary glycemic index have been more strongly related to coronary heart disease and diabetes than carbohydrate intake as reported by Liu et al. (2007). Total carbohydrate intake has a weak correlation with dietary glycemic index or dietary glycemic load, which takes into account the quality of carbohydrate. Barclay et al., in 2007 also conducted a prospective cohort study, to examine the link between glycemic index (GI) and incidence of type two diabetes in an older Australian population and reported that total carbohydrate was not correlated with type two diabetes.

### **CONCLUSION**

From the present study it can be stated that biscuits can be made with incorporation of nutraceuticals like flaxseed flour, foxtail millet flour, carrot flour without affecting the sensory attributes. These novel biscuits are nutritionally superior to that of refined wheat flour biscuits. The developed biscuits showed a low glycemic index which can be consumed by diabetic population. The outcome of the present research can be used as a valuable information for development of nutrient dense high fibre biscuits. Hence development and utilization of such functional ingredients will improve the overall health of the population as well as prevent the onset of degenerative disease and help in management of metabolic disorder like diabetes and obesity.

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