Development and Evaluation of Glycemic Index of Traditional Western Indian food *Khakhara* using Low Glycemic Ingredients

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Abstract: *Khakhara or flattened bread is a popular snack item in the Guajarati cuisine. It is traditionally baked product prepared from mainly wheat flour combined with millet flour. Low glycemic index foods have shown to have several therapeutic effects on diabetes, weight management and improvement in blood lipid concentration. Thus an attempt has been made to develop Khakhara using low glycemic ingredients like barley and sprouted soya flour and evaluate its glycemic index. Khakharas were prepared from a combination of barley flour, wheat flour and sprouted soya flour (3:2:1) in 5 different variations. The variations included fenugreek leaves, coriander leaves, Amaranthus leaves, mint leaves and coriander + mint leaves. These were evaluated for its acceptability using quantitative numerical scoring method by 20 semi trained panelists. Sensory and statistical analysis revealed coriander leaves Khakhara as the most acceptable. Proximate analysis revealed energy as 402.45kcals, protein 11.6g, fat 6.28g and dietary fibre as 3.38g.The product had a shelf life of 15 days under normal storage conditions. The glycemic index of the product estimated using internationally recognised methodology on 15 healthy subjects revealed a value of 56.8 indicating Khakhara as a low glycemic food. Hence the product is recommended for diabetics as well as a wholesome snack for kids and the normal population.*

Keywords: Khakhara, barley, Soya flour, Glycemic Index, Diabetes

1. Introduction

Khakharas are individually rolled out thin chapattis which are flattened and made crisp over a griddle or *tawa*. Originated from the state of Gujarat it is a part of the Jain Gujarati cuisine. *Khakharas* are made from mat bean, wheat flour and oil usually served during breakfast. It is also a popular tea time snack. It is eaten along with different chutneys and pickles. There are several varieties of *Khakhara* available like *methi, bajri, masala, pav bhaji, panipuri* etc.

Glycemic Index (GI) is defined as a measure of the blood-glucose-raising ability of the available carbohydrate in foods. It is expressed as a percentage of the Incremental Area under the Glycemic response Curve (AUC) elicited by a portion of food containing 50 g available carbohydrate in comparison with the AUC elicited by 50 g glucose in the same subject. A GI value of ≥70 is considered high, a GI value 56-69 inclusive is medium and a GI value ≤ 55 is low, where glucose = 100 (Brand Miller *et.al*, 2003).

Low-Glycemic Index foods may reduce the insulin demand, improve blood glucose control, reduce blood lipid concentrations and body weight and thus could help prevent diabetes-related cardiovascular events. Low GI foods have been found to increase satiety, reduce hunger and food intake in many short term feeding studies thus useful in weight management.

Ashwini Pande *et.al* (2012) reported significant hypoglycemic and hypolipidemic effects in type 2 diabetic subjects upon consumption of low glycemic load Indian vegetarian snacks and mixed meals for 4 continuous weeks. A decrease in 35.8% mean blood glucose, 79.8% decrease in Triglyceride levels and 39.9 mg% decrease in total cholesterol levels while an increase of 5.8 mg% in total High Density Lipoprotein fraction was observed. Very Low Density Lipoprotein fractions decreased to 16% while Low density lipoprotein levels decreased to 26.8 mg %.

Barley is a wonderfully versatile cereal grain with a rich nutlike flavour and an appealing chewy, pasta-like consistency, due to its gluten content. Barley β-glucan is a natural soluble dietary fibre that has been shown to lower glycaemic response when incorporated to a variety of foods.

Owing to the advantages of low glycemic index foods the aim was to develop *Khakhara* using low glycemic ingredients.

The objectives of the study were to identify the low glycemic ingredient, process the selected ingredient standardise and develop the product, subject it to analysis and then evaluate the glycemic index of the product.

2. Materials and Methods

The major raw materials used in the preparation of product include whole wheat flour, barley flour, soy flour and barley flour. Green leafy vegetables were used for variation purpose. These include coriander leaves, fenugreek leaves, mint leaves and amaranthus leaves.

1) Preparation of Flour

a) Whole wheat flour
Whole wheat was procured from the local market. It was cleaned to remove stones and extraneous matter, soaked overnight to reduce the phytate content, drained, and sundried. The dried grains were then milled into flour in a flour mill.

b) Barley Flour
Barley was procured from the local market. It was cleaned, washed to remove dirt and filth, and sundried. The dried grains were ground into flour in a flour mill. The flour was then slightly roasted to reduce the raw flavour. The glycemic index of barley is 37 (Int. table of glycemic index, 2002)

c) Sprouted Soybean Flour
Soybean also contains micronutrients, which include isoflavones, phytate, saponins, phytosterol, vitamins and minerals. Isoflavones may additionally have a beneficial role in lipid and glucose metabolism. Soybeans are the most abundant source of isoflavones in food. Amongst the soya products soya flour is reported to have high isoflavones content followed by soy milk, tofu, tempeh and miso. (Saidu, 2005).

Sprouted Soy flour was procured from the market. The glycemic index of soyabean is 15(Int. table of glycemic index, 2002)

d) Green Leafy Vegetables
Fresh, tender leafy vegetables were procured form the local vegetable market. The leaves were separated from stalks, unwanted leaves, wilted and ripe leaves. They were then washed thoroughly in water, drained completely, and chopped finely. The chopped vegetable was then weighed and added fresh.

2) Development of the product
Barley flour, wheat flour, and sprouted soy flour was used in a combination of 3:2:1. To the cereal – pulse flour blend (50 g) was added to other pre- processed vegetables (finely chopped green chillies – ½ t, 1 t ginger – garlic paste), (cleaned, washed and chopped green leafy vegetable -25g) turmeric powder- 1.0 g, salt – 2.0 g, . About 65ml of water was added and made into soft dough. The dough was then divided into small portions each weighing 35 g and rolled into a circular shape with the help of rolling pin and plate into 12 cm diameter and 0.2 cm. thickness. The circle was cooked on a pre-heated tawa on both sides using ¼ t clarified butter *(ghee)* until light pink colours spots appeared on the surface.

Using a folded muslin cloth the *Khakhara* was pressed from all sides and cooked over a slow flame until crisp.

3) Product Analysis
The organoleptic evaluation of the products was carried out by a panel of 20 semi- trained panelists using a numerical scoring method. The sensory parameters included colour, flavour, texture, taste and overall acceptability. The scores were then subjected to statistical analysis like mean score, 2 way ANOVA and coefficient of variation using MATLAB software 2007.The best variation was further evaluated for nutrient and shelf life analysis.

Nutrient Analysis
The nutrient analysis was carried using standardised protocols (AOAC, 2000). The parameters analysed include moisture, total ash, protein, total carbohydrate, total fat and dietary fibre content.

4) Shelf Life Analysis
Khakhara was subjected to microbiological analysis for the parameters – Total Viable Count and E-coli. The shelf life analysis was carried out by storing the product in the Low Density Polyethylene (LDPE) zip- lock pouches at room temperature. Sensory evaluation was carried out at definite time intervals, on the day of preparation (0 day), fifth day, tenth day and fifteenth day by a panel of 10 members using numerical scoring method from excellent to poor. The sensory parameters include colour, flavour, texture, taste and overall acceptability.

5) Evaluation of Glycemic Index
The most accepted product was taken for determination of glycemic index. Glycemic index was evaluated using the prescribed standardised internationally recognised methodology. Glycemic Index is defined as relation of the incremental area under the B-glucose response curve (IAUC) of a tested meal containing 50 g of digestible carbohydrates and the Incremental Area Under the B-glucose response Curve of the standard food, 50 g pure glucose. (IAUCS) (Jenkins DJ, *et.al,* 2002).

The students were first oriented about the concept of glycemic index prior to taking their consent. The students were enquired about their health status whether they were diabetic or any other complications or were taking any kind of medication etc. Girls falling under these criteria were not included in the study. Thus fifteen healthy subjects aged between 20- 25 years were selected for the study. The height of the selected subjects was measured using a measuring tape while weight of students was taken using LAICA Electronic personal scale, model PL8033. Basal Metabolic Index (BMI) was calculated as {Weight $(kg/Height\ m^2))$}. The average BMI of students was   22.7 kg/m^2.

The subjects were given general instructions to avoid any physical exertion, medication, fast or feast during the experimental period. The Glucose Tolerance Test (GTT) was carried on overnight fasted subjects at 7.00 a.m. with a glucose load of 50 g pure glucose (Glucon –D – Original powder, Heinz India). Pure glucose was given in the form of powder. This was used as the reference food and was administered on the first day. Fasting blood glucose level was checked prior to administration of reference food and test food. Blood sample was drawn after the initiation of food at specific intervals, at 30 minutes, 60 minutes, 90 and 120 minutes by finger prick method. The sample was collected on Glucocard ^{TM} sensor. The blood glucose was measured on glucometer brand Arkray Blood Glucose Monitoring Kit, Glucocard ^{TM} 01- mini. A time interval of 48 hours was maintained between the administration of the reference food and test food. The most acceptable product was served to the subjects in a fixed test portion containing 50 g digestible (available) carbohydrate. Similar procedure was followed for the test food too. Therefore, a total of five blood samples were collected from each subject on the same

day. The subjects were asked not to consume any sweetened drinks during the course of the test and avoid physical exertion. The Glycemic Index of the product was calculated by taking the average of the glycemic response of both the reference and test food at 30, 60, 90 and 120 minutes and a glycemic response curve was constructed. The incremental area under blood glucose response curve (iAUBGR) was calculated. The Glycemic Index value of each individual was calculated as

GI value for = $\dfrac{\text{iAUBGR curve after test food}}{\text{iAUBGR curve after reference food}}$ X 100
test food (%)

The average Glycemic Index of 15 subjects was taken as the Glycemic Index Value of the product. Student t - test was carried out to determine the significance.

3. Results and Discussion

From the sensory analysis *Khakhara* prepared from coriander leaves was found to be the most acceptable followed by fenugreek leaves. The results have been represented below.

Table 1: Mean Scores of Organoleptic Evaluation of *Khakhara*

Variations	Colour (5)	Flavour (5)	Texture (5)	Taste (5)	Overall Acceptability (5)
BWSS (Barley+Wheatflour+Sproutedsoybean flour)					
I	3.24±0.38	3.24±0.38	3.24±0.38	3.53±0.55	3.73±0.5
II	3.33±0.45	3.33±0.45	3.33±0.45	3.46±0.59	3.53±0.51
III	2.99±0.54	2.99±0.54	2.99±0.54	2.28±0.47	2.28±0.43
IV	3.03±0.43	3.03±0.43	3.03±0.43	3.13±0.44	3.14±0.34
V	3.08±0.26	3.08±0.26	3.08±0.26	3.13±0.3	3.43±0.5

I-Fenugreek , II- coriander , III- Amaranthus, IV- Mint V- Coriander + Mint

The mean scores from Table I reveal that Variation I scored high (3.53), (3.73) in taste and overall acceptability while Variation II has scored high mean scores (3.33) for colour flavour and texture parameters respectively. However, Variation III scored a poor score for its sensory parameters. Variation IV and V have scored an average score for acceptability.

Table 2: Anova and Coefficient of Covariation for *Khakhara*

Variations	Colour	Flavour	Texture	Taste	Overall Acceptability
BWSS					
I	7.00%	7.00%	7.00%	13.00%	12.00%
II	12.00%	12.00%	12.00%	12.00%	12.00%
III	15.00%	15.00%	15.00%	10.00%	7.00%
IV	11.00%	11.00%	11.00%	9.00%	7.00%
V	0.00%	0.00%	0.00%	0.00%	10.00%
Prob>F	0.0114**	0.0114**	0.0114**	0.0024***	0.0012***

***Significant at 1% level   **Significant at 5 % Level   * Significant at 10 % level   ns- not significant

Table II represents Coefficient of co variation and ANOVA of *Khakara* prepared from barley flour, whole wheat flour and sprouted soyabean flour. Variation V has scored least scores in terms of colour, flavour, texture, and taste parameters while Variation III and IV have scored least in terms overall acceptability. Hence Variation V is consistent in colour, flavour, texture and taste and Variations III and IV in overall acceptability. The colour, flavour and texture attribute showed significance at five per cent while taste and overall acceptability were significant at one per cent. Thus the sensory and statistical analysis revealed *Khakhara* prepared from coriander leaves was found to be the most acceptable.

Table 3: Nutrient Analysis of *Khakhra*

Sr. No.	Nutrient Parameter	Value (per 100g)	Method Used
1.	Moisture (%)	2.67	Hot – air oven method
2.	Total Ash (%)	4.49	Muffle furnace method
3.	Energy (Kcals)	402.45	By calculation method
4.	Total protein (g %)	11.55	Micro- Kjeldahl method
5.	Total Fat (g %)	6.28	Soxhlet extraction method
6.	Dietary Fibre (g %)	3.38	Dietary Fibre Assay Kit method (SIGMA)
7.	Total Carbohydrate (g%)	75.01	By Calculation

The nutrient analysis was carried out in triplicates. The nutrients analysed in the cereal – pulse based product, *Khakhara* had 2.67 % moisture indicating a very low moisture food product. The ash content of the product was 4.49%. The product provides approximately 402.45 kCal/100 g. The protein content was 11.55 g while the fat content was about 6.28 g / 100 g. The dietary fibre content was about 3.38 g and the carbohydrate content was 75.01g for each 100 g of the product. The available carbohydrate content (Total carbohydrate- dietary fiber) was 71.63g.

Table 4: Microbial Analysis of *Khakhara*

Product Name	Total Viable Count(cfu/ml)		E- Coli	
No.of days	0 day	II day	0 day	II day
Khakhara	7 X 10^{-7}	7 X 10^{-7}	No growth found	No growth found

Khakhara witnessed a microbial load of 7 colonies at 10^{-7} dilution on the first day while on the third day the number of colonies increased to 10 colonies. No growth of coli forms was found. No fungal growth was observed even after three days of storage. This could be attributed to very low moisture content of the product. This indicates that the

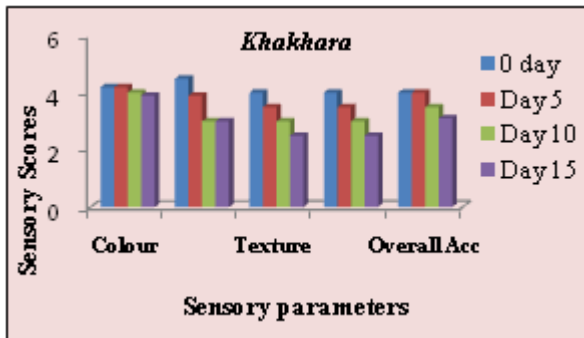product can be stored for more than three days at room temperature.

Figure 5: Changes in the Sensory Parameters During Storage Period

No changes took place in the sensory parameters during the storage period of fifteen days. Thus the product can be stored for a period of fifteen days.

Evaluation of Glycemic Index

The available carbohydrate content was calculated in the prepared product. Definite quantities providing 50 g of available carbohydrate (Total carbohydrate - dietary fiber) was provided to the subjects and the glycemic responses were taken. The total carbohydrate content in *Khakhara* was 71.63g and the serving size of the product served was 70 g.
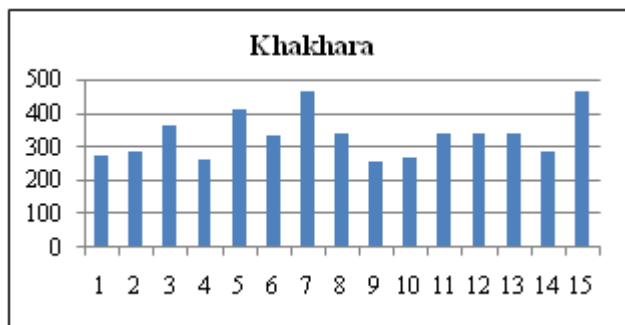

Figure 6: Individual Blood Glucose Responses of Subjects

The number of subjects used in the study was fifteen. Their individual responses have been represented in the above figure. Student t – test was used to statistically analyse the blood glucose response. The p value of **0.899** indicates a significant difference between the blood glucose response values. The mean glycemic response has been represented as follows.
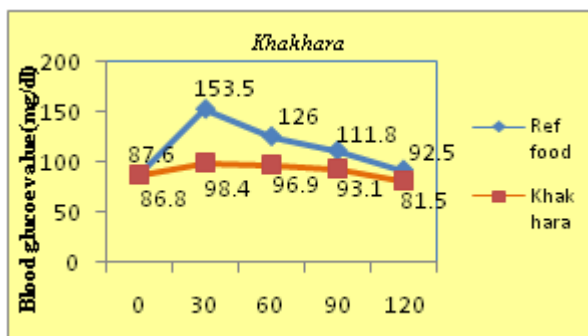

Figure 7: Mean Blood Glucose Responses of *Khakhara*

The Reference food reached a peak (153±39.3) in first 30 minutes after consumption and the peak declined rapidly at 90 minutes. This is because of rapid absorption and utilisation of pure glucose in the body. *Khakhara* showed a blood glucose response of (98.4 ±7.2) in the first 30 minutes upon consumption. No increase was observed in the next 60 minutes. The decrease in the blood glucose levels also took place very gradually.

Different factors can influence blood glucose response. These include the physical form of the food, degree and type of processing, cooking method and time, amount of heat or moisture used (Pi-sunyer, 2002), type of starch (that is, amylose versus amylopectin), and Co ingestion of protein and fat with test foods. (Manders *et al.,* 2005)

The Incremental Area Under The Blood Glucose Response Curve (iAUBGR) value of *Khakhara* was found to be (335.5±67.7) and Glycemic Index value (56.8±15.5). This indicates that *Khakhara* is a low Glycemic index food.

This may be attributed to the low glycemic ingredients namely barley and soya flour used in the preparation of the product. The method of cooking employed was roasting. Roasting involves application of dry heat to cause moisture loss. Although heat is employed but the temperature may not be sufficient enough to cause gelatinisation of starch thus offering resistance to α – amylase digestion. These factors are responsible to render a low GI value to the product. This is in complement with a study done by (Oboh and Erema, 2010) on roasted raw plantain. The roasted product had a low Glycemic Index of 56.87.

4. Conclusion

The product *Khakhara* prepared from barley flour, whole wheat flour and sprouted soyabean flour combined with coriander leaves and other vegetable was found to produce a glycemic index of 56.8 which is a low glycemic index food.

Hence it can be recommended as a snack or breakfast item for diabetics as well the general population in order to overcome the nutritional problems in future.

5. Scope

The scope of this research lies in bringing about more innovations in the development of the product with respect to; incorporation of other underutilised millet flours evaluating its sensory abilities and effect on the glycemic index. Conducting a comparative study on the glycemic index of different Khakhara samples available in the market. Evaluating the effect of glycemic upon consumption of plain Khakhara vs Khakhara consumed with different chutneys.

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