DEVELOPMENT OF LOW GLYCEMIC INDEX FOOD PRODUCTS INCORPORATING SORGHUM, MORINGA OLEIFERA, NIGELLA SATIVA FOR TYPE II DIABETIC PATIENTS

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Abstract
The widespread occurrence of Diabetes Mellitus has presented a dire need for innovative approaches to mitigate and treat this enduring condition. Glycemic Index plays an important role in maintaining blood glucose levels in normal population as well as in Diabetic patients.

The present study aimed to develop low glycemic index food products incorporating Sorghum flour, *Moringa oleifera* leaves powder, *Nigella Sativa* for Type II diabetic patients (age group 30-50 years) and to evaluate the sensory acceptability and nutritional content of the products through proximate analysis. Moringa leaves were collected, washed and shade dried for 24 hours and finally ground to fine powder to be used in the development of Tortilla Chips and Dip. Two variations of Tortilla Chips: Sample T1 (Sorghum, Moringa and Nigella Tortilla Chips) and Sample T2 (Sorghum and Nigella Tortilla Chips) were made. A dip rich in *Moringa* leaves powder was also developed as an accompaniment. A team of 50 trained panelists did the organoleptic evaluation. The statistical analysis of sensory evaluation was carried out using ANOVA and t-test (SPSS Software). The overall acceptability score of T2 was 8.04±1.19 and was more acceptable than T1 at 0.05% significance level. The *Moringa* dip was also accepted with an overall acceptability score of 8.04±1.19. The developed products present a good snacking option and a healthier alternative to processed snacks for Diabetic patients.

Key words: Diabetes Mellitus, Glycemic Index, Sorghum, Moringa oleifera, Nigella sativa, Tortilla Chips.

Introduction
Along with obesity, prevalence of diabetes is increasing in all parts of the world (Anthanont *et al*., 2016: 308-313). Glycemic Index is a list of carbohydrate containing foods based on the increase in blood glucose produced by them. Also, it is the area covered by a blood glucose response curve, on consumption of 50g of available carbohydrate from a sample food. Low GI diets improve glycemic control over and above that obtained by conventional or high-GI diets. There are certain foods that reduce post prandial blood glucose levels such as whole grain foods, millets (Bamosa *et al*., 2010: 344-54; Ruhembe *et al*., 2014: 53). Herbs and spices such as Fenugreek, Cinnamon, Ginseng, *Moringa*, Holy basil, *Nigella* seeds (Dayakar Rao *et al*., 2017: 112; Gopalakrishnan *et al*., 2016: 49-56). Out of the listed food items, *Sorghum*, *Moringa oleifera* leaves and *Nigella sativa* were used in the present study to develop low glycemic index food products and to assess the acceptability of the developed products through sensory evaluation and nutritional content through proximate analysis. The component of sorghum that is favourable for diabetes and high lipid levels in the body is slow digestible starch. It slows down the digestion process and carbohydrate absorption in the body (Mensah *et al*., 2012: 107-112). *Moringa* belongs to the Order Brassicales and Family Moringaceae and is known commonly by many names such as Horseradish tree, Drumstick Tree, Saijihan, Sajna. The fibre in *Moringa* leaves slows down gastric emptying. Polyphenols such as flavanoids and phenolic acids impart anti-diabetic activity to *Moringa* leaves. This is the reason for the decrease in plasma and urine glucose values. A dose of 4g *Moringa* leaves powder increases the secretion of insulin in normal subjects (Brand-Miller *et al*., 2003: 2261-2267; AbuKhader, 2012: 65-68). *Nigella sativa* also known as ‘Kalonji’ belongs to the family Ranunculaceae. *Nigella* seeds increase insulin secretion and slow down the absorption of glucose in the intestines. Therefore, it
forms an essential part of diabetes management and people having glucose intolerance (Vergara-Jimenez and Maria Luz Fernandez, 2017: 91).

**Materials and Methods**

Fig. 1 shows the study was conducted in the following phases:

**Phase 1: Procurement of Ingredients**

The ingredients that were used in the study were sorghum flour, wheat flour, bengal gram flour, *Moringa* leaves powder, *Nigella sativa*, coriander leaves, mint leaves, curd. *Moringa* leaves were collected from a tree in a local village of Uttrakhand.

- **Processing of *Moringa* Leaves:**

  After plucking of leaves, they were washed thoroughly and shade dried for 24 hours at room temperature (Rashmi and Shilpy, 2016: 1-11). Following this, the leaves were ground to a fine powder.

**Phase 2: Development of the products**

- **Development of Tortilla Chips:**
  - Recipe:
    1. Preparation of a soft dough by combining all the ingredients using lukewarm water and oil
    2. Rolling out the dough and pricking it evenly with a fork
    3. Cutting of triangular shapes
    4. Baking of chips in a pre-heated oven for 2 minutes

- **Development of Dip:**
  - Recipe:
    1. Washing and chopping of onions, coriander leaves, mint leaves, ginger, garlic.
    2. Grinding of the above ingredients into a fine paste.
    3. Addition of the paste to hung curd.
    4. Same method is used for *Moringa* dip with subsequent addition of *Moringa* powder also to hung curd.

  Table 1 shows the amounts of all the ingredients along with their carbohydrate content in Tortilla chips. All the samples of tortilla chips were developed so that they contain total of 20±0.5 g of available carbohydrate. (Available carbohydrates =Total Carbohydrate - Fiber).

  Table 2 shows the amounts of all the ingredients along with their carbohydrate content in Dip. Both the samples of dip contained 5-6g of available carbohydrates. In combination, Tortilla chips and dip contain approximately 25g of available carbohydrates and hence, can be treated as a test sample for estimating its Glycemic Index with glucose or white bread as reference foods containing an equal amount of available carbohydrates (25g).

**Phase 3: Sensory evaluation of the developed products**

- **Selection of Panel Members:**

  Selection of panel members involved the screening of 50 subjects from Manav Rachna International Institute Of Research And Studies, Faridabad, Haryana (MRIIRS). 50 trained panelists were selected on the basis of sensitivity threshold test.

  - **Sensitivity threshold test**
    Selection procedure of panelist
    
The test was conducted by preparing four dilution of sugar using 5g, 10g, 15g and 20g in 100 ml of water. The
members were asked to taste the solution and to give
description of taste accordingly. Selection of panel
members involved the screening of 50 trained subjects
from, Manav Rachna International Institute Of Research
And Studies, Faridabad (Haryana)

b. Preparation of composite score cards

Sensory evaluation using composite score card by
50 trained panelists was done in the Food Science
Laboratory, MRIIRS. It was based on taste, color, texture,
flavor, appearance, mouthfeel and aftertaste and overall
acceptability. The panelists were asked to rate the product
on the scale of 10 with 1 being the lowest and 10 being
the highest score.

Phase 4: Proximate analysis of the developed products

Proximate analysis was carried out by Opal Research
& Analytical Services (An ISO certified Laboratory),
Ghaziabad. The parameters which were estimated in the
present study were: Ash, Moisture, Protein, Fat,
Carbohydrates, Crude fibre, Total Phenolic Content and
Total Flavanoid Content.

• Ash Content (AOAC, 2005)

APPARATUS
Crucible, muffle furnace, desiccators.

PROCEDURE
10-15 g of the sample was weighed accurately into a
porcelain crucible (which has previously been heated to
about 600°C and cooled). Then the crucible was placed
in muffle furnace for 3-5 hours at about 60°C. It was
then cooled in desiccators and weighed. This was
repeated till two consecutive weights were same and the
ash was almost white or grayish white in color.

CALCULATIONS
Percentage of ash = \( \frac{W_2 - W_1}{W_1} \times 100 \).
Where,
\( W_1 \) = weight of crucibles (g)
\( W_2 \) = weight of crucible and ash (g)
\( W \) = weight of dry sample.

• Moisture (AOAC, 2005)

APPARATUS
Moisture dish, oven and desiccators

PRINCIPLE
The empty dish and the lid were dried for 15 minutes
in the oven at 100°C. Following which they were
transferred to the desiccators for the purpose of cooling
for 10-20 minutes. 10-15 g of the sample was transferred
to the dish. Then, the lid was replaced and the weight of
the dish with contents was measured quickly to the nearest
milligram. The lid was again removed and weight of the
dish from the oven was measured. After replacing the
lid, the dish was cooled in desiccators and re-weighed.
The dish was dried until the weight was constant.

CALCULATION
Moisture (%) = \( \frac{W_1 - W_2}{W_1 - W} \times 100 \)
Where,
\( W_1 \) = Weight in grams of the dish with the material
before drying
\( W_2 \) = Weight in grams of the dish with the material
after drying
\( W \) = Weight in grams of the empty dish

• Protein (AOAC, 2005)

APPARATUS
Kjehldal flask, volumetric flasks, burette and
condenser.

REAGENTS
Potassium sulphate, copper sulphate, concentrated
sulphuric acid, sodium hydroxide and boric acid.

PROCEDURE
10-15 g of the dried sample was taken in 500 ml

<table>
<thead>
<tr>
<th>INGREDIENTS</th>
<th>SAMPLE T (Standard)</th>
<th>SAMPLE T1 (Moringa Tortilla chips)</th>
<th>SAMPLE T2 (Sorghum and Nigella Tortilla chips)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Amount (Carbohydrate content)</td>
<td>Amount (Carbohydrate content)</td>
<td>Amount (Carbohydrate content)</td>
</tr>
<tr>
<td>Sorghum Flour</td>
<td>-</td>
<td>20 (11.4)</td>
<td>20g (11.4)</td>
</tr>
<tr>
<td>Wheat Flour</td>
<td>25g (13.18)</td>
<td>10g (5.27)</td>
<td>10g (5.27)</td>
</tr>
<tr>
<td>Bengal Gram Flour</td>
<td>18g (7.23)</td>
<td>10g (3.01)</td>
<td>10g (3.01)</td>
</tr>
<tr>
<td>Moringa leaves powder</td>
<td>-</td>
<td>4g (0.76)</td>
<td>-</td>
</tr>
<tr>
<td>Nigella sativa</td>
<td>-</td>
<td>2g (0.20)</td>
<td>2g (0.20)</td>
</tr>
<tr>
<td>Total available carbohydrates</td>
<td>20.41</td>
<td>20.56</td>
<td>19.8</td>
</tr>
</tbody>
</table>

1Alfenas and Paiva, 2007: 197-202; Taweerutchana et al., 2017; Gopalakrishnan et al., 2016: 49-56;
2Gopalakrishnan et al., 2016: 49-56;
kjehldal flask separately to which 1 g of digestion mixture (9.5 g of potassium sulphate and 0.5 g copper sulphate) and 20 ml concentrated sulphuric acid were added, glass buds were introduced to check bumping. The flask was then kept on the electric heater till the digested material becomes colorless or light bluish green in color. The flask was cooled overnight and material was diluted with distilled water to make the volume 250 ml in a volumetric flask 25 ml of aliquot was taken and 80-100 ml of saturated sodium hydroxide was added and was kept on distillation unit, which was connected to a condenser. The lower end of the condenser was dipped in solution of 25 ml of 2% boric acid containing Tashiro’s indicator (methyl red 80 mg, methyl blue 20 mg and methanol 100 ml) in a conical flask. Distillation was carried out for 45 minutes during which all the ammonia releases was tapped in the boric acid. The distillate was titrated in the conical flask against standard N/10 H\textsubscript{2}SO\textsubscript{4} solution taken in a burette till the red color just reappeared. The percent of nitrogen was calculated by the following formula:

\[
\text{Percent Nitrogen} = \frac{M \times 0.0014 \times \text{Dilution factor}}{2 \text{ g of sample taken for digestion}}
\]

The protein content was calculated by using factor 6.25.

- **Carbohydrates**

**PROCEDURE:**

Carbohydrate content of the sample was estimated by subtracting from 100 the sum of the volume (per 100 g) of the moisture, crude protein, crude fat, ash and crude fibre.

**CALCULATIONS:**

Formula for estimating carbohydrate was:

\[
\text{Carbohydrate contents} = 100 - \text{[moisture + crude protein + crude fat + ash (g/100g) + crude fibre content].}
\]

- **Crude Fibre (AOAC, 2005)**

**APPARATUS**

Crucible and muffle furnace

**REAGENTS**

Dilute sulphuric acid, sodium hydroxide solution and ethyl alcohol

**PROCEDURE**

10-15 g of moisture and fat free samples were weighed accurately into a 500-ml beaker and 200 ml boiling 0.25 N (1.25 percent) sulphuric acid added. The mixture was boiled for 30 minutes keeping the volume constant by addition of distilled water at frequent intervals. At the end of this period the mixture was filtered through a muslin cloth and the residue was washed with hot water till free from acid. The material was transferred to the same beaker and 200 ml of boiling 3.313 N (1.25 percent) NaOH was added. After boiling for 30 minutes (keeping the volume constant as before), the mixture was filtered through a muslin cloth and residue was washed with hot water till free alkali followed by a wash with some alcohol. It was then transferred to the crucibles dried overnight at 80-100°C and then weighed (W1). The crucible was heated in a muffle furnace at 600°C for 2-3 hours, cooled and weighed again (W2). The difference in weights (W1-W2) represents the weight of crude fibre, percentage of crude fibre was determined by the following formula:

\[
\text{Crude Fibre} = \frac{(W1 - W2)}{W} \times 100
\]

Where,

\[
W = \text{weight of dried samples (g)}
\]

**Total Phenolic Content**

The method used for estimation of total phenolic content was by using Folin-Ciocalteu (FC) reagent. The extract (0.5 ml) was mixed with 0.5 ml of FC reagent (1:1 diluted with distilled water) and incubated for 5 min at 22°C followed by addition of 2 ml of 20% Na\textsubscript{2}CO\textsubscript{3}. The mixture was then incubated further at 22°C for 90 minutes and the absorbance was measured at 650 nm. The total phenolic content (mg/ml) was calculated using gallic acid as standard (Longvah et al., 2017).

**Total Flavanoid Content**

The method used for estimation of total flavanoid content (mg/ml) was by using aluminum chloride (AlCl3)

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>Sample A (STANDARD)</th>
<th>Sample B (MORINGA DIP)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Amount</td>
<td>Amount</td>
</tr>
<tr>
<td></td>
<td>(Carbohydrate content)</td>
<td>(Carbohydrate content)</td>
</tr>
<tr>
<td>Hung Curd</td>
<td>70g (3.6)</td>
<td>70g (3.6)</td>
</tr>
<tr>
<td>Onion</td>
<td>15g (1.0)</td>
<td>15g (1.0)</td>
</tr>
<tr>
<td>Coriander leaves</td>
<td>8g</td>
<td>4g</td>
</tr>
<tr>
<td>Mint leaves</td>
<td>2g(0.3)</td>
<td>2g(0.3)</td>
</tr>
<tr>
<td>Moringa Leaves powder</td>
<td>-</td>
<td>4g(0.76)\textsuperscript{a}</td>
</tr>
<tr>
<td>Ginger</td>
<td>2.5g</td>
<td>2.5g</td>
</tr>
<tr>
<td>Garlic</td>
<td>2.5g(0.4)</td>
<td>2.5g(0.4)</td>
</tr>
<tr>
<td>Total available carbohydrates</td>
<td>53</td>
<td>60</td>
</tr>
</tbody>
</table>

\textsuperscript{a}Alfenas and Paiva, 2007: 197-202; Taweerutchana et al., 2017
method. The assay mixture consisting of 0.5 ml distilled water and 0.3 ml of 5% NaNO₂ was incubated for 5 minutes at 25°C. This was followed by addition of 0.3 ml of 10% AlCl₃ immediately. Two milliliters of 1M NaOH was then added to the reaction mixture and the absorbance was measured at 510 nm. Quercetin was used as a standard (Longvah et al., 2017).

Phase 5: Statistical Analysis of Data

The statistical analysis of sensory evaluation was done using ANOVA and T-test through SPSS software.

Results

Sensory Evaluation Of Developed Product

Table 3 determines the sensory evaluation of developed product. It tells the mean acceptability score of attributes between Sample T, Sample T1, Sample T2 by composite score test.

Regarding taste, Sample T (standard) (8.06±1.18) was more acceptable than Sample T1 and Sample T2. The difference was significant between Sample T and Sample T1. Sample T2 was more acceptable (8.02±1.15) than Sample T1 with the difference being statistically significant. In terms of color, Sample T was more acceptable (8.32±1.07) than Sample T1 and T2. The difference was statistically significant between Sample T and Sample T1. Out of Sample T1 and T2, Sample T2 was found to be more acceptable (8.06±1.09). In texture, Sample T was found to be more acceptable (8.021.15) than Sample T1 and Sample T2 with a statistically significant difference between Sample T and T1. Regarding Flavor, Sample T was more acceptable (8.1±1.18) than Sample T1 and Sample T2 with the difference being statistically insignificant. In Appearance, Sample T was more acceptable (8.00±1.17) than Sample T1 and Sample T2 but the difference was not statistically significant. In mouthfeel, Sample T was more acceptable (8.021.15) than Sample T1 and Sample T2 but the difference was not statistically significant. Regarding Aftertaste, Sample T was more acceptable (8.021.15) than the others with a statistically significant difference between Sample T and Sample T1. Sample T was more acceptable. In terms of overall acceptability (8.021.15) Out of Sample T1 and Sample T2, Sample T was more acceptable with a statistically significant difference.

Table 4 depicts the sensory evaluation of developed product. It tells the mean acceptability score of attributes between sample A and sample B by composite score test.

Regarding taste, sample A is more acceptable (8.02±1.15) than sample B but the difference was not statistically significant. Regarding color, sample A was more acceptable (8.01±1.15) than sample B and the difference was statistically significant. In terms of texture, sample A was found to be more acceptable (8.01±1.15) than sample B with the difference between the two being statistically significant. In flavor, sample A was found to be more acceptable (8.4±1.29) than sample B but the difference was not statistically significant. Regarding appearance, sample A was found to be more acceptable (8.01±1.15) than sample B and the difference was found to be statistically significant. In mouthfeel, sample A was found to be more acceptable (8.01±1.15) than sample B and the difference was statistically significant. Regarding aftertaste, sample A was more acceptable than sample B and the difference was statistically significant. Regarding overall acceptability, sample A was more acceptable than sample B but the difference was not statistically significant.

The Proximate Analysis of Fresh Moringa Leaves and the Dried Moringa Leaves Powder (100g)

The proximate analysis of fresh Moringa leaves and

<table>
<thead>
<tr>
<th>PARAMETERS</th>
<th>Sample T (Standard)</th>
<th>Sample T1 (Moringa Tortilla Chips)</th>
<th>Sample T2 (Sorghum, Nigella Tortilla Chips)</th>
<th>P-value (at 0.05% significance)</th>
</tr>
</thead>
<tbody>
<tr>
<td>TASTE</td>
<td>8.06±1.18</td>
<td>7.46±1.09a</td>
<td>8.02±1.15b</td>
<td>0.015</td>
</tr>
<tr>
<td>COLOR</td>
<td>8.32±1.07</td>
<td>7.52±1.03a</td>
<td>8.06±1.09b</td>
<td>0.001</td>
</tr>
<tr>
<td>TEXTURE</td>
<td>8.00±1.10</td>
<td>7.42±0.94a</td>
<td>7.90±94</td>
<td>0.019</td>
</tr>
<tr>
<td>FLAVOR</td>
<td>8.1±1.18</td>
<td>7.58±1.21</td>
<td>7.96±1.12</td>
<td>0.076</td>
</tr>
<tr>
<td>APPEARANCE</td>
<td>8.00±1.17</td>
<td>7.52±1.18</td>
<td>7.92±1.08</td>
<td>0.085</td>
</tr>
<tr>
<td>MOUTHFEEL</td>
<td>7.92±1.19</td>
<td>7.48±1.16</td>
<td>7.72±1.10</td>
<td>0.166</td>
</tr>
<tr>
<td>AFTERTASTE</td>
<td>8.00±1.27</td>
<td>7.32±1.25a</td>
<td>7.88±1.11</td>
<td>0.013</td>
</tr>
<tr>
<td>OVERALL ACCEPTABILITY</td>
<td>8.26±1.27</td>
<td>7.42±1.16a</td>
<td>8.04±1.19b</td>
<td>0.002</td>
</tr>
</tbody>
</table>

a denotes a statistically significant difference between Sample T and T1; 
b denotes a statistically significant difference between Sample T1 and T2
the dried Moringa leaves powder shows that since the powder is a concentrated form, there is a substantial increase in all the parameters namely: Energy, protein, fat, carbohydrate, fibre, moisture and ash. There was a 67.29% reduction in moisture content during shade drying. Due to a low moisture content, the powder has a good shelf life. Since ash content is a measure of mineral content, an ash content of 7.8g in dried moringa leaves powder is indicative of it being a rich source of minerals. After the estimation of total phenolic content and total flavonoid content in fresh and dried Moringa leaves powder, the result of the study indicates higher flavonoid content in dried Moringa leaves powder than fresh leaves. The total phenolic content did not increase considerably after shade drying of the leaves.

Test Samples of Tortilla Chips and Dip

Table 5 presents the proximate analysis of the two variants of tortilla chips; Sample T1 and Sample T2. The energy content was higher (417.42 Kcal) in sample T2 (Sorghum, Nigella tortilla chips) than Sample T1. There was a slight difference between the carbohydrate content of both the samples. Protein content was higher (11.13g) in Sample T1 (Moringa tortilla chips) than Sample T2. Fat content was higher (15.7 g) in Sample T2 (Sorghum, Nigella tortilla chips) than Sample T1. Both the samples contain approximately 50g of available carbohydrates per 100g. Moisture content was more in Sample T1 (3.21 g) as compared to 2.91g in Sample T2. There was a slight difference in total ash content between the two samples. The proximate analysis of dip enhanced with Moringa powder. It is a low calorie product (87.5 kcal in 100g of Moringa dip). The dip contains 5.2g of available carbohydrate per 100g.

Discussion

The products developed in the study, Sample T1 (Moringa Tortilla Chips) and Sample T2 (Sorghum, Nigella Tortilla Chips) and Sample B of Dip (Moringa Dip) were well accepted by the trained panelists. The overall acceptability score of T2 was 8.04±1.19 and was more acceptable than T1. The Moringa dip was also accepted with an overall acceptability score of 8.04±1.19. The proximate analysis values of Moringa leaves powder were in agreement with the study conducted by (Gopalakrishnan et al., 2016: 49-56). The powder contains 2.92 mg/ml of total phenolic content and 9.38 mg/ml of total phenolic content through the use of shade drying. These compounds are responsible for the antioxidant capacity and β-cell protection. The developed products present a good snacking option and a healthier alternative to processed snacks for diabetic patients. Further prospects of the study might include the estimation of Glycemic Index of Tortilla Chips and Dip in combination to study the synergistic effect of all the three anti diabetic ingredients. The subjects can be normal healthy individuals or those suffering from Type II Diabetes.

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