Final Report of Minor Research Project

SUPPLEMENTARY EFFECT OF PUMPKIN SEEDS ON MEMORY FUNCTION IN ANIMAL AND HUMAN MODELS

UGC Reference No: MRP(S)-0836/13 -14/

Submitted by

Dr. Sr. Shemi George
Asst. Professor
Department of Home Science
Morning Star College, Angamaly South P.O,
Ernakulam-683573

FINAL REPORT OF A MINOR RESEARCH PROJECT
SUBMITTED TO UNIVERSITY GRANTS COMMISSION. SOUTH WESTERN REGIONAL OFFICE, BANGALORE
# CONTENTS

<table>
<thead>
<tr>
<th>Chapter</th>
<th>Title</th>
<th>Page No</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>INTRODUCTION</td>
<td>1-5</td>
</tr>
<tr>
<td>2</td>
<td>REVIEW OF LITERATURE</td>
<td>6-16</td>
</tr>
<tr>
<td>3</td>
<td>METHODOLOGY</td>
<td>17-29</td>
</tr>
<tr>
<td>4</td>
<td>RESULTS AND DISCUSSION</td>
<td>30-55</td>
</tr>
<tr>
<td>5</td>
<td>SUMMARY AND CONCLUSION</td>
<td>56-58</td>
</tr>
</tbody>
</table>

BIBLIOGRAPHY

APPENDIX
---

**LIST OF TABLES**

<table>
<thead>
<tr>
<th>Sl.No</th>
<th>Title</th>
<th>Page No</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Proximate Composition of pumpkin seed powder</td>
<td>31</td>
</tr>
<tr>
<td>2</td>
<td>Mineral composition of Pumpkin Seeds</td>
<td>31</td>
</tr>
<tr>
<td>3</td>
<td>Amino acid composition of pumpkin seeds powder</td>
<td>32</td>
</tr>
<tr>
<td>4</td>
<td>Compound identified in raw pumpkin seed powder</td>
<td>34</td>
</tr>
<tr>
<td>5</td>
<td>Sensory Evaluation of Developed Product</td>
<td>35</td>
</tr>
<tr>
<td>6</td>
<td>Nutrient Analysis of Developed Products</td>
<td>36</td>
</tr>
<tr>
<td>7</td>
<td>Memory function of pumpkin seeds extract in animal model</td>
<td>38</td>
</tr>
<tr>
<td>8</td>
<td>Socio- Economic Background of Children</td>
<td>39</td>
</tr>
<tr>
<td>9</td>
<td>General Information</td>
<td>40</td>
</tr>
<tr>
<td>10</td>
<td>Anthropometric status of children</td>
<td>41</td>
</tr>
<tr>
<td>11</td>
<td>Food habit</td>
<td>41</td>
</tr>
<tr>
<td>12</td>
<td>Meal pattern per day</td>
<td>42</td>
</tr>
<tr>
<td>13</td>
<td>Skipping of meal</td>
<td>42</td>
</tr>
<tr>
<td>14</td>
<td>Favorite food</td>
<td>43</td>
</tr>
<tr>
<td>15</td>
<td>Dislike food</td>
<td>44</td>
</tr>
<tr>
<td>16</td>
<td>Food intake of children</td>
<td>44</td>
</tr>
<tr>
<td>17</td>
<td>Nutrient intake of children</td>
<td>45</td>
</tr>
<tr>
<td>18</td>
<td>Behavioral Attitude of Children</td>
<td>46</td>
</tr>
<tr>
<td>19</td>
<td>Children behavior attitude in school</td>
<td>46</td>
</tr>
<tr>
<td>20</td>
<td>Emotional behavior of children</td>
<td>49</td>
</tr>
<tr>
<td>21</td>
<td>Stress related event in children</td>
<td>50</td>
</tr>
<tr>
<td>22</td>
<td>Memory test score</td>
<td>53</td>
</tr>
</tbody>
</table>
23  Effect of supplementation on school going children  55

LIST OF FIGURES

<table>
<thead>
<tr>
<th>Figure No</th>
<th>Title</th>
<th>Page No</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>GS-MS chromatogram of pumpkin seeds powder</td>
<td>33</td>
</tr>
<tr>
<td>II</td>
<td>Sensory Evaluation of Developed Product</td>
<td>36</td>
</tr>
<tr>
<td>III</td>
<td>Nutrient Analysis</td>
<td>37</td>
</tr>
<tr>
<td>IV</td>
<td>Behavior attitude of children</td>
<td>47</td>
</tr>
<tr>
<td>V</td>
<td>Children behavior attitude in school</td>
<td>49</td>
</tr>
<tr>
<td>VI</td>
<td>Emotional behavior of children</td>
<td>50</td>
</tr>
<tr>
<td>VII</td>
<td>Stress related event in children</td>
<td>51</td>
</tr>
</tbody>
</table>
## LIST OF PLATES

<table>
<thead>
<tr>
<th>Plate No</th>
<th>Title</th>
<th>Page No</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Developed Products</td>
<td>22</td>
</tr>
<tr>
<td>Ii</td>
<td>Radial Arm Maze</td>
<td>24</td>
</tr>
<tr>
<td>Iii</td>
<td>T-Maze</td>
<td>24</td>
</tr>
<tr>
<td>Iv</td>
<td>Assessment Of Anthropometric Measurement</td>
<td>27</td>
</tr>
</tbody>
</table>

## LIST OF APPENDIX

<table>
<thead>
<tr>
<th>Sl.No</th>
<th>Title</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Questionnaire</td>
</tr>
</tbody>
</table>
1. INTRODUCTION

Let food be your medicine (Hippocrates). A strong memory depends on the health and vitality of your brain. Understanding the physiological basis of behavior is one of the major goals in contemporary neuroscience. Behavior is considered as the outcome of an interaction between inborn and environmental factors. The most important environmental factor in shaping the brain and behavior is learning. Accordingly, unraveling the physiological basis of learning and memory may lead to the neural basis of behavior.

In nature many plants and plants seed provided source of medicine at the earlier times. Plants have proven to be the most useful in curing diseases and provide an important source of pharma medicine. Plants have great significance to the health of individuals. The medicinal importance of these plants lies in some chemical substances that produce a distinct physiological action on the body of human (Anonymous, 2008). Pumpkin seeds are rich in medicinal and nutritive components, due to which reason they are applied in therapeutic purposes across the globe. Food is one of our most basic needs, which provides us energy for everything we do and also for all involuntary functions of our internal organs. All the vast variety of food we eat comes either from plants or animals. Plants provide oil seeds, grains, fruits and vegetables (Srinivasan et al., 2004).

Pumpkin belongs to genus *Cucurbita* of the family *Cucurbitaceae* is one of the largest families of vegetable kingdom. They are widely grown and consumed in many tropical and sub-tropical countries around the world (Juna et al., 2006). Pumpkin seeds are rich in medicinal and nutritive components, due to which reason they are applied in therapeutic purposes across the globe. Food is one of our most basic needs, which provides us energy for everything we do and also for all involuntary functions of our
internal organs. All the vast variety of food we eat comes either from plants or animals. Plants provide oil seeds, grains, fruits and vegetables (Srinivasan et al., 2004).

Pumpkins are grown all around the world for a variety of reasons ranging from agricultural purposes (animal feed) to commercial and ornamental sales. Out of the seven continents only Antarctica is unable to produce pumpkins, the biggest international producers of pumpkins include the United States, Mexico, India, and China. The pumpkin capital of the world is Morton, IL. The traditional American pumpkin is the Connecticut Field variety. Although native to the Western hemisphere, pumpkins are cultivated in North America, continental Europe, Australia, New Zealand, India and some other countries. The pumpkin is the state fruit of New Hampshire (Roberts, Tammy, 2008).

Pumpkin seeds, also known as peptides, are small, flat, green, edible seeds. Most pumpkin seeds are covered by a white husk, although some pumpkin varieties produce seeds without them. Pumpkin seeds have many health benefits, some of which include a good source of protein, zinc and other vitamins, and are even said to lower cholesterol. One gram of pumpkin seed protein contains as much tryptophan as a full glass of milk. The pumpkin seeds contained 39.25% crude protein, 27.83% crude oil, 4.59% ash, and 16.84% crude fiber; the corresponding values for the kernels were 39.22, 43.69, 5.14, and 2.13%, respectively. Pumpkin seed kernels contained moderate concentrations of minerals, especially P, Mg, and K. The amino acid profiles indicate that methionine and tryptophan were the most limiting amino acids, while arginine, glutamic, and aspartic acids were the most plentiful amino acids (http://colleges.ksu.edu.sa/).

Pumpkin seed oil supplementation can prevent changes in plasma lipids and blood pressure associated with inadequate estrogen availability (Gossell-Williams et al., 2008). Moreover, pumpkin seed protein components have anti peroxidative properties and its administration was effective in alleviating the detrimental effects associated with protein malnutrition and carbon tetrachloride (CCl4) intoxication (Nkosi et al., 2006).
Pumpkin (*Cucurbita pepo*) has received considerable attention in recent years because of the nutritional and health protective values of the seeds. The seed is an excellent source of protein and also has pharmacological activities such as anti-diabetic, antifungal, antibacterial, anti-inflammation activities and antioxidant effects (Nkosi *et al*., 2006). Besides, the pumpkin is economical and a nutrient dense source, the pumpkin seed flour fortified complementary food mix is economical, with highly acceptable sensory qualities and a rich nutritive value (Dhiman, 2009).

Pumpkin seeds are rich in oil and the variability in the oil content is very high resulting from a broad genetic diversity. Twelve pumpkin cultivars (*Cucurbita maxima* D.), cultivated in Iowa, were shown to contain oil ranging from 10.9 to 30.9% of high oxidative stability (Stevenson *et al*., 2007). Pumpkin seed oil (PSO) is commonly used in folk medicine. It was shown in several countries that the incidence of hypertension, atherosclerosis, prostatic hypertrophy and urinary bladder hyperplasia was reduced in people regularly consuming the seed oil. Also pumpkin seeds are used locally in Eritrea to treat tapeworm (Harvath, 1988; Schiebel-Schlosser and Friederich, 1998; Zuhair *et al*., 2000, Dreikorn, 2002). PSO is rich in many antioxidants and beneficial nutritional supplements such as essential fatty acids (FAs), vitamins, squalling, carotenoids, tocopherols, phytoestrogens, phytosterols, polyphenols, hydrocarbon, triterpenoids and selenium (Zambo, 1988; Murkovic *et al*., 1996; Fruehwirth and Hermetter, 2007; Gossell-Williams, 2008).

Pumpkin seed is used for cooking or as salad dressing; the oil is generally mixed with other oils because of its robust flavor (Tyler, 2001). It is used for cooking in central and Eastern Europe. It is considered a delicacy in Austria where a little is often added to traditional local cuisine on pumpkins soup or potato salad. In some restaurants in Vienna a few drops are added on vanilla ice cream. It combats benign prostatic hyperplasia because it has beta sitosterol rich in ß-carotene and vitamin C (Mateljjan, 2008). Pumpkin seed oil contains fatty acids which help maintain healthy blood vessels and nerves are loaded with essential fatty acids that help maintain healthy vessels, nerves and tissues (Levin and Rachel, 2008). Sap is used to treat snake bites, hypertension, coronary, stomach, eye and renal disorders.
Pumpkin seeds are rich in nutrients such as vitamin A, vitamin E, zinc, omega-3 and omega-6 fatty acids. The zinc found in pumpkin seeds plays an important role in improving memory and brain function. ([http://www.healthonlinezine.info/top-20-foods-to-improve-memory-brain-function.html](http://www.healthonlinezine.info/top-20-foods-to-improve-memory-brain-function.html))

Pumpkin seeds are actually one of the most nutritious (and delicious) seeds to eat. They are a true brain food containing Omega 3 and Omega 6, making them a smart snack for you and your kids. The Omega 3 fatty acids are known for their role in improving mental health, aiding memory and supporting healthy brain development. The benefits of pumpkin seeds go well beyond just omegas. They contain a high amount of magnesium, which has a calming effect on the brain. ” Pumpkin seeds are high in zinc, which is mega-important for the healthy functioning of the brain, immune system, skin functions and more ([http://inspiyr.com/pumpkin-seeds-good-for-you](http://inspiyr.com/pumpkin-seeds-good-for-you)).

Pumpkin is an excellent source of radish, an antioxidant that helps in keeping a healthy brain. Dutch studies found that more the daily dietary intake of food rich in radish more critical thinking can be maintained. Pumpkin seeds contain zinc which is also an important substance for brain functioning. Insufficient zinc is prone to memory loss and lack of attention. In University of Texas, a Ph.D. pointed out that even in healthy people lack a small amount of zinc can cause short-term memory to decline. As long as there is supplement of zinc, for example, women's vocabulary memory will improve by 12%. ([http://www.philcheung.com/Health/MYBS_e.htm](http://www.philcheung.com/Health/MYBS_e.htm))

Pumpkin seeds are rich in protein, magnesium and manganese, but zinc is the first important ingredient when it comes to improving your memory. A study conducted at Duke University Medical Center in North Carolina, published in the medical journal Neuron, found zinc helps the neurons within the hippocampus communicate. The other important component of pumpkin seed oil for memory is essential omega-3 fatty acids. Omega-3 is known to boost our cognitive function, and better cognitive function means improved concentration and less memory loss. A study funded in part by the National Center for Complementary and Alternative Medicine (NAACM) and published in Neurology, examined 104 adults, aged 65 years and older. Researchers found those with higher concentrations of omega-3 in their blood nutrient
Recently increased attention has been focused on underutilized products as food and feed. Pumpkin seed is an underutilized product with high nutritious and medical property. Due to many reason memory capacity has been decreased. Pumpkin seed can increase the memory power in school going children. With this in view; the present study is undertaken with the following aim and objectives;

Children with working memory impairments often fail in the classroom because the working memory loads are excessive for them. Working memory failure leads to inattentive behavior, simply because the child forgets what s/he is doing. Poor memory leads to poor academic progress, particularly in reading and math’s, normal social integration, reserved in groups, difficulties in following instructions, problems combining processing with storage, place-keeping difficulties, short attention span and distractibility. So the study of the memory problems and supplementation of the memory enriched food is very relevant.

Objectives

1. To analyze the nutrient compounds in pumpkin seeds
2. To study the memory Function of pumpkin seeds extract in animal models
3. To investigate the nutritional status of the selected school going children
4. To assess the supplementary effect of pumpkin seeds for increasing memory in selected school going children
2. REVIEW OF LITERATURE

The literature pertaining to the study entitled “Supplementary Effect of Pumpkin Seeds on Memory Function in Animal and Human Models” is reviewed under the following headings.

2.1. Pumpkin – General aspects

2.2. Constituents of the pumpkin seeds

2.3. Health benefit of pumpkin seeds

2.4. Memory function of pumpkin seeds

2.5. Pumpkin seed for brain development

2.6. Pumpkin seed for brain development in children

2.1 Pumpkin- General aspects

Classification of pumpkin includes;

\[\begin{align*}
\text{Family} & \quad : \quad \text{Cucurbitaceae} \\
\text{Botanical Name} & \quad : \quad \text{Cucurbita Maxima} \\
\text{Kingdom} & \quad : \quad \text{Plantae} \\
\text{Order} & \quad : \quad \text{Cucurbitales} \\
\text{Family} & \quad : \quad \text{Cucurbitaceae} \\
\text{Genus} & \quad : \quad \text{Cucurbita} \\
\text{Colour} & \quad : \quad \text{Light yellow-orange to bright orange} \\
\text{Best season} & \quad : \quad \text{Throughout the year}
\end{align*}\]

Pumpkin (English), kumbra (Bengali), kohul (Gujarati), kaddu (Hindi), kumbala (kannada), paarimal (kashmiri) mathan or chakkara kumbalanga (Malayalam), Lal Bhopal (Marathi), kakharu (oriya), sitaphal (Punjabi), purangikkai or pooshanik (tamil), gummadi kayi (telugu) dungaree (Sanskrit) (gopalan et al., 2011).

Pumpkin belongs to the family Cucurbitaceae which is an angiosperm, genus Cucurbita with different varieties (Alfawaz, 2004). Normally pumpkin seeds are thrown away as a waste, whereas it is a rich source of nutrients and oil and can be used as a food
After the elimination of pulp, pumpkin seeds and rinds which remain in large quantities as waste product could be consumed for edible purpose (Abd El-Aziz and El-Kalek, 2011). The antimicrobial activity of pumpkin has many applications, including preservation, pharmaceuticals, alternative medicine and natural therapies (Rajakaruna et al., 2002).

Pumpkin seeds indeed are an excellent source of dietary fiber and mono-unsaturated fatty acids, which are good for heart health. In addition, the seeds are concentrated sources of protein, minerals and health-benefiting vitamins. For instance, 100 g of pumpkin seeds provide 559 calories, 30 g of protein, 110% RDA of iron, 4987 mg of niacin (31% RDA), selenium (17% of RDA), zinc (71%) etc., but no cholesterol. Further, the seeds are an excellent source of health promoting amino acid tryptophan. Tryptophan is converted to GABA in the brain. (http://www.nutrition-and-you.com/pumpkin.html).

2.2 Constituents of pumpkin seeds

Nuts are nutrient dense foods. They contain high amounts of protein and fat, mostly unsaturated fatty acids. Nuts are also dense in a variety of other nutrients and provide dietary fiber, vitamins (e.g. folic acid, niacin, vitamin E, vitamin B6), minerals (e.g. copper, magnesium, potassium, zinc) and many bio-active constituents such as antioxidants, phytosterols and other phytochemicals (Dreher et al. 1996; USDA, 2006).

Pumpkin seeds contained relatively large amounts of magnesium Mg (5690); zinc Zn (113); copper Cu (15.4); molybdenum Mo (0.805) and another minerals: phosphorus P (15700); calcium Ca (346); iron Fe (106); manganese Mn (49.3); aluminum Al (9.21); barium (1.16); cobalt Co (0.29); strontium Sr (1.83); nickel Ni (0.53); arsenic (0.45) (in μg/g dry weight). Noteworthy are the low amounts of calcium in the seeds (Glew et al. 2006).

Pumpkin seeds are an excellent natural source of essential vitamins and minerals healthy oils and fiber. Boiled, baked or even raw, pumpkin seeds are packed full of vitamins, minerals and amino acid. Just one serving (about 1/4cup) pumpkin seed gives you almost half recommended daily amounts of manganese, magnesium, phosphorus, iron, copper, vitamin K and zinc. Pumpkin seeds are a good source of protein and Monounsaturated
fats([http://www.squidoo.com/pepitas](http://www.squidoo.com/pepitas)).

### 2.2.1 Chemical composition of pumpkin seed

Pumpkin active ingredients include essential fatty acids, amino acids, phytosterols (including beta-sitosterol), minerals and vitamins. Seed kernels of pumpkin are rich in fat soluble vitamins, oil and protein, containing considerable amounts of P, K, Mg, Mn, and Ca. Roasted pumpkin contains a significantly high amount of carotenoids; especially zeaxanthin. Pumpkin seed kernel flour has high values of chemical score, essential amino acid index, and in vitro protein digestibility. Pumpkin Seed Oil has high amounts of unsaturated fatty acids with linoleic and oleic acids as the major acids. Pumpkin possess an unusual amino acid known as cucurbitin has been chemically defined as (-)-3-amino-3-carboxy pyrimidine ([The Columbia E encyclopedia. 2004](http://www.columbia.edu/)).

The pumpkin seeds contained 39.25% crude protein, 27.83% crude oil, 4.59% ash, and 16.84% crude fiber; the corresponding values for the kernels were 39.22, 43.69, 5.14, and 2.13%, respectively. Pumpkin seed kernels contained moderate concentrations of minerals, especially P, Mg, and K. The amino acid profiles indicate that methionine and tryptophan were the most limiting amino acids, while arginine, glutamic, and aspartic acids were the most plentiful amino acids. The saturated fatty acid content was 27.73% and comprises of 16.41% palmitic acid and 11.14% stearic acid. The unsaturated fatty acid value was 73.03% and consisting mainly of 18.14% oleic acid and 52.69% linoleic acid. The oil obtained from the pumpkin seed kernels had a refractive index of 1.4656, specific gravity of 0.913, iodine value of 105.12 (g I$_2$/100g oil) saponification value of 185.20 (mg KOH/g oil), acid value of 0.53 (mg KOH/g oil), and peroxide value of 0.85 (meq peroxide/kg oil). Considering the lipid and protein content in the kernels, and their fatty and amino acid compositions, the pumpkin seed kernels appear to be quite promising for commercial exploitation (Mohammed A. Alfawaz, 2004).

Pumpkin flour was produced from mature pumpkin contain 6.01% moisture, 3.74% protein, 1.34% fat, 7.24% ash, 2.9% fiber, 78.77% carbohydrate and 56.04% alcohol insoluble solids. The sample has 7.29 mg/100 g beta-carotene with color values of L* 57.81, a* 8.31, b* 34.39 and 0.24 water activity (Pongjanta et al., 2006).
The nutritional value of pumpkin seeds is based on high protein content and high energy potential due to high percentage of oil. Oil content in the seeds is 40%–60%; 98%–99% of the oil is from the fatty acids oleic (up to 46.9%), linolenic (up to 60.8%), palmitic (up to 14.5%), and stearic (up to 7.4%), with a ratio of monounsaturated to polyunsaturated acids from 0.60 to 0.75 (Markova et al. 1996, Nakić et al. 2006). Because of its high protein content (61.4%±2.6%), and certain functional and electrophoretic properties (Lazos, 1992), pumpkin seed flour obtained after oil pressing might be a potential food (Mansour et al. 1993).

2.2.2. Fatty acid composition

The pumpkin seeds contained, saturated fatty acid content was 27.73% and Comprises of 16.41% palmitic acid and 11.14% stearic acid. The unsaturated fatty acid value was 73.03% and consisting mainly of 18.14% oleic acid and 52.69% linoleic acid (http://colleges.ksu.edu.sa/).

Pumpkin seed oil is comprised of mostly unsaturated fatty acids, primarily linoleic acid, with lesser amounts of oleic acid (Fruhwirth & Hermetter, 2007). Pumpkin seed contains fatty acids were oleic (up to 46.9%), lenolenic (up to 60.8%), palmitic (up to 14.5%), and stearic (up to 7.4%), with a ratio of monounsaturated to polyunsaturated acids from 0.60 to 0.75 (Nakic et al. 2006).

The fatty acid composition of the crude lipid fraction of seeds of Cucurbita spp. Nigerian origin with four fatty acids accounting for >97% of the fatty acid total: palmitic acid (C 16:0; 13.0%), stearic acid (C 18:0; 7.9%), oleic acid (C 18: 1 n-9; 45.4%) and the essential fatty acid linoleic acid (C 18:2n-6; 31%). On a percentage basis, oleic acid was the predominant fatty acid. Whereas linoleic acid accounts for nearly one-third of the total fatty acid in pumpkin seeds, only a trace amounts of α- and γ- linolenic acids were found (Glew et al. 2006).

The glyceride fraction contains over 80% unsaturated fatty acids, mainly linoleic (C18:2; 42-64%) and oleic (C18:1; 20-38%) acids, and 19% saturated fatty acids, mainly palmitic (C16:0; ca. 13%) and stearic acids (C18:0; ca. 6%) (Andrikopoulos et al. 2004, Schilcher 1986; Murkovic et al. 1996a; Vogel 1978; Tsaknis et al. 1997; Bombardelli and Morazzoni 1997).
Typical pumpkin seed oil is comprised of mostly unsaturated fatty acids, primarily linoleic acid, with lesser amounts of oleic acid. Its fatty acid profile is similar to that of soybean oil with the exception of linolenic acid, which is very low in pumpkin seed oil (Table 1). (Fruhwirth & Hermetter, 2007; Murkovic, Piironen, Lampi, Kraushofer, Sontag, 2004; Murkovic & Pfannhaus, 2000).

Unsaturated fats including monounsaturated and polyunsaturated fatty acids are recommended for a healthy daily diet rather than the consumption of saturated fats. Pumpkin seed oil contains high amounts of unsaturated fats including Omega 3 (linolenic) and Omega 6 (linoleic) essential fatty acids (Murkovic, & Pfannhauser, 2000).

2.2.3. Amino acid composition
In pumpkin pulp the amount of protein is negligible – on average 0.8–1.0% (Anon, 1976), but they contain almost all the essential and nonessential amino acids (Duke, 1998).

Rare amino acids have also been found in Cucurbita pepo seeds: m-carboxyphenylalanine, β-pyrazolalanine, γ-aminobutyric acid, ethyl-asparagine and citrulline. Particular attention has been paid to cucurbitine (3-amino-3-carboxypyrrololidine for its anthelmintic properties isolated from Cucurbita pepo (0.18-0.66%) and Cucurbita moschata Duch, 0.4-0.84%) seeds. Cucurbitin content varied even within the same species (Blagrove and Lilley 1980; Bombardelli and Morazzoni 1997; Bradley 2006; Bruneton 1995; Huang 1998; Mihranian and Abou-Chaar 1968, Rybałtowski 1966).

According to Mansour et al. (1993) the dehulled and defatted pumpkin seed (Cucurbita pepo Kakai 35), has an excellent pattern of amino acids, contain high levels of most essential amino acids including: isoleucine 2.66; leucine 6.13; lysine 5.20; cystine 1.52; methionine 1.25; tyrosine 2.94; phenylalanine 4.00; threonine 2.75; tryptophan 1.56; valine 3.40; histidine 3.62; and non-essential amino acids: arginine 16.70; aspartic acid 10.19; glutamic acid 18.13; serine 5.46; proline 4.34; glycine 5.86; alanine 4.29 (in g per 16 g N).

2.2.4. Minerals
The pumpkin seeds contained relatively large amounts of potassium (5790 μg/g dry weight) and chromium (approx.3 μg/g dry weight). However, the sodium content of pumpkin seeds was low (6.9 μg/g dry weight). Pumpkin seeds contained relatively large
amounts of magnesium Mg (5690); zinc Zn (113); copper Cu (15.4); molybdenum Mo (0.805) and another minerals: phosphorus P (15700); calcium Ca (346); iron Fe (106); manganese Mn (49.3); aluminum Al (9.21); barium (1.16); cobalt Co (0.29); strontium Sr (1.83); nickel Ni (0.53); arsenic (0.45) (in μg/g dry weight). Noteworthy are the low amounts of calcium in the seeds (Glew et al. 2006).

Particularly phosphorus, potassium, magnesium, calcium, iron, zinc and trace elements (Mansour et al. 1993). Selenium is of particular importance as its content ranges between 0.08 and 0.4 μg/g, one of the highest values found in plants (Bombardelli and Morazzoni 1997; Kreft et al. 2002). Other sources are reporting even higher amount of selenium: 1.29g/g (Glew et al., 2006). Mean estimated maximum intake: 0.4 mg/day and safe upper level was estimated for 0.45 mg total selenium/day for daily consumption over a lifetime (Scientific Opinion 2009).

Pumpkin seeds contain a good dose of zinc which is said to increase the sperm count they also contain essential fatty acids like Omega-3 which enhance the blood flow to sexual organs.

2.3. Health benefits of pumpkin seed

2.3.1. Prostate Cancer

The protective compounds present within the pumpkin seeds, called phytosterols can lower the risk of prostate cancer. These work by shrinking the prostate and stimulating the secretion of chemicals that protect against the transformation of testosterone into dihydrotestosterone (DHT). High DHT levels can cause enlargement of the prostate glands (http://harvestcoop.files.wordpress.com/2011/11/pumpkin-health-benefits.pdf).

2.3.2. Anxiety Relief

A study published in the Canadian Journal of Physiology and Pharmacology revealed that tryptophan, an amino acid abundant in pumpkin seeds, might help alleviate anxiety. Since tryptophan is converted to serotonin, a neurotransmitter that enhances mood and promotes well-being in the brain, researchers investigated whether consuming a tryptophan rich food could boost serotonin levels and reduce anxiety
symptoms. They discovered that subjects with anxiety disorder who consumed tryptophan rich gourd seeds with carbohydrates before an anxiety test experienced greater improvements in subjective and objective measures on the Endler Multidimensional Anxiety Scale compared with those who consumed only carbohydrates (J Physiol Pharmacol, 2007).

2.3.3. Diabetes prevention

Most of the evidence we've seen about pumpkin seeds and prevention or treatment of diabetes has come from animal studies. For this reason, we consider research in this area to be preliminary. However, recent studies on laboratory animals have shown the ability of ground pumpkin seeds, pumpkin seed extracts, and pumpkin seed oil to improve insulin regulation in diabetic animals and to prevent some unwanted consequences of diabetes on kidney function. Decrease in oxidative stress has played a key role in many studies that show benefits of pumpkin seeds for diabetic animals.

2.3.4. Protection for Men's Bones

In addition to maintaining prostate health, another reason for older men to make zinc-rich foods, such as pumpkin seeds, a regular part of their healthy way of eating is bone mineral density. Although osteoporosis is often thought to be a disease for which postmenopausal women are at highest risk, it is also a potential problem for older men. Almost 30% of hip fractures occur in men, and 1 in 8 men over age 50 will have an osteoporotic fracture. (http://www.balancedconcepts.net/Pumpkin_Seeds_Pros.pdf)

2.4. Pumpkin seed for brain development

Raw pumpkin seed oil contains healthy omega-6 and omega-9 fatty acids, phytosterols, and vitamins E and K. The linoleic acid contained in pumpkin seed oil promotes healthy brain function as well as skin suppleness. The oleic acid in pumpkin seed oil lowers "bad" cholesterol, which promotes heart and liver health. (Yadav M, et al., 2010).
Pumpkin seeds are extremely rich in nutrients, especially zinc, which makes them a wonderful immune system protector, especially for men who want to ensure a healthy prostate. ([http://www.dailymail.co.uk/health/article-435/Pumpkin-seeds.html](http://www.dailymail.co.uk/health/article-435/Pumpkin-seeds.html))

Zinc is a trace mineral that plays a central role in cellular growth, specifically in the production of enzymes necessary for the synthesis of RNA and DNA. Zinc is prevalent in the brain, where it binds with proteins, thus contributing to both the structure and function of the brain. (Bae YS, Hill ND, Bibi Y, et al.).

Pumpkin seeds have long been valued as a source of the mineral zinc, and the World Health Organization recommends their consumption as a good way of obtaining this nutrient. If you want to maximize the amount of zinc that you will be getting from your pumpkin seeds, we recommend that you consider purchasing them in unshelled form. ([http://www.whfoods.com/](http://www.whfoods.com/))

Magnesium is involved in biochemical reactions that help keep bones strong and promote healthy heart rhythms and nervous system function. ([http://www.encognitive.com/files/Pumpkin%20seed%20power.pdf](http://www.encognitive.com/files/Pumpkin%20seed%20power.pdf))

**2.5. Pumpkin seed for brain development in children**

Pumpkin seeds are also useful to treat children affected with learning disabilities. Many consider these seeds as a brain food due to its high magnesium content. Magnesium deficiency can slow down brain function, which may lead to learning problems. These seeds being a rich source of magnesium have a positive impact on the brain, which may help to improve learning disorders. ([http://www.buzzle.com/articles/pumpkin-seeds-benefits.html](http://www.buzzle.com/articles/pumpkin-seeds-benefits.html))

Pumpkin seeds are actually one of the most nutritious (and delicious) seeds to eat. They are a true brain food containing Omega 3 and Omega 6, making them a smart snack for you and your kids. The Omega 3 fatty acids are known for their role in improving mental health, aiding memory and supporting healthy brain development. Pumpkin seeds
are high in zinc, which is mega-important for the healthy functioning of the brain, immune system, skin functions and more in children (http://inspiyr.com/pumpkin-seeds-5-reasons-add-diet/).

Manganese plays an important role in all mental functions and aids in the transfer of oxygen from lungs to cells, it is important as an activator for enzyme reactions concerned with carbohydrate, fat and protein metabolism (Ahmed, U. and Birnin-Yauri, U. A. (2008)).

In a study of students with low working memory capacity, Michael Kane, at University of North Carolina, found that these students more often spent their time off task and daydreaming, especially when the task at hand was challenging. However, student with relatively high working memory kept their attention on the task, even when it was challenging. The Learning Brain , Working memory, brain training, and helping develop the minds of children, Torkel Klingberg, M.D., Ph.D, 2012. Significant improvement in blood levels of micronutrients has been reported in Indian schoolchildren after supplementation of a micronutrient-enriched beverage [Sivakumar, et al., Effect of micronutrient supplement on health and nutritional status of schoolchildren: biochemical status. Nutrition 22 (1 suppl): S15–S25, 2006.] or a food supplement (containing ferrous sulfate, calcium pantothenate, and vitamins) [Vinod Kumar M, Rajagopalan S, Impact of a multiple-micronutrient food supplement on the nutritional status of schoolchildren. although its effect on cognitive function was marginal and selective in the earlier study and significant in the latter. However, the effect of natural dietary modification on the micronutrient status and health of schoolchildren has been reported scarcely. Researchers from The Ohio State University extended summer learning gap research further by conducting a national study of 17,000 kindergarten and first grade children from the Early Childhood Longitudinal Study. The authors confirmed earlier findings of an unequal starting point, showing that a standard deviation’s advantage in SES predicts a 1.77 month advantage in initial reading skill on the first day of kindergarten (Downey, D., von Hippel, P., & Broh, B. 2004). The authors also confirmed that the SES achievement gap continues to grow after schooling starts, with summer learning accounting for the vast majority of the difference. While the average kindergarten
learning rate was 1.65 test points per month, a standard deviation’s advantage in SES predicted a relative gain of 0.16 points per month during summer, 0.07 points per month during kindergarten, and 0.05 points per month during first grade.

2.5.1. National status

The importance of education in India is indeed rising with passing time. Though India has always been a great source of learning for many years, it still needs to improve not just on the quality of education but also on the number of people being educated. In India, still many are deprived of education mainly due to poverty and less accessibility educational services. The lack of education, adds to the vulnerability of children for forcing them into social evils of child labour and crime. The loss in learning varies across grade level, subject matter, and family income. A common finding across numerous studies is that on average, students score lower on standardized tests at the end of the summer than they do at the beginning of summer (on the same test). In India the Mathematics - 2.6 months of grade-level equivalency loss, Reading- Varies across SES. Low income students generally lose about 2 months of reading achievement.

2.5.2. International status

International studies revealed that the loss was most acute in factual and procedural learning such as mathematical computation, where an average setback of more than two months of grade-level equivalency was observed among both middle- and lower-class students. In reading and language, however, substantial differences were found between middle- and lower-class students. Whereas middle-class students showed no significant gain in reading scores, lower-class students showed a significant loss that represented a gap of about three months of grade-level equivalent reading skills between middle- and lower-class students. These results are consistent with other researcher's findings that a family’s socioeconomic status affects children’s achievement scores almost exclusively when school is closed. Barbara Heyns’ 1978 landmark study of 2,978 6th and 7th graders in the Atlanta city public schools was the first thorough investigation of summer learning (.Heyns found that while poor children and black children came close
to keeping up with middle-class children in cognitive growth when school was in session, they lagged far behind during the summer.

Researchers Doris Entwistle and Karl Alexander extended Barbara Heyns’ line of research through the Beginning School Study (BSS) in 1982. BSS compared the school-year and summer achievement gains of 790 youth across 20 of Baltimore’s public schools from the beginning of first grade in 1982 through the end of elementary school. The study also tracked these students’ progress through high school and college. They found that in year nine, the low-socioeconomic status (SES) group’s Reading Comprehension average lagged 73 points behind the high-SES group’s on the California Achievement Test (CAT-V)( Alexander, Karl, Entwisle et al. 2004). About a third of the 73 points difference (27 points) was in place when the students started first grade. After the first grade, the low-SES students fall farther behind each year, with the gap reaching a plateau of around 70 points in the 5th grade. The remaining two-thirds of the 73 point gap accumulate over the course of the elementary and middle school years, with a staggering 48.5 points being attributed to the cumulative summer learning gap from the five elementary years.
3. METHODOLOGY

Methodology is a way to systematically solve the research problem. It may be understood as a science of studying how research is done. Scientifically in it we study the various steps that are generally adopted by a researcher in studying his research problem along with logic behind them (kothari2005).

Phase 1

3.1. Determination of Chemical Properties of Pumpkin Seeds
   3.1.1. Determination of proximate composition
   3.1.2. Determination of Mineral Composition of Pumpkin Seed
   3.1.3. Amino acid and of Pumpkin Seed
   3.1.4. Fatty acid profile of Pumpkin Seed

Phase 2

3.2. Product Development Using Pumpkin Seed
   3.2.1. Preparation of Product
   3.2.2. Sensory Evaluation of Developed Product
   3.2.3. Nutrient Analysis of Developed Product
   3.2.4. Selection of Developed Product

Phase 3

3.3. To study the memory Function of pumpkin seeds extract in animal models
3.4. To study the nutritional status of the selected school going children
   3.4.1. Selection of Area
   3.4.2. Selection of Subject
   3.4.3. Tools Selected for Data Collection
3.5. To study the memory Function of pumpkin seeds extract in human models
3.1. Determination of chemical properties of pumpkin seeds

   Chemical characteristics like proximate composition, mineral composition, fatty acid profile of the seed powder was done.
3.1.1 Proximate composition of pumpkin seeds

3.1.1.1. Moisture

A known amount of sample was weighed into a previously weighed moisture cup and dried in an oven at 60% to a constant weight (Anon, 1990) and moisture content calculated as follows:

\[
\text{Moisture} \% = \frac{\text{weight of sample before drying} - \text{weight of sample after drying}}{\text{Sample weight}}
\]

3.1.1.2. Crude protein

The nitrogen content of the processed pumpkin seeds powder was assessed by the Kjeldahl method using Pelican Kelpplus equipment. Crude protein was calculated by multiplying the titer value with a factor 6.25 (Monteiro et al., 1988).

\[
\text{Protein} \% = \frac{\text{Titer value} - \text{Blank} \times \text{No. of HCl} \times 14.007 \times 6.25}{\text{Sample weight} (g)}
\]

3.1.1.3. Crude fiber

Crude fiber was estimated from the moisture and fat-free sample. The residue obtained after digestion with acids and alkali was dried in a crucible and weighed. The difference in weight of the crucible before and after the ashing of digested residue was taken as weight of the crude fiber (Anon, 2002).

3.1.1.4 Carbohydrate content

The total carbohydrate content was determined (Egan et al., 1981), by the formula; Carbohydrate \( \% \) = 100-(% moisture + % proteins + % lipids + % ash)

3.1.1.5 Energy value

The energy value was calculated using the Atwater factor method \([9 \times \text{fat})] + (4 \times \text{carbohydrate}) + (4 \times \text{protein})\) as described by Osherine and Voogt (1978); Enahe (1991); Chinma and Agyor (2007). The proportion of protein, fat, and cho
were multiplied by their physiological fuel value of 4, 9 and 4 kcal, respectively and sum of the product was taken (Nwabaeze, 2007).

3.1.1.6. Nitrogen free extract

The NFE was calculated according to the following expression

$$\text{NFE} = 100 - (\% \text{ moisture} + \% \text{ crude protein} + \% \text{ crude fat} + \% \text{ ash})$$

3.1.2. Determination of Mineral Composition

Using the method describe by AOAC (2005) the ash of each sample was digested with 5 ml of 2 m HNO3 and heated to dryness on heating mantle. 5 ml of 2 m HON3 and added to it and heated to boil and filtered through a whatman no.1 filter paper into a 100 ml volumetric flask. The filtrate was determined using jenway digested flame photometer using (PFP7 modle) while other minerals apart from phosphorus were determined using an amino acid analyser according to the method of Schuster (1988). The sample were hydrolysed with 6N HCL under vaccum at 110°C for 24 h. The hydrolysates were dried in a rotary evaporator at 40°C under vaccum to remove the excess acid (6N HCL) the dry residue were then dissolved in a known quantity of and filtered to obtained a clean solution of the hydrolysate. An aliquot of it was injected into column (shim-pack ISC-07 IS 1504) of the HPLC-based amino acid analyser CRF-10AXL, Shimadzu corporation, Tokyo, Japan) equipped with fluorescence detector (FLD-6A) sodium hypochlorite and ophlha solution were used as reaction solution.

3.1.3. Amino acid composition

All the samples of processed pumpkin seeds were subjected to determination of amino acids composition using an amino acid analyzer according to the method of Schuster (1988). The samples were hydrolyzed with 6N HCl under vacuum at 110°C for 24 h. The hydrolysates were dried in a rotary evaporator at 40°C under vacuum to remove the excess acid (6N HCl). The dry residues were then dissolved in a known quantity of citrate buffer (2.2pH) and filtered to obtain a clean solution of the hydrolysate. An aliquot of it was injected into the column (Shim-pack ISC-07/S1504 Na) of the HPLC. Based amino acid analyzer (RF-10AXL, Shimadzu Corporation, Tokyo, Japan) Equipped with
fluorescence detector (FLD-6A). Sodium hypochlorite and ophthalaldehyde solutions were used as reaction solutions.

3.1.4. Fatty acids profile analysis by Gas Chromatography-Mass spectrometry (GC-MS)

GC-MS analysis of the fatty acid was carried out after methylation. GC-MS analysis was performed with GC Clarus 500 Perkin Elmer equipment. Compounds were separated on Elite-5MS capillary column (5% diphenyl /95% Dimethyl poly siloxane), 30× 0.25mm×0.25μm df). Oven temperature was programmed as follows: isothermal temperature at 110ºC for 2 min., later increased to 200ºC at the rate of 10ºC/minutes and then increased up to 280ºC at the rate of 5ºC/min and held for 9 min. Ionization of the sample components was performed in the EI mode (70eV). The carrier gas flow rate was 1ml/min, and 3 μl of sample was injected. The detector was Mass detector turbo mass gold-Perkin Elmer. The total running time for GC was 36 min. and software Turbo mass 5.2 was used in this GC-MS study.

The determination of the components was done by comparing their retention time with those of authentic specimens on the capillary column as well as peak enrichment.

Phase2

3.2. Preparation of Products using pumpkin seeds

3.2.1 Preparation of product

Sun dried pumpkin seed was powdered and this flour was used for developed product.

3.2.1.1 Preparation of bread

The composition of bread was 70g whole wheat flour, 40g pumpkin seed, 3.1 g yeast, 14 g salt, 30g butter, 1 cup water. The proportion was based on standard procedure. This is the flow chart of bread preparation.
Mixing the whole wheat with salt and add in the instant yeast

↓

Add Butter and pumpkinseed

↓

Mix well and adding water in intervals

↓

Kept in oven for 220 degrees C for 22-25 mins.

3.2.1.2. Preparation of cake

The composition of cake was pumpkin seed powder 40g, 3 Eggs, 20 g sugar, 30 g flour, 10g butter, Vanilla extract. The proportion was based on standard procedure. This is the flow chart of bread preparation.

Mix the egg yolks

↓

Add the pumpkin seed powder, vanilla extract and sugar

↓

Mix the flour

↓

Bake in oven for 40 minutes at 375 degrees F.

3.2.1.3. Preparation of nutrient ball

The composition of nutrient ball was pumpkinseed powder 40g, roasted rice flour 50g, 10g jaggery.

Roasted the rice flour

↓

Add pumpkin seed powder and mix well.

↓

Add jaggery in to it.
3.2.2. Sensory evaluation of developed product

Sensory evaluation of products was done by hedonic scale. The hedonic 9-point scale is a useful tool to have for any examiner of food preference or overall
liking of food. The scale is easy for the panelist to understand and use. The scale is self-explanatory with little instructions from the moderator of the test (Lawless and Heymann 1998). Sample selected for evaluation were at random from.

3.2.3. Nutrient analysis of developed product

The nutrient like energy, magnesium and zinc are calculated based on Nutritive Value of Indian Foods.

3.2.4. Selection of developed product

Select best product based on analyzing nutritive value and sensory evaluation.

3.3. To study the memory Function of pumpkin seeds extract in animal models

3.3.1. Experimental Animals and Design

The 40 male waster rats weighing between 120 to 140g body weight is selected for evaluate the supplementary effect of pumpkin seeds extract on memory functions of the brain in animals. Each experimental animal receive (100 mg/kg) pumpkin seeds extract daily for 30 days. Control group didn’t receive any supplements. All the behavioural experiments were carried out in three phases viz; orientation and training session, learning performance test (acquisition test) and memory performance test (retention test).

3.3.2. Tool selected for the study

The two tools selected for analyze the supplementary effect in animal model is T- MAZE and Radial arm Maze. The assessment will be carried out initial and final supplementation of the seeds in rats. Rats were grouped into four based on the behavioural task, with 10 rats in each group. First group performed only T maze task and designated as TM group, and second group had only radial arm maze task and designated as RAM group. Third and fourth groups performed both the behavioural tasks (Praveen K.V. & Joseph Kurian Mukkadan, 2009).
Plate II
Radial Arm Maze.

Plate III
T- MAZE
3.4. To study the nutritional status of the selected school going children

3.4.1. Selection of area and subjects

A total 100 school going children were selected in between the age of 6-8 years were selected for supplementation in purposive sampling method was done. Fifty of them were taken as experimental group and reaming fifty of them as control group.

3.4.2. Tools selected for data collection

3.4.2.1. Interview schedule

A detail interview schedule was developed by including socio economic status, dietary pattern, and mental health questionnaire.

The interview method of collecting data involves presentation of oral-verbal stimuli and reply in term of oral-verbal responses (Kothari 2005).

3.4.2.2. Assessment of Anthropometric measurement

Anthropometry is considered to be the most sensitive parameters for assessing the nutritional status of growing children. It is based on the concept that an appropriate measurement should reflect any morphological variation occurring due to a significant functional and physiological change (Rao, 2000).

3.4.2.2.1. Body weight

Body weight is the most widely used and the sensitive and simplest reproducible anthropometric measurements for the evaluation of nutritional status of adult women’s. It indicates the body mass and is a composite of all body constituents like water, mineral, fat, protein and bone. It reflects more recent nutrition than does height.

The body weight was determined using a level balance, adjusted to zero and the reading were noted. Weights should be taken with the individual under basal conditions with minimum clothing and without shoes. The zero error of the weighing scale should be checked before taking the weight and corrected as and when required (Sreelakshmi 2010).
3.4.2.2. Height

The height of an individual is influenced both by genetic and environmental factors. The maximum growth potential of an individual is decided by hereditary factors, while the environmental factors, the most important being nutrition and morbidity, determine the extent of exploitation of that genetic potential.

Height of the subjects was determined using a fiber tap. The subjects were made to straight on a flat floor against a wall, without slippers and with feet close together and head, shoulder, back and buttocks and heels touched the wall and their head was held erect. A horizontal scale was placed gently to touch the head of the subject and mark on the wall.
According to Bamji et al., (2004), diet is a vital determinant of health and nutritional status of people. Information regarding food consumption pattern of children were taken through questioner and also were collected using 24 hour recall
method. Based on this the mean food intake and mean nutrient intake of children were calculated and compared with the Recommended Dietary Allowances (RDA) value given by ICMR(2004).

### 3.4.2.4.1. 24 Hour Recall

Information regarding the food intake of obese women was collected using 24-hours recall method. Based on this, mean food intake and mean nutrient intake of obese women were calculated and compared with Recommended Dietary Allowances (RDA) values given by ICMR (2004). This method requires an interviewer to ask the respondent to remember in detail all the food and drink they consumed during a period of time in the recent past (often the previous 24 hours).

The primary limitation of this method is that recording consumption for a single day is seldom representative of a person’s usual intake due to day-to-day variation (U.S Department of agriculture, 2010).

### 3.5. Effect of supplementation of pumpkin seed on memory of school going children

Nutrient ball was used for supplementation due to its high nutritive value and sensory evaluation. The supplementation was done in twelve school going children. The supplementation was done for two months and Short term memory test were used as scale. The initial reading and final reading were taken by using short term memory scale.

Pumpkin seed will be supplemented as nutrient ball using other protein rich cereals and pulses. The amount of supplementation will be based on pilot study in 10 subjects.

#### 3.5.1. Mental health questionnaire

Information regarding the mental health of children were taken through questionnaire because the children not only school they were stay away from home.
questionnaire includes the questions such as behavior attitude of children, emotional behavior of children, stress related events in children etc.

3.5.2. Short term memory test

Short-term memory (STM) ability is related to a range of cognitive tasks such as learning to read, reading complex text and arithmetic skills (Gathercole & Baddeley, 1993).

Short term memory test were done on children in two times. Before supplementation the reading may be taken after two months once more the reading may take. Then score the experimental group and control group as separately

3.5.3. Mental health questionnaire

Information regarding the mental health of children were taken through questionnaire because the children not only school they were stay away from home. In mental health questionnaire includes the questions such as behavior attitude of children, emotional behavior of children, stress related events in children etc.
4. RESULTS AND DISCUSSION

The result of this study entitled “Supplementary Effect of Pumpkin Seeds on Memory Function in Animal and Human Models” is discussed as follows,

4.1. To analyze the nutrient compounds in pumpkin seeds

4.1.1. Determination of proximate Properties of Pumpkin Seeds

4.1.2. Determination of Mineral Composition of Pumpkin Seed

4.1.3. Determination of amino acid Composition of Pumpkin Seed

4.1.4. Determination of fatty acid Composition of Pumpkin Seed

4.2. To develop the products

4.2.1. Sensory Evaluation of Developed Product.

4.2.2. Nutrient Analysis of Developed Product.

4.3. To study the memory Function of pumpkin seeds extract in animal models

4.4. To investigate the nutritional status of the selected school going children

4.4.1. General Profile of Children

4.4.2. Anthropometric Status of Children

4.4.3. Dietary Pattern of Children

4.5. To assess the supplementary effect of pumpkin seeds for increasing memory in selected school going children

4.5.1. Mental Health Questionnaire

4.5.2. Practical Interpretation of Memory Test Score

4.5.3. Short Term Memory Test

4.1. Proximate Composition of Pumpkin Seed Powder

Proximate composition of pumpkin seed are shown in table 1
The result shows that the raw pumpkin seed powder contains 1.9% moisture, 4.9% of total ash, 0.04% acid in soluble ash, 29.54% of curd protein, 3.9% of curd fiber, 49.85% curd fat, 15.71% of carbohydrate, 629.65% of energy, 9.91% of nitrogen free extract.

According to Pongjanta et al., 2006, Pumpkin flour was produced from mature pumpkin contain 6.01% moisture, 3.74% protein, 1.34 % fat, 7.24% ash, 2.9% fibre, 78.77% carbohydrate and 56.04% alcohol insoluble solids. The sample has 7.29 mg/100 g beta-carotene with colour values of L* 57.81, a* 8.31, b* 34.39 and 0.24 water activity.

### 4.2 Mineral composition of Pumpkin Seed

Mineral composition of pumpkin seed are shown in table 2

<table>
<thead>
<tr>
<th>Minerals</th>
<th>Amount (mean±SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Calcium (%)</td>
<td>0.139±0.01</td>
</tr>
<tr>
<td>Magnesium (%)</td>
<td>1.35±0.10</td>
</tr>
<tr>
<td>Phosphorus (%)</td>
<td>1.35±.10</td>
</tr>
<tr>
<td>Iron (%)</td>
<td>354.8±2.</td>
</tr>
<tr>
<td>Zinc (%)</td>
<td>109±1.00</td>
</tr>
<tr>
<td>Manganese (%)</td>
<td>212±0.10</td>
</tr>
<tr>
<td>Copper (%)</td>
<td>30±3.00</td>
</tr>
</tbody>
</table>

![Table 1](#)

**Table 1**

**Proximate Composition of pumpkin seed powder**

<table>
<thead>
<tr>
<th>Nutrients</th>
<th>Amount</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moisture (%)</td>
<td>1.9±0.30</td>
</tr>
<tr>
<td>Total ash (%)</td>
<td>4.9±0.59</td>
</tr>
<tr>
<td>Curd protein (%)</td>
<td>29.54±1.00</td>
</tr>
<tr>
<td>Crude fat (%)</td>
<td>3.9±0.02</td>
</tr>
<tr>
<td>Carbohydrate (%)</td>
<td>49.85±0.25</td>
</tr>
<tr>
<td>Energy(kcal)</td>
<td>629.65±0.10</td>
</tr>
<tr>
<td>Nitrogen free extract (%)</td>
<td>9.91±0.03</td>
</tr>
</tbody>
</table>
The table 2 shows that pumpkin seed contains 0.139% of calcium, 1.35% of magnesium, 1.35% of phosphorus, 354.8% of iron, 109% of zinc, 212% of manganese and 30% of copper.

According to Glew et al. 2006, Pumpkin seeds contained relatively large amounts of magnesium, zinc, copper, molybdenum and another minerals: phosphorus, calcium, iron, manganese, aluminum, barium, cobalt, strontium, nickel and arsenic.

4.3. Determination of amino acid Composition of Pumpkin Seed

Table 3
Amino acid composition of pumpkin seeds powder

<table>
<thead>
<tr>
<th>Processed Seed</th>
<th>Methionine (mean±SD) (g)</th>
<th>Threonine (mean±SD) (g)</th>
<th>Phenylalanine (mean±SD) (g)</th>
<th>Tryptophan (mean±SD) (g)</th>
<th>Arginine (mean±SD) (g)</th>
<th>Serine (mean±SD) (g)</th>
<th>Tyrosine (mean±SD) (g)</th>
<th>Proline (mean±SD) (g)</th>
<th>Isoleucine (mean±SD) (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Raw</td>
<td>1.2±0.06</td>
<td>3.22±0.12</td>
<td>9.45±1.02</td>
<td>2.60±0.70</td>
<td>5.10±0.30</td>
<td>1.27±0.07</td>
<td>3.0±0.20</td>
<td>3.45±0.02</td>
<td>6.56±0.01</td>
</tr>
<tr>
<td>f- ratio</td>
<td>46.935</td>
<td>52.875</td>
<td>7.038</td>
<td>.956</td>
<td>4.913</td>
<td>142.927</td>
<td>23.922</td>
<td>95.020</td>
<td>2.777</td>
</tr>
<tr>
<td>p-value</td>
<td>0.000</td>
<td>0.00</td>
<td>0.006</td>
<td>0.472</td>
<td>0.019</td>
<td>0.000</td>
<td>0.000</td>
<td>0.000</td>
<td>0.087</td>
</tr>
</tbody>
</table>

Each value represents the mean of 3 replications and expressed as Mean ± Standard Deviation. The amino acid present in pumpkin seeds are 1.2 g of methionine, 3.22g of threonine, 9.45g of tryptophan, 5.10g of arginine, 1.27g of serine, 3g of tyrosin, 3.45g of proline and 6.56g of isoleucine.

4.5. Determination of fatty acid Composition of Pumpkin Seeds

The maximum amount of fatty acid constituents present in the raw pumpkin seeds extract were benzonic acid, 4-ethoxy ethyl ester (C_{11}H_{14}O_{3}) with Retention Time(RT) of 8.17 with a peak area of 40.00%, followed by nonane, 4, 5-dimethyl (C_{11}H_{24}) with RT of 3.03 with a peak area of 23.21%. The minimum amount of fatty acid constituents were phthalic acid, Retention Time (RT) of 11.88, with the peak area of 1.29% the total unsaturated fatty acid present in raw pumpkin seeds was 44.82% (MUFA- 44.82%) and saturated fatty acid was 54.18%.
Figure 1
GC-MS Chromatogram of pumpkin seeds powder
Table 4
Components identified in raw pumpkin seeds powder [GC MS study]

<table>
<thead>
<tr>
<th>No.</th>
<th>RT</th>
<th>Name of the compound</th>
<th>Molecular Formula</th>
<th>Peak Area %</th>
<th>Probable compound structure</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>3.03</td>
<td>Nonane, 4,5-dimethyl</td>
<td>C_{11}H_{24}</td>
<td>23.21</td>
<td><img src="image1" alt="Structure" /></td>
</tr>
<tr>
<td>2.</td>
<td>4.94</td>
<td>Undecane, 2-methyl-</td>
<td>C_{12}H_{26}</td>
<td>13.82</td>
<td><img src="image2" alt="Structure" /></td>
</tr>
<tr>
<td>3.</td>
<td>5.52</td>
<td>Dodecane, 1-fluoro-</td>
<td>C_{15}H_{25}F</td>
<td>15.58</td>
<td><img src="image3" alt="Structure" /></td>
</tr>
<tr>
<td>4.</td>
<td>8.17</td>
<td>Benzoic acid, 4-ethoxy-, ethyl ester</td>
<td>C_{11}H_{14}O_{3}</td>
<td>40.0</td>
<td><img src="image4" alt="Structure" /></td>
</tr>
<tr>
<td>5.</td>
<td>8.93</td>
<td>Hexadecane</td>
<td>C_{16}H_{34}</td>
<td>1.29</td>
<td><img src="image5" alt="Structure" /></td>
</tr>
<tr>
<td>6.</td>
<td>10.04</td>
<td>Heptadecane, 2, 6,10,15-tetramethyl</td>
<td>C_{21}H_{44}</td>
<td>2.01</td>
<td><img src="image6" alt="Structure" /></td>
</tr>
<tr>
<td>7.</td>
<td>11.07</td>
<td>1-Docosene</td>
<td>C_{22}H_{44}</td>
<td>1.29</td>
<td><img src="image7" alt="Structure" /></td>
</tr>
<tr>
<td>8.</td>
<td>11.88</td>
<td>Phthalic acid, butyloctyl ester</td>
<td>C_{20}H_{30}O_{4}</td>
<td>1.29</td>
<td><img src="image8" alt="Structure" /></td>
</tr>
<tr>
<td>9.</td>
<td>13.34</td>
<td>17-Pentatriacontene</td>
<td>C_{35}H_{70}</td>
<td>1.53</td>
<td><img src="image9" alt="Structure" /></td>
</tr>
</tbody>
</table>
4.2. To develop the products

4.2.1. Sensory Evaluation of Developed Product

The sensory evaluation of product is shown in table 5. Standard Hedonic scale is used for evaluate product. Evaluation was done by 10 persons.

Table 5

Sensory Evaluation of Developed Product

<table>
<thead>
<tr>
<th>Type</th>
<th>Appearance</th>
<th>Colour</th>
<th>Flavor</th>
<th>Texture</th>
<th>Taste</th>
<th>Overall acceptability</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bread</td>
<td>3</td>
<td>4</td>
<td>4</td>
<td>3</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>Nutrient ball</td>
<td>9</td>
<td>8</td>
<td>9</td>
<td>9</td>
<td>9</td>
<td>9</td>
</tr>
<tr>
<td>Cake</td>
<td>7</td>
<td>5</td>
<td>5</td>
<td>6</td>
<td>5</td>
<td>5</td>
</tr>
</tbody>
</table>

Sensory Evaluation Result shows that bread have 3% of appearance, 4% of colour, 4% of flavor ,3% of texture, 4% taste, the overall acceptability of bread is 4%. Nutrient ball have 9% of appearance, 8% of colour, 9% of flavor, 9% of texture, 9% taste, the overall acceptability of bread is 9 %. Cake have 7% of appearance, 5% of colour, 5% of flavor ,6% of texture, 5% taste, the overall acceptability of bread is 5%. By comparing three product nutrient ball have high sensory evaluation.
Sensory Evaluation of Developed Product

4.2.2. Nutrient Analysis of Developed Products

Nutritive values of the developed product are given in table 6 and figure 3.

Table 6

<table>
<thead>
<tr>
<th>Nutrients / product</th>
<th>Bread (100g)</th>
<th>nutrient ball (100g)</th>
<th>Cake (100g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Energy (kcal)</td>
<td>272</td>
<td>599.2</td>
<td>518</td>
</tr>
<tr>
<td>Magnesium (mg)</td>
<td>27.3</td>
<td>330.2</td>
<td>252</td>
</tr>
<tr>
<td>Zinc (mg)</td>
<td>0.18</td>
<td>4.2</td>
<td>3.2</td>
</tr>
</tbody>
</table>
The nutrient content of bread was energy 272 k.cal, magnesium 296.5(mg) and zinc 4.1(mg). The nutrient ball contains 599.2 k.cal of energy, 330.2 mg of magnesium and 5.1(mg) of zinc. The cake contains energy 518 k.cal, magnesium 252 (mg), and zinc 3.2 (mg). As the result show nutrient ball has highest nutritive value.

Figure 3

Nutrient Analysis

4.6. To study the memory Function of pumpkin seeds extract in animal models

Table 7

Memory Function of pumpkin seeds extract in animal models
Effect of pumpkin seed on memory function of school going children

**T-maze task**

**Acquisition**: Number of trials required for acquisition of TM task of TM Group (Control) shows 15 trials whereas TM experimental group during acquisition and TM experimental group during retention shows 12.55 and 15.55 trials for acquisition of TM task respectively.

**Retention**: Number of trials required during retention was less compared to acquisition within all groups. TM Group (Control) shows 12.22 trials for retention of TM task, whereas TM experimental group during acquisition and TM experimental group during retention shows 11.58 and 7.55 trials for acquisition of TM task respectively.

**Radial arm maze task**

**Acquisition**: The number of trials required for acquisition of RAM task was similar in control and RAM experimental group during retention. It was 20.99 in RAM control group and 20.99 in RAM experimental group during retention.
**Retention:** Number of trials required during retention was less compared to acquisition within all groups. RAM Control group shows 16.15 trials for retention of RAM task, whereas TM experimental group during acquisition and TM experimental group during retention shows 16.44 and 8.99 trials for acquisition of TM task respectively.

The result of this study reveals that the number of trials required for retention of memory both in T maze and RAM analyse was less in experimental group compared to control group. This was proved that pumpkin seeds supplement (100 mg/kg) increase the memory of rats which is similar in human being.

4.4. To investigate the nutritional status of the selected school going children

4.4.1 Soci- Economic Background of Children

Details regarding the socio-economic background of children were elicited in terms of their age, sex, religion, type of family and birth order.

Table 8 show socio- economic background of children at the age of 6-8 years

<table>
<thead>
<tr>
<th>Sl.no</th>
<th>Socio-economic factor</th>
<th>Criteria</th>
<th>Total number of children</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Age</td>
<td>6</td>
<td>29</td>
<td>29</td>
</tr>
<tr>
<td></td>
<td></td>
<td>7</td>
<td>33</td>
<td>33</td>
</tr>
<tr>
<td></td>
<td></td>
<td>8</td>
<td>38</td>
<td>38</td>
</tr>
<tr>
<td>2</td>
<td>Sex</td>
<td>Male</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Female</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>3</td>
<td>Religion</td>
<td>Hindu</td>
<td>54</td>
<td>54</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Muslim</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Christian</td>
<td>46</td>
<td>46</td>
</tr>
</tbody>
</table>
Among the selected children 29 percentages of them were in the age group of 6 years and 33 percentages were in age of 7 years and remaining 33 percentages were in age of 8 years. 100 percentages of children selected for the study were females. Among the selected children 54 percentages were belonging to Hindu religion and remaining 46 percentages were Christian.

4.5.2. General Information

Table 9 shows the general information of the selected children.

Among selected children 75 percentage of them belong to nuclear family. Remaining 25 percent sample were belongs to joint family. The birth order of the selected sample shows that 29 percent of the samples were first child and 42 percent of sample were second child and 21 percent of sample were third child and 8 percent of them are of fourth child.

### Table 9
**General Information**

(\(N=100\))

<table>
<thead>
<tr>
<th>General information</th>
<th>Criteria</th>
<th>Total number of children</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Type of family</td>
<td>Nuclear</td>
<td>75</td>
<td>75</td>
</tr>
<tr>
<td></td>
<td>Joint</td>
<td>25</td>
<td>25</td>
</tr>
<tr>
<td>Birth order</td>
<td>First</td>
<td>29</td>
<td>29</td>
</tr>
<tr>
<td></td>
<td>Second</td>
<td>42</td>
<td>42</td>
</tr>
<tr>
<td></td>
<td>Third</td>
<td>21</td>
<td>21</td>
</tr>
<tr>
<td></td>
<td>Fourth</td>
<td>8</td>
<td>8</td>
</tr>
</tbody>
</table>

4.6. Anthropometric Status of Children

Anthropometric status of children is shown in Table 10

The table 8 shows that the mean weight of children in the age group of 6 years was 15.2. The mean weight of children in the age group of 7 years was 15. The mean weight of children in the age group of 8 was 18. The mean weight of children in all age group is below than the
standard weight given by ICMR (2004). The mean height of children in age group of 6 years was 112. The mean height of children in the age group of 7 years was 110.5. The mean height of children in the age group of 8 was 117.5. The mean height of children in all age group is below than the standard weight given by ICMR (2004)

Table 10  
Anthropometric status of children

<table>
<thead>
<tr>
<th>Age</th>
<th>Weight</th>
<th>Height</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>*Standard weight</td>
<td>Mean</td>
</tr>
<tr>
<td>6</td>
<td>19.5</td>
<td>15.2</td>
</tr>
<tr>
<td>7</td>
<td>21.8</td>
<td>15</td>
</tr>
<tr>
<td>8</td>
<td>24.8</td>
<td>18</td>
</tr>
</tbody>
</table>

(ICMR 2004)

4.7. Dietary Pattern of Children

The dietary pattern of children was studied in terms of their food habit, meal pattern, skipping of meal, favorite food, dislike food etc.

4.7.1. Food habit

Table 11 gives total food habit of the sample.

Table 11  
Food habit  (N=100)

<table>
<thead>
<tr>
<th>Categories</th>
<th>Number</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vegetarian</td>
<td>2</td>
<td>8</td>
</tr>
<tr>
<td>Non –vegetarian</td>
<td>22</td>
<td>92</td>
</tr>
</tbody>
</table>

Among the sample selected 8 percentage of the sample were vegetarian and 92 percentages were non-vegetarian.
According to the Scientific Advisory Committee on Nutrition (SACN), a diet lacking vegetables is likely to have adverse consequences for brain function and thus mental health and behavior.

### 4.7.2. Meal pattern per day

Table 12 gives the information of meal pattern per day

<table>
<thead>
<tr>
<th>Meal pattern</th>
<th>Number</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>One meal</td>
<td>Nill</td>
<td>_</td>
</tr>
<tr>
<td>Two meal</td>
<td>Nill</td>
<td>_</td>
</tr>
<tr>
<td>Three meal</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>Four meal</td>
<td>Nill</td>
<td>_</td>
</tr>
</tbody>
</table>

Among the sample selected 100 percentage of the sample were taking 3 meals per day. According to (Alaimo2001) eating the right nutrients at the right time during growth increases a child's potential. Yet, the physical, behavioral and cognitive development.

### 4.7.3. Skipping of meal

Table 13 gives the information of skipping of meal.

<table>
<thead>
<tr>
<th>Criteria</th>
<th>Number</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yes</td>
<td>Nill</td>
<td>Nill</td>
</tr>
<tr>
<td>No</td>
<td>100</td>
<td>100</td>
</tr>
</tbody>
</table>

Among the selected sample 100 percentage of were not skipping meals.
4.7.4. Favorite food

Table 14 shows the information of the favorite food of the selected sample.

**Table 14**

**Favorite food**

(N=100)

<table>
<thead>
<tr>
<th>Sl.no</th>
<th>Categories</th>
<th>Number</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Rice</td>
<td>16</td>
<td>16</td>
</tr>
<tr>
<td>2</td>
<td>Biryani</td>
<td>29</td>
<td>29</td>
</tr>
<tr>
<td>3</td>
<td>Ice cream</td>
<td>79</td>
<td>79</td>
</tr>
<tr>
<td>4</td>
<td>Chocolate</td>
<td>83</td>
<td>83</td>
</tr>
<tr>
<td>5</td>
<td>Fish</td>
<td>12</td>
<td>12</td>
</tr>
<tr>
<td>6</td>
<td>Milk</td>
<td>11</td>
<td>11</td>
</tr>
<tr>
<td>7</td>
<td>Egg</td>
<td>16</td>
<td>16</td>
</tr>
<tr>
<td>8</td>
<td>Chicken</td>
<td>20</td>
<td>20</td>
</tr>
</tbody>
</table>

Among the 24 samples 16% of them like rice, 29% of them like biryani, 79% of them like ice cream, 83% of them like chocolate, 12% of them like fish, 11% of them like milk, 16% of them like egg, 20% of them like egg. Chocolate and ice cream is most favorite food of children.

4.7.5. Dislike food

The table 15 shows the information of unfavourite food.
Table 15

<table>
<thead>
<tr>
<th>Dislike food</th>
<th>(N=100)</th>
</tr>
</thead>
</table>

<table>
<thead>
<tr>
<th>Criteria</th>
<th>Number</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bitter guard</td>
<td>29</td>
<td>29</td>
</tr>
<tr>
<td>Dosas</td>
<td>8</td>
<td>8</td>
</tr>
<tr>
<td>Uppma</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>Cabbage</td>
<td>12</td>
<td>12</td>
</tr>
<tr>
<td>Putt</td>
<td>16</td>
<td>16</td>
</tr>
<tr>
<td>Mango curry</td>
<td>16</td>
<td>16</td>
</tr>
</tbody>
</table>

Among the sample selected 29% of them dislike bitterguyard, 8% of them them dislike dosa, 20% of them dislike uppma, 12% of them them dislike cabbage ,16% of them them dislike putt , 16% of them dislike mango curry.

4.7.6. Food intake of children

The food intake of children is shown in the table 16.

Table 16

<table>
<thead>
<tr>
<th>Food intake of children</th>
</tr>
</thead>
</table>

<table>
<thead>
<tr>
<th>Food group</th>
<th>*Recommended Allowance</th>
<th>Actual intake</th>
<th>Percentage deficit or excess</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cereals</td>
<td>270</td>
<td>180</td>
<td>-66.6</td>
</tr>
<tr>
<td>Pluses</td>
<td>30</td>
<td>35</td>
<td>-10</td>
</tr>
<tr>
<td>Green leafy vegetable</td>
<td>100</td>
<td>10</td>
<td>+116.6</td>
</tr>
<tr>
<td>Roots and tubers</td>
<td>100</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>
The above table reveals that the food intake of children showed that 66.6% deficit in cereals, 10% in pulses, 30% in other vegetables, 30% in fruits, 34% in milk and milk products, 60% in fish/meat/poultry, 40% in fats and oil and 33% in sugar and jaggery compared to the recommended allowance of ICMR.

4.7.7. Nutrient intake of children

The mean nutrient intake of children is given in table 17.

Table 17

<table>
<thead>
<tr>
<th>Nutrient</th>
<th>*Recommended allowance</th>
<th>Actual intake</th>
<th>Percentage deficit or excess</th>
</tr>
</thead>
<tbody>
<tr>
<td>Energy (kcal)</td>
<td>1950</td>
<td>1408</td>
<td>-72</td>
</tr>
<tr>
<td>Protein (g)</td>
<td>41</td>
<td>97.2</td>
<td>-237</td>
</tr>
<tr>
<td>Fat (g)</td>
<td>25</td>
<td>31.1</td>
<td>+124</td>
</tr>
<tr>
<td>Iron (mg)</td>
<td>26</td>
<td>6.3</td>
<td>-24</td>
</tr>
<tr>
<td>Calcium (mg)</td>
<td>400</td>
<td>218.6</td>
<td>-55</td>
</tr>
<tr>
<td>B-carotene</td>
<td>2400</td>
<td>605</td>
<td>-25.2</td>
</tr>
<tr>
<td>Thiamin (mg)</td>
<td>1.0</td>
<td>0.54</td>
<td>-54</td>
</tr>
<tr>
<td>Riboflavin (mg)</td>
<td>1.2</td>
<td>0.59</td>
<td>-49.1</td>
</tr>
</tbody>
</table>

*ICMR2004
The mean intake of children revealed that the fat consumption exceed the RDA value by 124 per cent. The intake of other nutrient like Energy, Protein, Iron, Calcium, B-carotene, thiamin, Riboflavin intake were less than RDA values (ICMR 2004) by 72, 237, 24, 55, 25, 2, 54, 49.1 respectively.

Wolf and Burkman (2000) stated several dietary components support brain function and neurotransmitter activity, and that scientists recommend a wide range of foods as nutrient sources; the most important known today are protein, fat, B vitamins, iron, chlorine, and antioxidants.

Erickson (2006) discussed vitamins and minerals as an important substance for the functioning of the brain. Most important are the vitamins A, C, E, and B complex vitamins. Manganese and magnesium are two minerals essential for brain functioning; sodium, potassium and calcium play a role in message transmission and the thinking process.

4.8. Mental Health Questionnaire

4.8.1 Behavioral Attitude of Children

The behavioral attitude of children is given in Table 18

<table>
<thead>
<tr>
<th>No.</th>
<th>Behavioral Attitude</th>
<th>Categories</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Present</td>
</tr>
<tr>
<td></td>
<td></td>
<td>%</td>
</tr>
<tr>
<td>1</td>
<td>Trouble in paying attention</td>
<td>Nill</td>
</tr>
<tr>
<td>2</td>
<td>Often seem distrustful of others</td>
<td>33</td>
</tr>
<tr>
<td></td>
<td></td>
<td>33%</td>
</tr>
<tr>
<td>3</td>
<td>Often seem to express strange thoughts</td>
<td>Nill</td>
</tr>
<tr>
<td></td>
<td></td>
<td>-</td>
</tr>
<tr>
<td>4</td>
<td>Often seem blame others</td>
<td>79</td>
</tr>
<tr>
<td></td>
<td></td>
<td>79%</td>
</tr>
</tbody>
</table>

Table 18
Behavioral Attitude of Children
(N=24)
The above table shows that nobody had any trouble in paying attention. 33% of children may often seem too distrustful of other and 67 per cent had not.

No children are often seemed to express strange thought. 79% of children found to blame others and reaming 21% had not. According to (Kevin M, 2002) . Blaming is often intuitive and automatic, driven by a natural, impulsive desire to express and defend social value and expectation.

![Image of bar chart showing behavior attitudes of children]

**Figure 4**

**Behavior attitudes of children**

**4.8.2. Children behavior attitude in school**

Children behavior attitude in school is given in table 19.
Table 19
Children behavior attitude in school
(N=100)

<table>
<thead>
<tr>
<th>No</th>
<th>Problem at School</th>
<th>Categories</th>
<th>Present</th>
<th>%</th>
<th>Absent</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>School Grades</td>
<td></td>
<td>63</td>
<td>63</td>
<td>37</td>
<td>37</td>
</tr>
<tr>
<td>2</td>
<td>Skipping Classes</td>
<td>Nill</td>
<td>_</td>
<td>_</td>
<td>Nill</td>
<td>_</td>
</tr>
<tr>
<td>3</td>
<td>Behavior</td>
<td>17</td>
<td>17</td>
<td>83</td>
<td>83</td>
<td></td>
</tr>
</tbody>
</table>

The above table show that 63% of children’s had problem in school with grade and 37% had not. And anyone had no problem with skipping the class.

Seventeen percent of children had the problem at school with behavior 83% had not. According to Ainsworth & Filmer, 2006, orphans do have systematically lower educational outcomes than non-orphans; the significance of the relationship tends to be minimal after controlling for socio-economic status.

According to Minde (1988), orphans stress may be shown in symptoms of confusion, anxiety, depression, and behavioural disorders such as disobedience. The same symptoms may cause learning problems.
Figure 5
Children behavior attitude in school

4.8.3. Emotional behavior of children

Table 20 shows the emotional behavior of children.

**Table 20**

**Emotional behavior of children**

<table>
<thead>
<tr>
<th>No.</th>
<th>Emotional behavioral</th>
<th>Categories</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Present</td>
</tr>
<tr>
<td>1</td>
<td>Sadness</td>
<td>4</td>
</tr>
<tr>
<td>2</td>
<td>Angry</td>
<td>12</td>
</tr>
<tr>
<td>3</td>
<td>Nervous</td>
<td>21</td>
</tr>
</tbody>
</table>
The above table shows that 4% of children often seem sad and 96% had not. 12% of children may seem angry and remaining 96% may had not. 21% of children may seem nervous and remaining 79% had not.

4.8. 4. Stress related event in children

The stress related event is given in Table 21

Table 21

Stress related event in children

<table>
<thead>
<tr>
<th>No.</th>
<th>Criteria</th>
<th>Categories</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Present</td>
<td>%</td>
<td>Absent</td>
</tr>
<tr>
<td>1</td>
<td>Stress moving from one place to another place</td>
<td>71</td>
<td>71</td>
<td>29</td>
</tr>
<tr>
<td>2</td>
<td>Stress on separation of</td>
<td>54</td>
<td>54</td>
<td>46</td>
</tr>
</tbody>
</table>
The study shows that 71% of children had stress to moving from one place to another place and 29% had not.

54% of children have stress on separation of their parents and 46% had not. According to Tatek Abebe (2008). The losses of the parents continue to affect the children’s developmental stages.

Figure 7
Stress related event in children

4.9 practical interpretation of memory test score
4.9.1 Memory test score (by observation)

Memory test score of children is given in the table 22.
<table>
<thead>
<tr>
<th>Sl. no</th>
<th>Criteria</th>
<th>Before slowly</th>
<th>After Slowly</th>
<th>Before Medium</th>
<th>After Medium</th>
<th>Before Quickly</th>
<th>After Quickly</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Ability to learn and retain new material</td>
<td>21</td>
<td>12</td>
<td>25</td>
<td>17</td>
<td>54</td>
<td>71</td>
</tr>
<tr>
<td>2</td>
<td>Ability to learn and retain auditory versus visual information</td>
<td>8</td>
<td>-</td>
<td>21</td>
<td>12</td>
<td>71</td>
<td>87</td>
</tr>
<tr>
<td>3</td>
<td>Ability to store newly learned information after a delayed interval</td>
<td>29</td>
<td>8</td>
<td>21</td>
<td>37</td>
<td>50</td>
<td>54</td>
</tr>
<tr>
<td>4</td>
<td>Individual benefit from hints, cues, or in remembering material</td>
<td>13</td>
<td>-</td>
<td>33</td>
<td>21</td>
<td>54</td>
<td>79</td>
</tr>
</tbody>
</table>
The above table shows that before supplementation ability to learn and return new material 21% of children were slowly, 25% of children was medium and 54% was quickly. After supplementation 12% was slowly, 17% was medium and 71% was quickly.

The ability to learn and retain auditory versus visual information before supplementation 8% of children was slowly, 21% of children was medium and 71% was quickly. After supplementation nobody was slowly, 12% was medium and 87% was quickly.

The Ability to newly learned information store after a delayed interval in before supplementation 29% of children was slowly level, 21% of children was medium and 50% was quickly. After supplementation 8.3% was slowly, 37.5% was medium and 54% was quickly.

Individual benefits from hints, cues, or choice in remembering material 13% of children was slowly, 33% was medium and 54% was quickly. After supplementation nobody was slowly, 21% was medium and 79% was quickly.

4.10 Effect of supplementation on school going children (short term memory test)

Effect of supplementation is given in table 23

The result reveals that after 2 months of supplementation significantly increased the memory level. The memory function of children in experimental group was significantly (5%) increased compare to control group. The mean difference of experimental group was 16.7. The initial value of experimental group was 76.08±8.70 and increased as 92.25±6.21.
Table 23

Effect of supplementation on school going children

<table>
<thead>
<tr>
<th>Experimental group</th>
<th>Control group</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial (mean+S D)</td>
<td>Initial (mean+SD)</td>
</tr>
<tr>
<td>Final 92.2±6. 21</td>
<td>68.25±11. 39</td>
</tr>
<tr>
<td>Mean Difference 16.17</td>
<td>1.864</td>
</tr>
<tr>
<td>‘t’ value 8.377</td>
<td>0.089^{NS}</td>
</tr>
<tr>
<td>Sig 0.01*</td>
<td></td>
</tr>
</tbody>
</table>

NS – Non significant,*- significant at 5% level, ** significant at 1%level
SUMMARY AND CONCLUSION

The present study entitled *Supplementary Effect of Pumpkin Seeds on Memory Function in Animal and Human Models*” is summarized as follows. The important nutrients were analyzed using standard procedures. A total of 24 school going children in the age group of 6-8 years were purposively selected from at Edakkunu. The General Profile, Dietary Pattern and Anthropometric Status were assessed. Among the total twenty four school going children 12 were selected as experimental group and remains as control. Developed nutrient ball was supplemented to experimental group. Piratical Interpretation of Memory test Score and, Short Term Memory Test were taken before and after supplementation .The result of study indicate following,

- The raw pumpkin seed powder contains 1.9% moisture, 4.9% of total ash, 0.04% acid in soluble ash, 29.54% of curd protein, 3.9% of curd fiber, 49.85% curd fat, 15.71% of carbohydrate, 629.65% of energy, 9.91% of nitrogen free extract. The pumpkin seed contains 0.139% of calcium, 1.35% of magnesium, 1.35% of phosphorus, 354.8% of iron, 109% of zinc, 212% of manganese and 30% of copper.

- The amino acid present in pumpkin seeds are 1.2 g of methionine, 3.22g of threonine, 9.45g of tryptophan, 5.10g of arginine, 1.27g of serine, 3g of tyrosin, 3.45g of proline and 6.56g of isoleucine. The raw pumpkin seeds extract were benzoic acid, 4-ethoxy ethyl ester (C_{11}H_{14}O_{3}) with Retention Time (RT) of 8.17 with a peak area of 40.00%, followed by nonane, 4, 5-dimethyl (C_{11}H_{24}) with RT of 3.03 with a peak area of 23.21%. Retention Time (RT) of 11.88, with the peak area of 1.29% the total unsaturated fatty acid present in raw pumpkin seeds was 44.82% (MUFA- 44.82%) and saturated fatty acid was 54.18%.

- The Sensory Evaluation Result of the that bread have 3% of appearance, 4% of colour, 4% of flavour, 3% of texture, 4% taste, the overall acceptability of bread is 4%. Nutrient ball have 9% of appearance, 8% of colour, 9% of flavour, 9% of texture, 9% taste, the overall acceptability of bread is 9%. Cake have 7% of appearance, 5% of colour, 5% of flavour, 6% of texture, 5% taste, the overall acceptability of bread is 5%. By comparing three products nutrient ball have high sensory evaluation.
The nutrient content of bread was energy 272 k.cal, magnesium 296.5(mg) and zinc 4.1(mg). The nutrient ball contains 599.2 k.cal of energy, 330.2 mg of magnesium and 5.1(mg) of zinc. The cake contains energy 518 k. cal, magnesium 252 (mg), and zinc 3.2 (mg). As the result show nutrient ball has highest nutritive value.

In T-maze test the Acquisition of the Number of trials required for acquisition of TM task of TM Group (Control) shows 15 trials whereas TM experimental group during acquisition and TM experimental group during retention shows 12.55 and 15.55 trials for acquisition of TM task respectively.

In T-maze test the Retention of the Number of trials required during retention was less compared to aacquisition within all groups. TM Group (Control) shows 1222 trials for retention of TM task, whereas TM experimental group during acquisition and TM experimental group during retention shows 11.58 and 7.55 trials for acquisition of TM task respectively.

In Radial arm maze task the Acquisition of the number of trials required for acquisition of RAM task was similar in control and RAM experimental group during retention. It was 20.99 in RAM control group and 20.99 in RAM experimental group during retention.

In Radial arm maze task the Retention of the number of trials required during retention was less compared to acquisition within all groups. RAM Control group shows 16.15 trials for retention of RAM task, whereas TM experimental group during acquisition and TM experimental group during retention shows 16.44 and 8.99 trials for acquisition of TM task respectively.

Among the selected children most of them in between the age group 7 and 8 years.

Anthropometric measurement of the selected subject indicated that the mean height weight of children in all age group was below than the standard weight.

Food intake of children revealed that the fat consumption exceed the RDA value by 124 per cent. The intake of other nutrient like Energy, Protein, Iron, Calcium, B-carotene, thiamin and riboflavin were less than RDA values (ICMR 2004) by 72,237,24,55,25.2,54,49.1 respectively

Among the selected children most of them (67%) seen to distrustful of others. Majority of 79% of children found to blame others.
Most of them (63%) had problem in school with grade. Majority (83%) of them had not any the problem at school with behavior.

Majority 96% of children had not seemed sadness in nature. Most of the 79% of children had not seen angry in nature.

The study shows that most of the (71%) children had stress to moving from one place to another and half of them 54% had stress on separation of their parents.

After supplementation, 71% of them improved their ability to learn and memories new material.

After supplementation, 87% of children improved ability to learn and retain auditory versus visual information.

Supplementation increased (54%) the ability to newly learned information store after a delayed interval.

After supplementation 79% of children benefits from hints, cues, or choice in remembering material.

The memory function of children in experimental group was significantly (5%) increased compared to control group. The mean difference of experimental group was 16.7. The initial value of experimental group was 76.08±8.70 and increased as 92.25±6.21.

The effect of supplementation shows that, there was increased in memory level as the result of supplementation of pumpkin seed nutrient ball. The memory function of children in experimental group was significantly increased compare to control group.

Conclusion

Pumpkin seed is a nutrient nutritional power house. Pumpkin seeds are rich in protein, magnesium and manganese, but zinc is the first important ingredient when it comes to improving memory. Zinc helps the neurons within the hippocampus communicate. The hippocampus is like a big filing cabinet in our brain, storing all your memories and information you learn. 40gm of pumpkin seed supplemented daily for the period of one month help to increase brain function. Hence the study concludes that food trends may come and go but the excellent natural nutrient source of pumpkin seed are ever powerful weapon for fighting to enhance the brain function of children.
BIBLIOGRAPHY


Effect of pumpkin seed on memory function of school going children


- Committee on Herbal Medicinal Products (HMPC) 13 September 2011
  EMA/HMPC/136022/2010


The pumpkin patch,(2007). Halloween oneline. ([http://www.pumpkin-patch.com](http://www.pumpkin-patch.com)).


NET REFERANCE

- [http://colleges.ksu.edu.sa/](http://colleges.ksu.edu.sa/)
Effect of pumpkin seed on memory function of school going children

http://inpiyr.com/pumpkin-seeds-5-reasons-add-diet/
http://inpiyr.com/pumpkin-seeds-5-reasons-add-diet/
http://inpiyr.com/pumpkin-seeds-good-for-you/.
http://www.buzzle.com/articles/pumpkin-seeds-benefits.html
http://www.buzzle.com/articles/pumpkin-seeds-benefits.html
http://www.encognitive.com/files/Pumpkin%20seed%20power.pdf
http://www.houseofnutrition.com/pumpkin.html
http://www.squidoo.com/pepitas
http://www.weekendbakery.com/cooking-conversions/
APPENDIX I

INTERVIEW SCHEDULE

A. Social-Economic Profile

1. Name of investigator : 
2. Name of interviewee : 
3. Age in year : 6 ☐ ☐ ☐ 
4. Sex : Male ☐ Female ☐ 
5. Religion : Hindu ☐ Muslim ☐ Christian ☐ 
6. Type of family : Nuclear ☐ Joint ☐ 
7. Birth order : 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 

B. Anthropometric Measurement

8. Height : 
9. Weight : 

C. Food consumption pattern of the children

10. Food habit of the subject : Vegetarian ☐ Non-vegetarian ☐ 
11. Number of meals per day : 1 ☐ 2 ☐ 3 ☐ 4 ☐ 5 ☐ 
12. Do you skip meal : Yes ☐ No ☐ 
13. List your favorite food : 
14. List the food dislike most : 
15. 24 Hours recall method
Effect of pumpkin seed on memory function of school going children

<table>
<thead>
<tr>
<th>Time</th>
<th>Food item</th>
<th>Approximate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Early morning</td>
<td>Breakfast</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Lunch</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Evening snacks</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Dinner</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Bed time</td>
<td></td>
</tr>
</tbody>
</table>

D. Practical Interpretation Of Memory Test Scores

16. What is the individual’s ability to learn and retain new material?

   Slowly □  Medium □  Quickly □

17. Are there differences in the individual’s ability to learn and retain auditory versus visual information?

   Slowly □  Medium □  Quickly □

18. How well is newly-learned information stored after a delayed interval?

   Slowly □  Medium □  Quickly □

19. Does the individual benefit from hints, cues, or choices in remembering material?
Effect of pumpkin seed on memory function of school going children

Slowly □    Medium □    Quickly □

E. Mental Health Questionnaire

20. Is the subject has trouble paying attention?
   Yes □  No □

21. Is the subject has often seem distrustful of others?
   Yes □  No □

22. Is the subject has often seem to express strange thought?
   Yes □  No □

23. Is subject has often seem blame others?
   Yes □  No □

24. Is the subject has problem at school with behavior?
   Yes □  No □

25. Is the subject has problems at school with grade?
   Yes □  No □

26. Is the subject has problem at school with skipping classes?
   Yes □  No □

27. Is the subject has often complain of “not feeling well”?
28. Is the subject has often seem sad?
   Yes ☐ No ☐

29. Is the subject has often seem angry?
   Yes ☐ No ☐

30. Is the subject has often seem nervous of afraid?
   Yes ☐ No ☐

31. Is the subject has any recent stress on the family or child?
   Yes ☐ No ☐

32. Is the subject has any recent stress on family or child?
   Yes ☐ No ☐

33. Is the subject has any stress on moving from one place to another place?
   Yes ☐ No ☐

<table>
<thead>
<tr>
<th>Score/ Rating</th>
<th>Standard Hedonic scale</th>
<th>Judge</th>
<th>Appearance</th>
<th>colour</th>
<th>Texture</th>
<th>Flavor</th>
<th>Taste</th>
<th>Overall accepta nce</th>
</tr>
</thead>
<tbody>
<tr>
<td>9</td>
<td>I like extremely</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>8</td>
<td>I like very much</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>I like moderately</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>I like slightly</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>I either like or dislike</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
### D. Standard Hedonic scale

#### E. Short Term Memory Test

<table>
<thead>
<tr>
<th>Trial</th>
<th>Total number of letters in the set</th>
<th>Correct letters</th>
<th>Total number of letter you remembered</th>
<th>% you remembered</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>2</td>
<td>UM</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>4</td>
<td>TZLD</td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>6</td>
<td>KXCEJO</td>
<td></td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>8</td>
<td>AVCYISEH</td>
<td></td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>10</td>
<td>LBFQRPMAUX</td>
<td></td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>12</td>
<td>ZQECTBUMONRV</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>