Objectives:

• To identify the most commonly consumed carbohydrate rich foods that constitute typical Indian meals.

• To standardize the recipe of the selected cooked food items

• To carry out proximate analysis of selected test food to quantify the total carbohydrate content (starch, reducing, non reducing sugars), protein, fat and moisture content of the selected food samples

• To determine the Glycemic Bread Equivalent (GBE) value of the selected, commonly consumed carbohydrate rich foods

• To find the insulinergic effects (secretagogue) of these selected foods

• To assess the relation of Insulinergic response and glycemic response of different foods

• To study the dose response of the selected foods (50 & 100grams) on the postprandial blood glucose and insulin level
Carbohydrates (CHO) are defined chemically as aldehyde or ketone derivatives of the higher polyhydric alcohols, or compounds which yield these derivatives on hydrolysis (Chatterjee & Shinde 2005). Plant carbohydrates vary widely in sweetness, rate of digestion and the degree of absorption. As a dietary component, they contribute a major percentage of energy requirements serving as the basic fuel for the body. In India, 65-70% of energy consumed is in the form of carbohydrate (Sudha et al 2004).

In 2002, the National Academy of Sciences–Food and Nutrition Board recommended that diets provide 45–65% of calories from carbohydrate, with a minimum intake of 130 g carbohydrate/day for adults. Raghuram, Pasricha, & Sharma (2003) suggested that 60-65% of total energy requirement can be derived from carbohydrate of the desirable kind. The major sources of carbohydrate are cereals; Rice and wheat, being the staple food item in South and North respectively representing over 50% of all carbohydrate consumed in developing countries, with sugar crops the next major source, followed by root crops, fruits, vegetables, pulses and milk products (FAO/WHO 1998).

Recent analysis of the consumption expenditure data collected from National Sample Surveys of 27th (1972-73), 43rd (1987-88) and 50th (1993-94) rounds, it was observed that the consumption expenditure in cereals and cereal substitutes have decreased with Indian ruralites spending more on pulses, milk & milk products, edible oils, meat, egg & fish and vegetables (Turan 2001). In some of the developing countries much of the carbohydrate is derived from a single food source such as Rice, cassava or maize. Carbohydrate foods are an important vehicle for protein, micronutrients and other food components, like phytochemicals, which have important benefits for health.
Traditional Asian diets are cereal-based, rich in fiber and low in saturated fat, cholesterol and meat, but as societies move up the socio-economic scale, changes take place in both dietary structures and patterns (Gopalan 1992). A trend towards greater consumption of animal as compared to vegetable sources in the diet has led to an increased energy intake. Popkin et al (2001) noted a large shift from consumption of coarse grains such as sorghum, barley, rye, maize and millet to more refined cereals, like polished Rice and wheat especially among the urban population and higher income groups. These changes have resulted in a significant decrease in the overall fiber content of the diet. Hindu vegetarians from India whose diet is composed largely of low-fat grains and pulses (legumes) maintain CHD rates equal to (Begom et al 1995) or higher (Miller et al 1988) than those in the USA and countries of Europe, despite their diets' lower total fat content when compared to American and European diets. In a typical Indian diet, the coefficient of digestibility is 98% for CHO and 1 gm of absorbed or available CHO gives about 4 kcal of energy. This absorption and availability of CHO depends on the quality of CHO.

**Classification of carbohydrates**

All carbohydrates are compounds of carbon, hydrogen and oxygen. Carbohydrates are classified according to their degree of polymerization as: sugars (mono- and disaccharides), oligosaccharides (contain thee to nine monosaccharide units), and polysaccharides (contain ten or more monosaccharide units) (Pigman & Horton 1972) (Table 2.1).
### Table 2.1: Chemical classification of dietary carbohydrates

<table>
<thead>
<tr>
<th>Class (DP)</th>
<th>Subgroup</th>
<th>Principal components</th>
<th>Physiology</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Sugars (1-2)</strong></td>
<td>Monosaccharides</td>
<td>Glucose, Fructose, galactose</td>
<td>Absorbed in the ileum; glucose gives a more rapid response than fructose</td>
</tr>
<tr>
<td></td>
<td>Disaccharides</td>
<td>Sucrose, lactose, maltose, trehalose</td>
<td>Absorbed form the duodenum. Sucrose produces more rapid response then fructose</td>
</tr>
<tr>
<td></td>
<td>Polyols (sugar alcohols)</td>
<td>Sorbitol, Mannitol, lactitol, xylitol, erythitol, isomalt, maltitol</td>
<td>Poorly absorbed and partially fermented</td>
</tr>
<tr>
<td><strong>Oligosaccharides (3-9 short chain)</strong></td>
<td>Malto-oligosaccharides (alpha – glucans)</td>
<td>Maltodextrins</td>
<td>Physiological behavior can differ</td>
</tr>
<tr>
<td></td>
<td>Non alpha-glucans oligosaccharides</td>
<td>Raffinose, Stachyose, fructo and galacto oligosaccharides, polydextrose, inulin</td>
<td>Not hydrolyzed by human digestive enzymes. Fermented in the colon</td>
</tr>
<tr>
<td><strong>Polysaccharides (≥10)</strong></td>
<td>Starch (alpha-glucans)</td>
<td>Amylose, amyllopectin, modified starches</td>
<td>Raw starch is not digested (Cummings and Englyst 1995) When gelatinized it is digested easily and speed of digestion is affected by many factors (Beyer 2004) Cooked and cooled starch may become resistant due to retrogradation (Asp &amp; Bjorck 1992)</td>
</tr>
<tr>
<td></td>
<td>Non-starch Polysaccharides (NSP)</td>
<td>Cellulose, hemicellulose, pectin, arabinoxylans, beta-glucan, glucomannans, plant gums and mucilages, hydrocolloids</td>
<td>Not hydrolyzed by human digestive enzymes. Fermented in the colon</td>
</tr>
</tbody>
</table>

DP- Degree of Polymerization or number of monomeric units

Monosaccharides: These are the simplest carbohydrate molecules. The most commonly occurring monosaccharides in food are glucose, fructose and galactose. Glucose (also called dextrose) is found in varying amounts in honey, maple syrup, fruits, berries, and vegetables. Glucose and fructose are often formed from the hydrolysis of sucrose, as in honey, maple sugar, and invert sugar. Glucose is also present in foods containing starch hydrolysis products, such as corn syrups and high-fructose corn syrups. Fructose may also be present in food products, such as soft drinks, bakery products, and candies from the use of invert sugar, crystalline fructose or high-fructose corn syrups (HFCS).

Disaccharides: These sugars are formed when two monosaccharide molecules join together with the removal of one molecule of water. Examples of disaccharides are sucrose (glucose and fructose), lactose (glucose and galactose) and maltose (2 molecules of glucose). Sucrose (alpha-D-glucopyranosyl beta-D-fructofuranoside) is a non reducing sugar and is the major disaccharide in most diets. It is present in honey, maple sugar, fruits, berries, and vegetables. Sucrose can provide a number of desirable functional qualities to food products including sweetness, mouth-feel, and the ability to transform between amorphous and crystalline states (Chinachoti 1995). Lactose, a reducing sugar also known as milk sugar, occurs in milk and milk products. Whey, obtained from milk, is used as an ingredient in foods and is high in lactose content.

Oligosaccharides: Oligosaccharides are not widely occurring except for a series of galactosyl sucroses (often designated as beta-galactosides) and fructo-oligosaccharides. The galactosyl sucrose family of oligosaccharides include raffinose (a trisaccharide), stachyose (a tetrascaccharide), and verbascose (a pentasaccharide). In vegetables, such as peas, beans,
lentils, the content of these oligosaccharides can range from 5-8% on a dry matter basis (Asp1995). Raffinose, stachyose, and verbascose are not digested in the small intestine by human gastrointestinal enzymes hence they pass into the large intestine where they are fermented by intestinal microflora with the production of gas. Fructo-oligosaccharides occur in wheat, rye, triticale, asparagus, onion, and Jerusalem artichoke and a number of other plants (FAO/WHO 1998)

**Polysaccharides:** Polysaccharides such as starch, glycogen, cellulose, beta glucan and pectin are made up of many monosaccharide molecules (usually glucose), joined together. Common food starches are derived from seed (wheat, maize, Rice, barley) and root (Potato, cassava/tapioca) sources. Most common cereal starches contain 20-30% amylose and the rest is amyllopectin. Waxy starches (maize, Rice, sorghum, barley) have no amylose and contain essentially 100% amyllopectin.

Major polysaccharide compounds:

A) Starch (found in Potatoes, bread, Rice and pasta).

B) Non-starch polysaccharides (NSP), e.g. cellulose, pectins and gums, found in fruits, vegetables, beans and whole-grain cereals.

A) Starch: Starch being the most common polysaccharide is made up of amylose and amyllopectin chains. The structural and molecular arrangement is peculiar for amylose and amyllopectin fractions (Table 2.2) and their ratios in starch vary depending on the source.
Table 2.2: Properties of amylose and amylopectin fractions of starch

<table>
<thead>
<tr>
<th>Properties</th>
<th>Amylose</th>
<th>Amylopectin</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chain length</td>
<td>Long, straight</td>
<td>Branched</td>
</tr>
<tr>
<td>Arrangement</td>
<td>Densely packed</td>
<td>More open, lower density</td>
</tr>
<tr>
<td>Size</td>
<td>Small</td>
<td>Large</td>
</tr>
<tr>
<td>Rate of digestion</td>
<td>Slow</td>
<td>Faster</td>
</tr>
</tbody>
</table>

**Source:** McWilliams 2001, Bennion & Scheule 2000, Charley & Weaver 1998, Brand Miller et al, 1996

Amylose are straight chain glucose molecules having alpha 1-4 glucosidic linkages whereas amylopectin constitutes branched chain with both alpha 1-4 and alpha 1-6 linkages. Amylose forms a helical open structure with unbranched α-glucan chains which can form complexes with small hydrophobic molecules especially lipids. Foods may contain small amounts of free fatty acids that could complex with amylose during eating. Complexes could also form in the small intestine by the interaction of linear alpha glucan fragments from partially digested amylose and amylopectin with free fatty acids released from triglycerides by the action of lipases (Crowe, Seligman, and Copeland 2000). These amylose-lipid complexes change functional properties of starch such as reduced solubility, increased gelatinization temperature and retarded retrogradation during storage (Eliasson et al. 1981, Krog 1971)

Starch in its native form is insoluble in cold water but can be solubilised by heating with excess water. The change in the conformation during application of moist heat results in the loss of the crystallization of amylopectin followed by swelling, hydration & solubilisation.
The process is called gelatinization. The extent of gelatinization of starch during cooking will depend on the amylose: amylopectin ratio, starch granule properties, degree of milling, type of wheat (soft/durum) and the level of damaged starch (Lang 2004, Colonna et al 1990, Colonna, Leloup & Buleon 1992). Amylose tends to quickly undergo retrogradation to form semi-crystalline structure which is more resistant to enzyme attack thereby producing a lower glycemic response (Bjo¨rck et al. 1994, Goddard et al. 1984, Berry 1986, Biliaderis 1991). Re-crystallization of molecules rarely occurs for amylopectin fractions hence foods containing higher proportion of these undergo rapid digestion and absorption. Proximate composition along with amylose content of some common starches is given in Table 2.3.

<table>
<thead>
<tr>
<th>Nutrient</th>
<th>Potato</th>
<th>Cassava</th>
<th>Wheat</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moisture,%</td>
<td>19</td>
<td>13</td>
<td>13</td>
</tr>
<tr>
<td>Ash,%</td>
<td>0.4</td>
<td>0.2</td>
<td>0.2</td>
</tr>
<tr>
<td>Protein,%</td>
<td>0.06</td>
<td>0.1</td>
<td></td>
</tr>
<tr>
<td>Lipid,%</td>
<td>0.05</td>
<td>0.1</td>
<td>0.8</td>
</tr>
<tr>
<td>Phosphorus%</td>
<td>0.08</td>
<td>0.01</td>
<td>0.06</td>
</tr>
<tr>
<td>Amylose,%</td>
<td>21</td>
<td>17</td>
<td>28</td>
</tr>
</tbody>
</table>

**Source:** Muhbeck & Eliasson 1987.

Wheat and Rice are the major cereals being consumed in India. It is important to study their starch characteristics (Table 2.4) and their impact on health.
Table 2.4: Characteristics of starch in wheat and Rice

<table>
<thead>
<tr>
<th>Cereal</th>
<th>Type of Granule</th>
<th>Diameter (µm)</th>
<th>Reference</th>
<th>Amylose (%)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wheat B</td>
<td>Small-spherical</td>
<td>4.12</td>
<td>Soulaka &amp; Morrison 1985a</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rice</td>
<td>1.5-7.5</td>
<td></td>
<td>Yeh &amp; Li 1996</td>
<td>25.6</td>
<td>Lii, Shao &amp; Tseng 1995</td>
</tr>
</tbody>
</table>

*Nutritional classification of starch:*

Englyst and Cummings (1987a) were the first to propose a nutritional classification of starches based on the rapidity with which glucose is released from a food source. The intrinsic properties of starch such as chemical structure and hydration as well as extrinsic factors such as fat and fiber content of the product influence the rate of starch digestion (Liljeberg, Granfeldt, & Bjorck 1992, Granfeldt et al 1995).

Three sub-fractions of starch have been identified:

1) *Rapidly digestible starch (RDS)* – It consists mainly of amorphous and dispersed starch. Starchy foods cooked by moist heat eg bread and Potatoes contain these in high amounts. These starches get digested very quickly and release glucose within 20 min.

2) *Slowly Digestible Starch (SDS)* – It is digested slowly but completely. This constitutes physically inaccessible amorphous and raw starch. It releases glucose 100 min after enzyme digestion.
iii) Resistant starch-Resistant starch has been defined as "the sum of starch and products of starch degradation not absorbed in the small intestine of healthy individuals" (Muir et al 1993). Resistant starch is digested only upto 50% in the small intestine as observed though exogenous glucose response in serum and the CO$_2$ excretion in breath (Vonk et al 2000). The amount of RS present may be influenced by several factors such as water, amylose, amylose:amylopectin ratio, pH, heating or cooking temperature, number of heating & cooling cycles, freezing & drying (Berry 1986).

Three types of resistant starch have been identified (Englyst, Kingman, & Cummings 1993).

1. RS1 - Physically trapped starch: These starch granules are physically trapped within a food matrix so that digestive enzymes are prevented or delayed from having access to them. This can occur in whole or partly ground grains, seeds, cereals, and legumes. The amount of type 1 resistant starch will be affected by food processing and can be decreased or eliminated by milling.

2. RS2 - Resistant starch granules: Certain raw (native) starch granules, such as Potato and green banana, are known to resist attack by alpha -amylase. This is probably related to the crystalline nature of the starch (i.e., crystalline regions of the starch granule are less susceptible to attack by acid and enzymes than the amorphous regions). Gelatinized starch is much more rapidly digested by enzymes than is raw starch hence gelatinized Potato and green banana starch are digested by alpha -amylases.

3. RS3 - Retrograded starch: Retrogradation of starch involves re-crystallization of the amylopectin fraction upon cooling of the gelatinized starch. The amylose and amylopectin
components of starch undergo the process of retrogradation in a time dependent process after starch has been gelatinized or cooked. Re-association of the amylose and amylopectin occurs with separation of the mixture into polymer-rich and polymer deficient phases. The rate at which amylose retrogrades is much higher than that for amylopectin which has much shorter chain lengths. Amylose can be retrograded to a form that resists dispersion in water and digestion with α-amylase (Muir et al 1993).

Retrograded starch resists digestion by alpha amylase and pullulanase enzymes thereby may be measured as dietary fiber (Englyst et al 1983). Retrogradation is enhanced at low temperatures. Freshly cooked Potato was reported to contain less than 2% resistant starch; however cooling resulted in an increase up to 3% (Englyst & Cummings 1987)

*Effect of processing technique on Resistant Starch (RS)*

The botanical source of starch, its molecular structure and starch interactions due to processing, cooking and eating may play a role in determining the amount and form of resistant starch (Englyst & Cummings 1982, Englyst et al 1996, Muir & O’Dea 1992). Generation of RS from raw foods require profound disruption of native granular structure resulting in a more crystalline conformation. This may be interpreted as the need to release amylose from its native associations, either with itself, from lipids, from amylopectin as a precondition for its subsequent self association to form crystalline RS3 with the property of resisting amylolysis (Berry 1986).

Gelatinization & retrograduation are important processes that govern the formation of RS. The formation of RS in food can be regulated by selecting specific processing techniques (Katyal
et al 2005). Pressure cooking increases the total RS content of legumes. Steam cooking decreases digestibility of starch in legumes, increasing the levels of RS (Tovar & Melito 1996). Cooking results in release of encapsulated starch thereby causing a reduction in RS1, but simultaneously amylose retrogradation markedly increases RS3 content (Brighenti et al 1998).

The resistant starch in bread is formed immediately on baking and remains very stable for more than a week of storage due to the high starch suspension formed whereas for boiled Potato with a relatively dilute starch suspension the development of resistant starch is time dependent (Englyst, Wiggins & Cummings 1982).

Nutritional classification of carbohydrates is the most effective means of understanding its metabolic effects. The study of resistant starch and resistant oligosaccharides as functional ingredients separate from dietary fiber is more relevant in the current context.

According to Englyst , Liu & Englyst (2007), structured approach to the characterization of nutritionally relevant features of dietary carbohydrates can provide the basis for establishing population reference intakes, nutrition claims and food labeling will assist the consumer with properly informed dietary choices.
B) Non starch Polysaccharides

*Dietary fiber:* Dietary fiber occurring in foods and food products can be considered to consist of cellulose, hemicelluloses, pectic substances, hydrocolloids (gums and mucilages), resistant starches, and resistant oligosaccharides. In March 2001, the Food and Nutrition Board, Institute of Medicine of the National Academy of Sciences released proposed definitions for dietary fiber as consisting of non-digestible carbohydrates and lignin that are intrinsic and intact in plants (Slavin 2003).

*Cellulose:* It the major cell wall structural component in plants and has been used as a bulking agent in food due to its water-absorbing ability and low solubility. Hemicellulose may be present in soluble and insoluble forms and is comprised of a number of branched and linear pentose- and hexose-containing polysaccharides.

*Beta-glucans:* Mixed linkage beta –glucans, have generated considerable interest in recent years due to their physiological response as soluble dietary fiber. While these glucans are found in relatively small quantities in wheat, they are a major component of cell-cell material in barley and oats. These glucans form viscous aqueous solutions and have been shown to be effective in reducing serum cholesterol concentrations (Anderson & Bridges 1993). Oat bran, a rich source of beta -D-glucan, has been incorporated into many food products, particularly cereals, as a source of the soluble fiber that has been touted for cholesterol reduction.

*Pectins:* Pectins are used in jams and jellies because of their ability to form stable gels. Hydrocolloids (gums, mucilages) are used in small amounts in food products for their
thickening (viscosity increasing), gelling, stabilizing, or emulsifying ability. They are derived from seaweed extracts, plant exudes, seeds, and microbial sources.

**High Carbohydrate and High Fibre diet (HCHF):** Extensive clinical and experimental studies by Prof. Viswanathan and colleagues (1991) showed that the HCHF diet is suitable for Indian diabetics, as it helped in achieving rapid and sustained control of hyperglycemia for several years and consistent reduction in the plasma cholesterol and triglyceride levels. Increased peripheral sensitivity to insulin was also observed, perhaps by improving the insulin receptor number and or affinity at post receptor level. Studies in experimental animals have indicated that high carbohydrate diets improve glucose metabolism by enhancing the activity of glycolytic enzymes and depressing the activity of glucogenic enzymes in the liver and thereby decreased the incidence of long term vascular complication (Viswanathan & Mohan 1991).

Foods rich in fiber delay glucose absorption and regulate postprandial hyperglycemia. Studies have shown that increasing the non starchy vegetables containing 3-4% of energy as carbohydrate (green leafy vegetables, and other vegetables like cucumber, cauliflower, ladies finger etc) in the diet contributed to bulk as well as greater satiety. Similar recommendations are also made by the study group of the European Association for the Study of Diabetes (EASD) (Giacco et al 2000). It has been well documented that viscous fiber (soluble fiber - vegetables and legumes) intake flattens the postprandial glycaemic profile in diabetic subjects. Studies have also confirmed its role in reducing postprandial insulin levels (Lafrance et al 1998).
Other classification:

1) Available and unavailable carbohydrates: In terms of their physiological or nutritional role, carbohydrates are often classified as available and unavailable. Available carbohydrates are those that are hydrolyzed by enzymes of the human gastrointestinal system to monosaccharides. These are absorbed into the small intestine and enter the pathways of carbohydrate metabolism. Unavailable carbohydrates are not hydrolyzed by endogenous human enzymes, although they may be fermented in the large intestine to varying extents (FAO/WHO 1998).

2) Readily available glucose (RAG) and slowly available glucose (SAG): A similar classification of starch based on RAG & SAG has been studied to understand its physiological effects on glucose metabolism. The glycemic index (GI) and insulin index (II) correlates positively and negatively with the RAG and SAG content in given food items respectively. Low GI foods that cause a slow release of glucose in the blood have high SAG and are proposed to have several health benefits (Englyst et al 2003).

3) Glycemic and non-glycemic carbohydrates: Glycemic carbohydrates include most mono and disaccharides, some oligosaccharides (maltodextrins) and rapidly and slowly digested starches. Non-glycemic carbohydrates include remaining oligosaccharides, NSP’s and RS (Cummings & Stephen 2007). Glycemic carbohydrate also called available CHO is available for metabolism and is the summation of analytical values of mono, di- and oligosaccharides, starch and glycogen but excludes fructo-oligosaccharides and other non digestible oligosaccharides and resistant starch (Department of Health, 2002).
4) **Indigestible carbohydrates**: The carbohydrates that escape digestion in the small intestine undergo fermentation in the colon. The reduced digestibility of such fermentable CHO’s is attributed to the dietary fiber or resistant starch content. Insoluble fibers (e.g., lignans, cellulose, and some hemicelluloses) that are resistant to colonic fermentation may carry with them fermentable carbohydrate substrate, including starches and sugars, although their major role is in fecal bulking. Soluble fibers (e.g., pectins, gums, mucilages, some hemicelluloses, as well as inulin-type fructans) are generally more completely fermented with little effect in increasing fecal bulk. Most fiber-containing foods contain about one-third soluble and two-thirds insoluble fiber (Cummings 1981). The colonic fermentation results in the production of short chain fatty acids (SCFA) - acetic, butyric, and propionic acids (60:20:20) together with gases (CO₂, CH₄, and H₂) and heat (Topping & Clifton 2001).

The end products of fermentation depend upon the substrate utilized. For example, starch fermentation primarily yields acetate and butyrate, whereas fermentation of pectin and xylan yields acetate alone (Englyst, Hay, & Macfarlane 1987).

Once absorbed, SCFA are metabolized at 3 major sites in the body:

1) Caeco-colonic cells use butyrate for energy production

2) Uptake of these SCFA’s by the liver (for e.g. propionate used as a substrate for gluconeogenesis)

3) Oxidation of acetate to provide energy for the muscle cells
The generation of these SCFA’s promotes the growth of favourable microflora in the intestine such as bifidobacteria, reduce potential pathogenic clostridia, and decrease solubility of bile acids and increase absorption of minerals (Roberfroid 2005).

Butyrate acts as a major fuel for the colonocytes, their differentiation and proliferation. Although it stimulates normal colonic cell proliferation (Roediger 1982), it suppresses the multiplication of carcinoma cells (Bornet et al 2002). The decrease in pH caused due to formation of organic acids inhibit the bacterial enzyme 7α-dehydroxylase, which degrades primary bile acids to secondary bile acids that have potential tumor promoter activity (Grubben et al 2001, Thornton 1981)

Breakfast with barley or rye kernels have higher content of indigestible carbohydrates which undergo colonic fermentation and hence improve glucose tolerance for a prolonged period i.e. during the course of the whole day or overnight when replaced by standardized meals (Nilsson et al 2008)

Controversy surrounds the role of SCFA in reducing blood lipid levels. Some studies have stated, though inconclusively, that propionate may inhibit the utilization of acetate for cholesterol biosynthesis (Wolever, Spadafora, and Eshuis 1991). Decreased acetate: propionate ratio induced by inulin type fructans may confer hypolipidemic effects (Bush and Milligan 1971). Also acetate producing lactulose has shown to increase serum cholesterol levels due to enhanced hepatic lipogenesis (Jenkins et al 1991)
**Inulin:** Inulin is a naturally occurring polysaccharide found in plants such as onion, garlic, chicory, Jerusalem artichokes, and wild yam. It consists of fructose sugar (20 to several thousand) linked together by beta 2-1 glucosidic linkages classified as fructo-oligosaccharides or fructans. Its physical properties such as bland to slightly sweet taste and viscosity make it suitable for fat, sugar and flour replacement in different foods.

Inulin is a rich source of soluble fiber and also is a prebiotic. Its fermentation in the colon leads to formation of carbon dioxide, hydrogen and/or methane. Inulin fermented by microflora in the large intestine produce lactic acid and other short chain fatty acids (SCFA). These SCFA’s such as butyrate serve as fuel for the colonocytes, also called as the bifidogenic nature or prebiotic effect (Roberfroid et al 1998; Bouhnik et al 1999). Inulin consumption results in a stable butyrate producing colonic ecosystem (Perrin et al 2001) which is very vital to protection against risk of colon cancer.

AOAC suggested that resistant starch and inulin, which cannot be categorized as dietary fiber can be considered as ‘functional fiber’ since they exert reasonable beneficial physiological effects in humans (Slavin 2003).

Carbohydrates can be classified in several ways based on its structure, chemical bonds, functional and nutritional properties, etc. We shall now discuss the digestion and absorption of carbohydrates in the body to gain a better understanding of its physiological effects.
**Digestion and absorption of carbohydrates**

Carbohydrate digestion is initiated in the mouth. Mastication causes breakdown of the cellulose covering providing smaller particles for further digestion. The salivary amylase called ptyaline is a starch splitting enzyme that results in formation of some malto-dextrins. The enzyme action ceases in the acidic environment of the stomach as ptyaline functions best at pH 4-9.

Starch hydrolysis is mainly completed in the jejunum by the action of pancreatic alpha-amylase which hydrolyzes amylose to maltose and amylopectin to a mixture of maltose, iso-maltose and alpha-limit dextrins. Brush border disaccharidases initiate rapid breakdown of maltose and iso-maltose leading to significantly higher absorption of glucose from maltose than that from free glucose (Southgate 1995). The end products of carbohydrate digestion are mainly glucose, together with some fructose and galactose.

Carbohydrates reaching the caecum (large bowel) undergo fermentation to produce short chain fatty acids such as acetate, butyrate and propionate. “The amount and types of carbohydrate that reach the caecum are unknown, but are probably between 20 and 40 g/day in countries with 'westernized' diets, whereas they may reach 50 g/day where traditional staples are largely cereal or diets are high in fruit and vegetables” (Elia & Cummings 2007).

The rate of starch hydrolysis is dependent on several factors (Englyst & Kingman 1990)-

- Physical state- state of division with finely ground starches being hydrolyzed more rapidly than coarsely ground ones
- Types of granules- Raw cereal starches have a slower hydrolysis compared to cooked starches, while some others like raw Potato and banana are indigestible (resistant starch)
- Presence of cellular structures (outer covering) that inhibit enzyme access.

Absorption
Glucose absorption occurs via carrier-mediated active transport which is sodium dependent. The sites for co-transporter systems are highest and most active in upper jejunum and duodenum. Fructose is absorbed though carrier-mediated facilitated diffusion. The absorption of fructose from sucrose seems to be more efficient than absorption of fructose alone in presence of glucose (Riby, Fujiwasa & Kretchmer 1993).

Disaccharides are hydrolyzed before passing though the mucosal cell membrane. Hydrolysis of sucrose occurs easily in weak acidic conditions (in stomach) and may be attenuated by heating. Lactose and maltose are more resistant to acid hydrolysis. Brush border enzymes – disaccharidases facilitate hydrolysis of the disaccharides. Glucose derived from disaccharide hydrolysis seems to be absorbed more quickly than glucose alone which may be due to non-sodium dependent transporters for products of disaccharide hydrolysis. Lactase activity is low in adult humans. Lactase enzyme breaks down lactose to glucose and galactose which are then absorbed by active transport. Maltose and isomaltose are rapidly hydrolyzed by disaccharidases significantly increasing rate of glucose absorption from maltose than that from glucose alone.

Oligosaccharides such as Raffinose, stachyose, fructo-oligosaccharides cannot be hydrolyzed in the human small intestine and are passed on to the colon for fermentation.
The absorption of glucose and its appearance in blood determines the postprandial responses. Plasma glucose stimulates insulin secretion which further facilitates uptake of glucose by the tissues thereby reducing blood glucose levels (Antia & Abraham 1997).

Under normal circumstances carbohydrate intake is matched with carbohydrate oxidation as the carbohydrate stores represent only a small proportion of daily carbohydrate intake and there is no net *denovo* lipogenesis from carbohydrates (Schwarz et al 1995, Hudgins et al 1996). Excess energy from fat promotes greater storage in the body rather than excess intake from carbohydrates (Horton et al 1995). The thermic effect of carbohydrates is higher leading to increased energy expenditure. However, this effect may be altered after several weeks of overfeeding.

**Factors affecting starch digestibility**

Intrinsic factors such as physical structure, molecular distribution, physical state of food, and food anti-nutrients and extrinsic factors such as chewing, transit time of food, amount of starch present, and diet anti-nutrients determine enzyme activity and subsequent digestibility of carbohydrates (Luz Sdos et al 1997).

**Nature of starch**- The ratio of amylose: amylopectin impact the digestibly of starch to a great extent. The amylopectin being a branched structure with open chains has a much larger surface area per molecule and hence may be digested more quickly than amylose units. The strong intermolecular hydrogen bonds in amylose render it more compact leading to slow and gradual amylolysis.
Carbohydrate foods usually contain a higher proportion of amylopectin (70-75%) to amylose (25-30%) except for the high amylose varieties such as high-amylose cornstarch. The presence of thick cell walls of plant structures (eg. Legumes) make it mechanically resistant to enzyme hydrolysis by serving as a physical barrier that prevents complete swelling of starch granule during gelatinization (Tivar, Granfeldt & Bjork 1992).

Gelatinised starch provides more readily accessible glucose linkages for enzymes hydrolysis releasing more glucose units which are readily absorbed into the small intestine contributing to glycemia (Nantel 2003).

Urooj and Puttraj (1999) studied the digestibility index (DI) of foods based on sugars released after 3 hs incubation of foods with human saliva and porcine pancreatin. The degree of gelatinization and percentage of starch digested was also determined. The DI of Chapatti was found to be much lower than that of Rice flakes and was inversely correlated with protein, fat and energy. Higher proportion of insoluble and total dietary fiber was associated with lower percent starch digestion.

**Lipid-amylose complex**- Starch digestibility is reduced in presence of amylose-lipid complexes with monoglycerides and FFA having greater affinity to form complexes than triglycerides when added to amylose-rich mixture (Bhatnagar & Hanna 1994a). Amylose complexed with lipids displayed a substantially reduced susceptibility to α-amylase *in-vitro* but was hydrolyzed and adsorbed to the same extent as free amylose *in-vivo* though somewhat slower (Holm et al 1983). Cui & Oates (1999) observed that the bioavailability of native and freshly gelatinised sago starch was decreased in the presence of lipids, while retrograded starch lipid samples showed higher digestibility than starch control.
**Protein starch interaction**- The interaction between protein and starch in carbohydrate containing foods may influence its digestibility and glycemic response. Starch may also form insoluble complex with proteins as in case of Maillard’s reaction rendering it unavailable for digestion and absorption (Pi-Sunyer 2002).

**Anti-nutrients other than fiber**- In its native form, legumes mainly contain anti-nutrients such as phytic acid and polyphenols. Phytates and lectins may serve as enzyme inhibitors thereby producing hypoglycemia. The levels of these is reduced greatly with processing such as soaking, dehulling, fermentation and germination of legumes which in turn enhances the starch and protein digestibility (Yadav & Khetarpaul 1994, Preet & Punia 2000). Amylase and sucrase inhibitors are now being used in treatment of diabetes by reducing rate of carbohydrate digestion and absorption.

**Cooking method**- Uncooked raw starch retains the outer cell wall and fiber thereby resulting in reduced digestibility. Cooking causes disruption of cell wall, splitting the starch granules and increasing its susceptibility to enzyme action. Similarly, boiling and pressure cooking enhance digestibility compared to roasting.

During baking, starch undergoes gelatinization to various degrees depending on presence of adequate water, time and temperature conditions. In bread, the starch in the crumb undergoes complete gelatinization whereas the crust portion contains a mixture of starch in stages from slightly damaged starch granule to completely dispersed starch gel (Southgate 1995). Even upon gelatinization, food matrix may influence digestibility as seen in pasta products which
despite being gelatinized from moist cooking is digested slowly due to presence of dense food matrix (Thomsen et al 1994, Englyst et al 1999).

Amylose tends to form secondary structures hard to disperse both in starch granule and after processing affecting starch digestibility. However, Gallant et al (1992) have shown that food processing may negate the influence of amylose: amylopectin ratio.

Dry heat application in some dry baked products such as biscuits without using water results in starch granules remaining intact and therefore digested slowly (Englyst et al 2003)

Particle size, blending and grinding- The physical form of foods affects the starch digestibility. Whole foods which are minimally processed are slowly digested and absorbed compared to foods that are ground or blended before cooking. The resulting smaller particles provide a greater surface area for enzyme action. Thorne, Thompson & Jenkins (1983) showed that cooked beans that were ground later were more resistant to enzymatic hydrolysis compared to beans that were first ground and then cooked. The factors affecting resistance of starch to breakdown is given in Table 2.5.
Table 2.5: Factors contributing to resistance of starches

<table>
<thead>
<tr>
<th>Nature of the granule and starch</th>
</tr>
</thead>
<tbody>
<tr>
<td>Resistant granules (raw Potato and under-ripe banana with GI 30; overripe banana with GI 52 (Powell &amp; Miller 1996))</td>
</tr>
<tr>
<td>Raw starch especially starch with B-, but also C-type X-ray diffraction pattern</td>
</tr>
<tr>
<td>Starch which is encapsulated in the plant cell or plant tissues</td>
</tr>
<tr>
<td>Retrograded amylose (Asp &amp; Bjorck 1992) and amylopectin (Toufeili et al 1999)</td>
</tr>
<tr>
<td>Modified starch</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Processes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Canning of starchy foods such as Potatoes (Soh &amp; Brand Miller 1999) may cause retrogradation due to cooling after gelatinization.</td>
</tr>
<tr>
<td>Staling bread due to retrogradation</td>
</tr>
<tr>
<td>Competition for moisture: fiber retains moisture when cooking or baking cereal products (McWilliams 2001) and limits the available water required for gelatinization; sucrose retains water when cooking or baking cereal products and limits the available water required for gelatinization (Bennion &amp; Scheule 2000)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Complex formation during modification or formulations</th>
</tr>
</thead>
<tbody>
<tr>
<td>Amylose lipid complexes (Asp &amp; Bjorck, 1992)</td>
</tr>
<tr>
<td>Starch protein complexes (Vosloo &amp; Davel, 1991)</td>
</tr>
</tbody>
</table>

**Source:** Vosloo 2005

The digestion and absorption of carbohydrates is followed by uptake of the sugar released by the different tissues in the body. This uptake is facilitated by several hormones which help to maintain the plasma glucose concentrations within normal range.
Hormonal regulation of fuel metabolism

The minute-by-minute adjustments that keep the blood glucose level near 4.5 mM (81mg/dL) involve the combined actions of insulin, glucagon, and epinephrine on metabolic processes in many body tissues, but especially in liver, muscle, and adipose tissue (Nelson & Cox 2004).

Insulin signals these tissues that the blood glucose concentration is higher than necessary; as a result, the excess glucose is taken up from the blood into cells and converted to storage compounds, glycogen and triacylglycerols. Glucagon carries the message that blood glucose is too low, and the tissues respond by producing glucose through glycogen breakdown and gluconeogenesis and by oxidizing fats to reduce the use of glucose. Epinephrine is released into the blood to prepare the muscles, lungs, and heart for a burst of activity. Insulin, glucagon, and epinephrine are the primary determinants of the metabolic activities of muscle, liver, and adipose tissue.

Epinephrine Signals Impending Activity

When an animal is confronted with a stressful situation that requires increased activity-fighting or fleeing -in the extreme case, neuronal signals from the brain trigger the release of epinephrine and norepinephrine from the adrenal-medulla. Both hormones increase the rate and strength of the heartbeat and raise the blood pressure, thereby increasing the flow of O₂ and fuels to the tissues, and dilate the respiratory passages, facilitating the uptake of O₂.
In its effects on metabolism, epinephrine acts primarily on muscle, adipose tissue, and liver. It activates glycogen phosphorylase and inactivates glycogen synthase (by cAMP-dependent phosphorylation of the enzymes), thus stimulating the conversion of liver glycogen into blood glucose, the fuel for anaerobic muscular work.

Epinephrine also promotes the anaerobic breakdown of the glycogen of skeletal muscle into lactate by fermentation, thus stimulating glycolytic ATP formation. The stimulation of glycolysis is accomplished by raising the concentration of fructose-2,6-bisphosphate, a potent allosteric activator of the key glycolytic enzyme phosphofructokinase-1. Epinephrine also stimulates fat mobilization in adipose tissue, activating (by cAMP-dependent phosphorylation) the triacylglycerol lipase. Finally, epinephrine stimulates the secretion of glucagon and inhibits the secretion of insulin, reinforcing its effect of mobilizing fuels and inhibiting fuel storage.

**Glucagon Signals Low Blood Glucose**

Even in the absence of significant physical activity or stress, several hours after the intake of dietary carbohydrate, blood glucose levels fall to below 4.5 mM because of the continued oxidation of glucose by the brain and other tissues. Lowered blood glucose triggers secretion of glucagon and decreases insulin release. Glucagon causes an increase in blood glucose concentration in two ways.
i. Like epinephrine, glucagon stimulates the net breakdown of liver glycogen by activating glycogen phosphorylase and inactivating glycogen synthase; both effects are the result of phosphorylation of the regulated enzymes, triggered by cAMP.

ii. But, unlike epinephrine, glucagon inhibits glucose breakdown by glycolysis in the liver and stimulates glucose synthesis by gluconeogenesis.

Both of these effects result from lowering the level of fructose-2,6-bisphosphate, an allosteric inhibitor of the gluconeogenic enzyme fructose-1,6-bisphosphatase (FBPase-1) and an activator of phosphofructokinase-I. Glucagon also inhibits the glycolytic enzyme pyruvate kinase (by promoting its cAMP dependent phosphorylation), thus blocking the conversion of phosphoenolpyruvate to pyruvate and preventing oxidation of pyruvate via the citric acid cycle; the resulting accumulation of phosphoenolpyruvate favours gluconeogenesis.

By stimulating liver glycogen breakdown, preventing glucose utilization in the liver by glycolysis, and promoting gluconeogenesis, glucagon enables the liver to export glucose to the blood, restoring blood glucose to its normal level.

Although its primary target is the liver, glucagon (like epinephrine) also affects adipose tissue, activating triacylglycerol lipase by causing its cAMP-dependent phosphorylation. This lipase liberates free fatty acids, which are exported to the liver and other tissues as fuel, thus sparing glucose for the brain. The net effect of glucagon is therefore to stimulate glucose synthesis and release by the liver and to cause the mobilization of fatty acids from adipose tissue, to be used instead of glucose as fuel for tissues other than the brain. All of these affects of glucagon are mediated by cAMP-dependent protein phosphorylation.
During starvation, metabolism shifts to provide fuel for the brain

The fuel reserves of a normal adult human are of three types: glycogen stored in the liver and in muscle in relatively small quantities; larger quantities of triacylglycerols in adipose tissues; and tissue proteins, which can be degraded when necessary to provide fuel.

After an overnight fast, almost all of the liver glycogen and most of the muscle glycogen have been depleted within 24 hours, the blood glucose concentration begins to fail, insulin secretion slows, and glucagon secretion is stimulated. These hormonal signals result in the mobilization of triacylglycerols, which become the primary fuels for muscles and liver. To provide glucose for the brain, the liver degrades certain proteins. Their amino groups are converted into urea in the liver; the urea is exported via the bloodstream to the kidney and is excreted. Also in the liver, the carbon skeletons of glucogenic amino acids are converted into pyruvate or intermediates of the citric acid cycle. These intermediates, as well as the glycerol derived from triacylglycerols in adipose tissue, provide the starting materials for gluconeogenesis in the liver, yielding glucose for the brain.

Eventually the use of citric acid cycle intermediates for gluconeogenesis depletes oxaloacetate, preventing the entry of acetyl coA into the cycle. Acetyl-coA produced by fatty acid oxidation accumulates, favouring the formation of acetoacetyl-coA and ketone bodies in the liver. After a few days of fasting, the levels or ketone bodies in the blood rise as these fuels are exported from the liver to heart and skeletal muscle and the brain, which use them instead of glucose. The triacylglycerols stored in the adipose tissue of an adult of normal
weight provide enough fuel to maintain a basal metabolism for about three months; a very obese adult has enough stored fuel to endure a fast of more than a year. However, such a fast would be extremely dangerous; it would almost certainly lead to severe overproduction of ketone bodies, and perhaps to death. When fat reserves are gone, the degradation of essential proteins begins; this leads to loss of heart and liver function, and death.

**Insulin Signals High Blood Glucose**

When glucose enters the bloodstream from the intestine after a carbohydrate-rich meal the resulting increase in blood glucose causes increased secretion of insulin and decreased secretion of glucagon.

Insulin stimulates glucose uptake by muscle tissues, where the glucose is converted to glucose-6-phosphate. Insulin also activates glycogen synthase and inactivates glycogen phosphorylase, so that much of the glucose-6-phosphate is channelled into glycogen. As a consequence of accelerated uptake of glucose from the blood, the blood glucose concentration falls to the normal level, slowing the rate of insulin release from the pancreas. Thus there is a closely adjusted feedback relationship between the rate of insulin secretion and the blood glucose concentration. The effect of this regulation is to hold the blood glucose concentration nearly constant in the face of large fluctuations in the dietary intake of glucose.

Insulin also stimulates the storage of excess fuel as fat. It activates both the oxidation of glucose-6-phosphate to pyruvate via glycolysis and the oxidation of pyruvate to acetyl-CoA. Acetyl-CoA not oxidised further for energy production is used for fatty acid synthesis in the
liver, and these fatty acids are exported as the triacylglycerols of plasma lipoproteins (VLDL’s) to the adipose tissue. Insulin stimulates triacylglycerol synthesis in adipocytes, using fatty acids released from the VLDL triacylglycerols. These fatty acids are ultimately derived from the excess glucose taken from the blood by the liver. In summary, the effect of insulin is to favour the conversion of excess blood glucose into two storage forms: glycogen (in the liver and muscle) and triacylglycerols (in adipose tissue)
Dietary carbohydrate metabolism

The following four mechanisms may be involved in role of carbohydrates on metabolism:

i) Nature of monosaccharide absorbed

ii) Rate of absorption

iii) Amount of CHO consumed

iv) Colonic fermentation

Nature of monosaccharide absorbed-

The type of monosaccharide unit present will influence the glycemic response. Starch breakdown yields mainly glucose units and some maltose. Glucose has shown to elicit highest postprandial response followed by bread, sucrose and fructose (Lee & Wolever 1998). Though the blood sugar responses tended to flatten as the quantity increased from 50 to 100g, the insulin responses showed a somewhat linear increase. Fructose induced the least glycemic and insulinemic response due to its rapid clearance and metabolism in the liver (Crapo 1980). Wolever & Brand Miller (1995) suggested that fructose is converted to glucose in the liver slowly and only some of this glucose is released into circulation which results in lower glucose and insulin response than glucose.

Food sources of starch and sugars that are non-glycemic, slow-release naturally fiber-rich have more beneficial effects on health (Jenkins et al 2002) by reducing serum triglycerides and improving lipid metabolism (Granfeldt et al 1994)
Rate of absorption-

The nature of the starch and sugars, and the presence of vegetable proteins, fats, viscous fiber, and anti-nutrients, including lectins and phytates reduce the rate of absorption. It can also be manipulated by the use of specific enzyme inhibitors and by increasing the number and frequency of meals while holding caloric intake constant. Hence, slow rate of absorption can be achieved by use of the slow-release carbohydrates or "lente carbohydrates" which are effective in long term control and management of blood sugars. The rate of absorption of sugars is further affected by the form in which it is consumed, effects of individual food matrices on gastric emptying and physical properties of intestinal contents. It has been suggested that once the food is absorbed; transport and metabolism converge and the effect of dietary source of sugar is of little significance (Southgate 1995).

Increasing meal frequency or nibbling throughout the day tends to slow down the small intestinal absorption of carbohydrates. This has been associated with reduced postprandial insulin secretion and lower concentrations of low-density lipoprotein (LDL) cholesterol and apolipoprotein-B. Reducing rate of absorption also facilitates faster delivery of the undigested particles to the colon where they undergo fermentation to produce short chain fatty acids which may inhibit cholesterol synthesis in the liver (Jenkins et al 1995).
Amount of CHO consumed-

Jenkins (1983) showed that ingestion of equal carbohydrates quantities in the form of sugars or Potato starch lead to rapid absorption resulting in high postprandial glycemia and insulinemia likely to cause rebound hypoglycemia after a meal. This fall in blood sugars soon after carbohydrate-rich meal leads to desire to eat more food (Anand 1974). In NIDDM subjects, an increase in dosage of starch lead to the glucose response areas to meals of varying quantities of starch (20, 40, and 60 g carbohydrate as white Rice) to differ significantly (p <0.05) (Rasmussen 1993)

Colonic fermentation -

Colonic fermentation of the undigested carbohydrates leads to production of short chain fatty acids such as acetate, propionate and butyrate. The beneficial effects of resistant starch i.e. starch that resists digestion by alpha-amylases can be attributed to these SCFA’s. SCFA (from oligofructose and inulin) suppress FFA release, improve glucose tolerance and may promote increased insulin secretion (Wolever et al 1989, Wolever, Spadafora, and Eshuis 1991). Colonic propionate increase blood glucose concentrations by acting as gluconeogenic substrate (Woelver 2003). Scheppach et al (1988) showed that oral acetate made no detectable difference to glucose tolerance or to levels of free fatty acids, 3-hydroxybutyrate, lactate, insulin, glucagon and gastric inhibitory polypeptide.
Glycemic response to foods

Macronutrients in the diet impact the blood glucose response with carbohydrates exercising the greatest influence. Glycemic response may be largely determined by the rate of amylolytic digestion of carbohydrates in the small intestine (Jenkins et al 1987) leading to absorption of glucose and then its clearance from the blood stream. The rate of disappearance of glucose is largely influenced by insulin secretion and its action on target tissues (DeFronzo & Ferrannini 1982).

Meals that produce a high glycemic response tend to increase intake at subsequent meals. Flint et al (2006) tested effect of breakfast meals containing 50g carbohydrate with varying energy and macronutrient content on subsequent energy intake at lunch. It was observed that glycemic response was positively associated with energy intake at lunch whereas insulin response was unrelated to the same

Bornet et al (1987) studied glycemic and insulinemic response to starchy foods (white bread, lentils, beans, spaghetti, Potato and Rice) alone and as mixed meals with constant CHO(50g, 43%), fat (20g, 37%), protein (24g, 20%), energy (475kcal) based on 50g available CHO to 18 type 2 diabetic subjects. When fed alone the GI responses were in the decreasing order from bread (95±15), Potato (74±12), spaghetti (64±15), Rice (56±2), lentils (30±15) and beans (23±1). However the glycemic responses decreased by upto 20% with mixed meals. The insulin index followed the same hierarchy with lowest for beans (69±13), lentils (103±37), Rice (126±16), spaghetti (172±38), Potato (274±164) and bread (278±18).
Glycemic response curve

Figure 2.1: Glycemic response curve without hypoglycemia

Figure 2.2: Glycemic response curve with rebound hypoglycemia
The entry of glucose from gut and liver into peripheral circulation is reflected as initial rise in blood glucose levels upon consuming food (Figure 2.1). The events occurring in the gut lumen bring about changes in hormonal concentrations. Sometimes insulin secretion may rapidly bring down the blood glucose levels to below baseline seen as hypoglycemia (Figure 2.2).

Meal related stimuli including glucose, amino acids, gut hormones and neurotransmitters influence insulin secretion from the pancreas leading to fall in blood glucose levels (Morgan 1988). Insulin facilitates glucose uptake by respiring cells and tissues. Specialized sites for glucose transport within muscle and fat cells are insulin dependent. Rapid assimilation of carbohydrates stimulates secretion of temporary excess of insulin relative to need resulting in blood sugars levels to drop below fasting levels before returning to baseline.

As glucose is absorbed, small rise in blood sugars (not exceeding about 50 mg/100 ml blood) is observed during 1st half h of glucose ingestion. Afterwards, the rate of utilization exceeds the rate of absorption leading to fall in the blood sugar levels within 2h (Yudaken & Bloomberg 1973). Depending upon the specific carbohydrate with the nature of starch present, this utilization time may vary.
Preparation for Glucose tolerance test

- A liberal carbohydrate diet should be taken for at least 3 days before the test. This should be at least 300 g daily. A simple way of doing this is to instruct the patient to take a full diet plus extra carbohydrate with each meal, e.g. cereals, toast and jam at breakfast, Potatoes and Rice at lunch, and 'sweets' at dinner.

- All drugs should, if possible, be stopped for an arbitrary minimum of 3 days before the test. Oral contraceptives, which may raise blood sugar, should be stopped preferably for one cycle.

- No food, liquid or solid, should be taken for 8 - 16 h preceding the test, which should commence at 0800 - 0900 h. (Glucose tolerance tests should not be conducted in the afternoon, since diurnal variations in blood sugar may influence the results.)

- Patient should fast for 10 - 12h before the test. Subject should not take food after 2000 - 2200’ h on the night preceding the test.

- Alcohol should not be taken after the evening meal. Black coffee or tea, and smoking should not be allowed before or during the test though Water is permitted

Sources of blood:

Yudaken & Bloomberg (1973) suggested that venous blood samples be preferred over capillary blood samples for the following reasons

- Technical difficulties are less, results are more reproducible, variations between successive samples are less, collection of samples is more easily controlled, e.g. stasis of blood can be avoided, and plasma instead of whole blood can be analyzed.
The advantages of using plasma are that 'saccharoids', which produce false high sugar values, are mainly in erythrocytes; glycolysis is less active in plasma than in whole blood (when the specimen stands for some pours between sampling and analysis); and differences in haematocrit values (which may affect the results) are avoided.

The lower water content in erythrocytes as compared to plasma, results in the whole blood glucose being less than the plasma glucose (approximately 10 - 20 mg/dL). The difference should be borne in mind when assessing plasma glucose values.

Caution to be exercised with use of venous blood samples

- The circulation though the hand and forearms on cold and warm days
- The degree of stasis induced by the application of a tourniquet and aggravated by contraction of forearm muscles to distend the veins
- Possible differences in blood sugar levels in superficial and deep veins in the antecubital fossa, and elsewhere in the fore- arm or hand.

Capillary blood levels may be affected by differences in rate of blood flow though the fingers, by volume errors in pipettes, and by errors in transferring capillary blood into diluting solutions.

Glucose concentrations in arterial blood are much higher after a meal than in venous plasma due to peripheral tissue utilization of glucose leading to greater concentrations of glucose in capillary blood. Wolever & Bolognesi (1996) validated though their study that measuring glycemic response using capillary blood or venous plasma did not affect the GI of foods.
Factors affecting glycemic response to foods

The glycemic response is affected by not only the nutrient quantity but also its type, presence of non-nutrient chemicals - anti-nutrients and the unique physical arrangement within the food (Sud et al 1988). For example, legumes are high in fiber, protein, anti-nutrients, and slowly digested starch resulting in relatively small blood glucose rises in postprandial period (Thorne, Thompson & Jenkins 1983). These unique structural food matrix properties inherently present in unprocessed foods once lost cannot be restored by addition of fiber supplements also.

Particle size: Smaller particles provide greater surface area for enzyme action and are emptied more quickly from the stomach compared to larger ones (Heaton et al 1988, Read et al 1986). O’Dea et al (1980) reported higher 60 minute blood glucose responses to ground Rice (both white and brown) compared to the meals containing whole Rice. However, in a separate study using bread made with traditional white, conventional whole-grain wheat (WWF), or ultra-fine whole-grain wheat (UFWF) flour, the particle size did not seem to affect the glycemic area under the curve (Behall, Scholfield & Hallfrisch 1999).

Disruption of cell wall integrity: The physical and botanical structure of natural foods is responsible for their glycemic and insulin responses to foods (Bjorck et al 1994). The fibrous coating of legumes and seeds act as physical barriers against digestive enzymes thereby delaying digestion and lowering blood sugar response.

To elucidate the effect of change in food form on the satiety, plasma glucose and insulin response, Haber et al (1977) fed test meals of apples as juice, puree and whole fruit each
providing 60g carbohydrates to 10 normal subjects. The rate of ingestion was equalized for all. It was noted that although initial rise in plasma glucose was similar in all thee, the levels fell rapidly for juice followed by puree and then whole fruit. However, whole apples did not lead to a rise in insulin levels as much as juice and puree. Satiety ratings were also highest for whole fruit followed by puree and least for juice. With the decrease in fiber and modification of physical structure the insulin responses rather increased and satiety decreased. This can have important metabolic implications in diabetic patients in the long term.

Source of sugar: The source of sugar plays a role in determining blood glucose response. Lee & Wolever (1998) observed highest glycemic response to glucose followed by white bread, sucrose and fructose but no disproportionate increase in postprandial insulin response with change in source. Carbohydrates in fruits and fruit juices (mainly fructose) have shown to produce glycemic responses similar to that produced by sucrose but lesser than refined starchy carbohydrate containing foods (Wolever & Miller 1995). Fructose has a sparing effect on glucose metabolism since fructose 6 phosphate gets converted to glucose replacing rather than adding to glucose production (Gurr 1995).

In healthy individuals, the chain length of glucose does not impact the rate of rise and fall in blood glucose concentration, the dietary form in which starch is administered is more important determinant of the glycemic response (Wahlqvist et al 1978). When same amounts of starch and glucose were administered it was observed that in the first 1 h postprandial period blood sugar rise after starch was only slightly less than that of glucose but the total glycemic effect was much larger (i.e. hyperglycemia was more prolonged) in case of starch.
(Koehler, Rapp & Hill 1934). When carbohydrate load containing 50 gm. of glucose was administered to subjects as drink or with other nutrients as a meal, it was observed that though glucose and sucrose induced similar plasma glucose response, the plasma insulin response curve elicited by sucrose was 20% higher than that of glucose (Crapo, Reaven & Olefsky 1976). This increased insulin response may be attributed to the presence of some percentage of fructose in sucrose which stimulates insulin with little effect on plasma glucose (Delarue et al 1993).

Miller and Lobbezoo (1994) noted that sucrose-free Puffed Rice cereal containing rapidly digestible starch demonstrated the highest plasma glucose and insulin response compared to cereal samples containing added sucrose (21g and 43g) despite keeping the available carbohydrate constant indicating that sweetened breakfast cereals may not compromise glycaemic control more than the unsweetened counterpart.

Reaven et al (1979) showed that attempts to reduce carbohydrate intake lead to increased calorie intake from fat. However administration of carbohydrate as slowly digestible starch lowers the glucose and insulin responses compared to an equivalent amount of glucose administered as either dextrose or sucrose. Slowly digested starchy foods blunt the gut hormone responses and prolong the suppression of free fatty acid and ketone body production. These factors along with enhanced production of short chain fatty acids due to passage of indigestible components into the caecum favourably modify the carbohydrate and lipid metabolism in the postprandial period (Jenkins et al 1987). Hence the traditional starchy foods with increased fiber in the form of resistant starch producing a flatter glycemic response may be promoted over fat in the regular diets. This concept also strengthens the
recommendations of diabetes association, heart foundations and cancer institute that recommend increase in carbohydrate intake from starch, by reducing fat intake (Jenkins et al 1987).

**Method of cooking or processing:** Starch can be hydrolyzed at pH of 4 or low upon heating and gelatinization (Bennion & Schedule 2000) which result in formation of low molecular compounds that increase blood glucose response (Cummings et al 1997). Gelatinization of starch granule leads to loss of birefringence, disruption of the ordered structure, increased soluble starch fraction and thereby increased susceptibility to enzyme hydrolysis. Holm et al (1988) found that the rate of hydrolysis by alpha amylases *in vitro* is positively correlated with the glycemic and insulin responses.

Some factors may hinder complete gelatinization of starch such as:

- low processing temperature
- low moisture content
- enclosure of ingredients in intact botanical structure
- amylose-lipid complex
- surface area of granule (Bjorck et al 1994)

Soaking can improve *in vitro* starch digestibility for horse gram and moth beans, whereas sprouting decreased the resistant starch content and increased the Glycemic Index greatly (Bravo, Siddhuraju, & Saura-Calixto 1998). When the glycemic response to freshly cooked, frozen and re-warmed foods was compared, it was seen that freshly cooked Rice and Potato produced the highest glycemic response than the refrigerated and re-warmed forms. The
lower glycemic response to refrigerated food can be attributed to the development of resistant starch (Kanan et al 1998).

**Starch bioavailability:** The modern methods of processing lead to a loss of nutrients and fiber rendering foods more readily digestible and absorbable. A shift to pre-agricultural diets emphasizing increased fiber intake from minimally processed cereal grains with intact plant cell wall structures (Sánchez-Castillo et al 2002) may go a long way in prevention of the new-age lifestyle disorders. Gastric emptying time is found to be inversely related to plasma glucose and insulin response at 120 min (Horowitz et al 1993). The amount and type of macronutrients in a meal, meal volume, food particle size, viscosity and pH affect the gastric emptying time (Rayner et al 2001). Different starchy foods such as mashed Potatoes, bread, Rice and spaghetti were fed to 12 healthy young adults to examine relationship between gastric emptying (GE) time and glycemic response. It was found that GE half time had a significant negative correlation with blood glucose response with fastest emptying of mashed Potatoes followed by bread, then Rice and slowest for spaghetti (Mourot et al 1988).

Physical characteristics of starch present influence extent and rate of hydrolysis by enzymes. Legumes, containing high amylose release less malto-triose and more maltose + glucose than bread, hence showing less digestibility and lower blood glucose response (Jenkins et al 1982).

Sugars and oligosaccharides released *in vitro* upon hydrolysis of 50 g carbohydrate portions of cooked lentils and soya beans were only 39% and 8% of wholemeal bread. Lentils (42%)
and soyabeans (14%) caused small rise in blood glucose levels compared to bread due to slow rate of digestion in vivo (Jenkins et al 1980). Mixing of cereal flours lead to lower readily digestible starch (RDS) and starch digestion index (SDI) than observed in single cereal flours. Amylose content influences these factors in foods hence it is suggested that characterization of carbohydrate containing foods based on its digestibility can be facilitated by a simple in vitro measurement of the amylose and amyllopectin fractions (Aarathi, Urooj & Puttaraj 2003). High amylose containing Rice (23-25%) showed significantly lower glycemic response and insulin at 30 min compared to 0% amylose Rice. This effect is attributed to differential enzymatic hydrolysis of amylose and amyllopectin and the presence of lipid-starch complexes (Goddard, Young & Marcus 1984). Amylose lipid complexes lead to slower digestion due to reduced rate of amylolysis (Holm et al 2006) which in turn lowers the glycemic response.

Yet another observation was that cellulose when added to glucose did lower the glycemic response but an elevation in insulin response was observed. Glucose when combined with pectin produced lower response compared to glucose with cellulose.

**Presence of fat and protein:** Dietary fat may delay the blood glucose response by slowing down glucose absorption (Collier & O’Dea 1983, Collier, McLean, O’Dea 1984, Nuttall & Gannon 1991) whereas amino acids (proteins) act as insulinagogue and augment glucose clearance from circulation (Floyd et al 1970, Nuttall et al 1984, van Loon et al 2000). The presence of fat delays gastric emptying and thereby lowers blood glucose response. An increase in the carbohydrate: fat ratio leads to higher peak glucose and insulin responses and
significantly increases peak area under the postprandial glucose curve (van Amelsvoort et al 1989).

Saturated fat such as butter causes a delay in blood glucose response but does not alter area under the curve significantly; however, unsaturated oils such as olive and corn oil totally blunted the glucose response. The insulin responses did not differ significantly (Gatti et al 1992). Shah et al (2007) studied the effect of test meals (1,000 kcal each) rich in various fatty acids such as palmitic acid, linoleic acid, oleic acid, and eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) on glycemic and insulin response of 11 type 2 diabetic subjects. Although a significant difference for insulin response was observed between the test meals, plasma glucose responses did not differ much. Increasing dose of fat within normal range of intakes (17-44% of energy) does not alter the Glycemic response significantly in healthy subjects (Owena & Wolever 2003)

Protein ingestion along with carbohydrate has an effect on lowering postprandial blood glucose area and increasing the insulin response area compared to protein free meal however the amount of protein consumed (dose) may not influence the extent of response upto 2 h period. Insulin levels also do not reach baseline levels within 2h (Spiller et al 1987) of consuming protein-rich meal.

**Oxidative damage:** The state of acute hyperglycemia in postprandial period is associated with generation of free radicals which induce oxidative damage. Efforts towards reducing postprandial glucose spikes and hyperglycemia can help in prevention of cardiovascular disease (CVD) (Antonio 2005). The role of nuts such as almonds in reducing risk of CHD
can be attributed to its antioxidative effect and decrease in the glycemic excursion postprandially (Jenkins et al 2006).
Glycemic index and its limitations

In 1981, Dr David Jenkins from University of Toronto in Canada proposed the concept of Glycemic Index (GI) concept to classify carbohydrates qualitatively based on their glycemic response induced in the body. GI is expressed as the percentage increase in blood glucose, produced by specific amount of available CHO in a test food as compared to the same weight of available CHO from a reference food such as glucose. GI provides “numeric physiologic classification of relevant carbohydrate foods” (Jenkins 2002). Initially glucose was used as standard which was then replaced by white bread in order to make more meaningful comparisons with real foods. GI relates to rate of digestion and entry of glucose into systemic circulation (Schenk et al 2003).

\[
\text{GI}_{\text{carb}} = \frac{\text{Incremental blood glucose response to 50g available carbohydrate in food}}{\text{Incremental response to glucose equal in weight to the food carbohydrate}} \times 100
\]

Scientists who emphasize the importance of carbohydrate quality suggest that it is a vigorous measurement tool (Wolever et al 2003), helps predict the glycemic response to mixed meals (Wolever and Jenkins 1986), is easy to implement and follow (Brand Miller, Colagiuri, Foster-Powell 1997), and produces consistent improvements in glycemic control when used in diabetics (Miller 1994). In contrast, the supporters of carbohydrate quantity argue that GI is highly variable (Pi-Sunyer 2002), not physiological (Gannon & Nuttall 1987), cannot reliably predict mixed meal responses (Coulston & Reaven 1997, Hollenbeck, Coulston & Reaven 1986), difficult to learn and follow (Franz 2001). The American Diabetes Association (ADA)
(2003) in their most recent evidence based guidelines maintained their position that carbohydrate quantity is a more important consideration than quality, dismissing the value of the GI in diabetes therapy. In contrast, most other major International diabetes organizations have interpreted the available data differently, supporting the application of the GI concept in the management of diabetes in their most recent guidelines. These organizations include the Canadian Diabetes Association (CDA) (Wolever et al 2000), Diabetes Australia (DA) [2003], Diabetes UK [2003], and the European Association for the Study of Diabetes (EASD) [2000].

The glycemic index is useful in characterizing the quality of carbohydrates beyond dietary fiber. Low GI, high unavailable CHO interventions improved insulin sensitivity in persons with type 2 diabetes, overweight and obese subjects (Geoffrey et al 2008b). They may help reduce quantity of available CHO and metabolizable energy along with a variable increase in unavailable CHO and proteins without increasing fat levels (Livesey et al 2008a). High GI/GL diets may lead to hyperinsulinemia >100pmol/L in some overweight or obese persons but not all (Geoffrey et al 2008b).

The amount of carbohydrate accounts for 57-65% of the variability in glucose response while 60% variability resulted from glycemic index or the source of carbohydrate. Together they accounted for ~90% of the total variability in blood glucose response (Wolever and Bolognesi 1996). One study showed that the outcomes of blood glucose response on consuming high or low GI foods were mixed indicating that amount of carbohydrate may be more important than the source (Franz 2001).
The major limitation for the practical use of classification of CHO on the basis of GI is that it is calculated on the basis of equal amount of CHO where as in actual food, quantification of CHO is difficult, as in the same quantity of food, there are other macro and micronutrients present with different moisture content. As GI denotes an absolute value, it fails to respond to the changes in the amounts of carbohydrate consumed (Monro 1999).

The concept of “Glycemic load” (GL) comes into the picture here. Introduced by the researchers at the Harvard School of Public health in 1997, Glycemic Load is calculated as the product of the Glycemic Index and the amount of carbohydrate in a serving. It serves as an aid to determine overall glycemic effect of a diet.

\[
\text{Glycemic Load (GL)} = \frac{\text{Glycemic Index} \times \text{CHO content of food /serving}}{100}
\]

Glycemic load has been implicated as an independent risk factor for diabetes (Salmeron et al 1997, 1997a), cardiovascular risks and certain types of cancers. It may also be correlated to other risk factors such as HDL cholesterol, triglycerides and C - reactive protein levels (Liu et al 2002). In a study conducted by Beulens et al (2007), it was reported that high GL of food is an independent causal factor for the incidence of cardiovascular attack (CVA) and coronary heart disease (CHD) but GI alone increased the CHD risk. This shows the importance of the amount of carbohydrates consumed along with the form of CHO. Thus characterization of carbohydrates is necessary in order to avoid unnecessary curtailment of carbohydrates in the diet and proper management of postprandial glycemic effect.

But GL is a mathematical expression and practical use is still limited. For example, watermelon, with a high GI value indicating the types of sugar present in watermelon is very
hyperglycemic in nature but by virtue of its high moisture content, has a low GL, and hence can be consumed in moderate amounts without causing a drastic increase in blood sugar levels. Daly (2003) proposed that the term glycemic load may be mistakenly taken to represent the overall 24 h glucose supply though its connotation as “the glycemic load of a diet”. This can be misleading because glycemic load is defined by only 2 h postprandial definition of GI. The studies that proposed negative impact of high GL diets seem to be designed such that starches provided major component of Glycemic load rather than sugars.

While initially being propagated as an effective dietary tool for the management of diabetes, the concept of GI has still remains under constant scrutiny and doubt with respect to its reliability and consistency. The GI is influenced by the inherent characteristics of carbohydrate containing foods such as fiber, moisture content, presence of fat, proteins, etc. however, its applicability is undermined by the very fact that the absolute figures derived have little meaning for the diabetic unless it boils down to actual quantity of food as a whole. Hence we see that GI needs to be further developed and standardized. Low GI foods are likely to be high in fiber hence most of the metabolic merits of low GI diets can be attributed to the higher fiber content (Björck & Elmståhl 2003) rather than just available carbohydrate content.
Limitations of GI

Glycemic Index (GI) is calculated using 50g available carbohydrate portions analyzed chemically but is poorly predictive of direct physiological response. This phenomenon was exemplified by work of Jenkins et al (1981) wherein glycemic responses for 56 test foods were different even for foods within the same food group.

When carbohydrates with different glycemic indices are incorporated into conventional meals, the glycemic responses differ significantly. Coulston, Hollenback & Reaven (1984) suggested that since neither the normal nor diabetic individuals consume carbohydrate containing foods alone but as a part of mixed meals, great caution needs to be exercised before propagating the use of GI in clinical practice for diabetes management. In general, the glycemic response to mixed meals can be predicted with some accuracy by summing up the glycemic index of the component foods (Wolever et al 1985, Gulliford, Bicknell & Scarpello 1989), although not all studies have found a direct relationship between calculated and measured glycemic index of mixed meals (Coulston et al 1984, Hollenbeck, Coulston, & Reaven 1986, Laine et al 1987).

Differences in fat, protein and calorie content in context of mixed meals act as confounding variables influencing glycemic responses. These are not accounted for by the GI as comparison of mixed meals is also based on available carbohydrate content with variable amounts of fat, protein, and calories. When meals composed of Rice, Potato and lentils were fed at 50g available carbohydrate with differing energy distribution, the area under the curve
of glycemic responses were similar for all which showed that GI is a poor indicator of glycemic response to mixed meals (Calle-Pascual et al 1988)

In vitro study comparing effect of 50g available carbohydrates from legumes (lentils and soya) versus bread on postprandial glycemia showed that although legumes contain slowly absorbed carbohydrates, glucose released from digestion of legumes were cleared by the tissues more efficiently in later stages of glucose absorption leading to a flat and prolonged response. However, no alteration in area under the curve compared to bread was observed (Jenkins et al 1982). This shows that the use of available carbohydrate as comparative measure has limited responsiveness and applicability in terms of type of carbohydrate source.

Four carbohydrate-equivalent meals (50 g available CHO) of french fries, boiled Potatoes served with and without addition of oil, and white wheat bread (reference) reported no differences in subjective satiety. However, French fries resulted in a significantly lower glycemic response (glycaemic index (GI=77) than boiled Potatoes either with or without addition of oil (GI=131 and 111, respectively) (Leeman, Ostman & Björck 2008). This shows that GI may not be directly associated with satiety.

Siddhu et al (1992) have shown that foods containing largest number of nutrients have lowest glycemic response reiterating the fact that glycemic response of natural foods is not predicted based only on carbohydrate content but overall structural and nutritional composition.
To determine whether Glycemic Index or Glycemic Load have an effect on heart failure (HF) hospitalization or death, nine year follow up study on 36,019 women 48–83 years old without baseline HF, diabetes, or myocardial infarction was conducted. It was observed that although GL was somewhat related to HF, neither GI nor GL showed statistically significant association with heart failure in this population (Levitan, Mittleman & Wolk 2010).

Dietary Guidelines Advisory committee, (2010, May 12th) rejected the assertion that glycemic index and/or glycemic load are associated with body weight gain, cancer, or type 2 diabetes. It concluded that, when selecting carbohydrate foods, there is no need for concern about glycemic index or glycemic load.

Properties of GI/ limitations

1) In order to allow direct comparison of glycemic responses to foods using GI, they must contain equal quantities of carbohydrates.

2) GI represents a static absolute value indicating glycemic potency as percentage of that of glucose i.e. relative to standard and not independent of it. E.g. effect of 50g available carbohydrate dose relative to 50g glucose. This intrinsic inflexibility and lack of adaptability makes it non-responsive to changing food intakes.

3) Effect of adding, replacing or substituting the glycemic ingredient (available carbohydrate) with non-carbohydrate (protein, fat, etc) or non-glycemic (fiber) ingredient will not be observable as a change in the GI value because available CHO values may remain constant. However, the actual glycemic potency of the food product may increase or decrease due to the above.
This can be exemplified by the study reported by Monro (2002) wherein glycemic impact of hypothetical test meal with calculated GI was compared with meal to which additional serving of carbohydrate food was added. Meal GI was calculated as the sum of average GI weighted by proportion of carbohydrate provided by each food. A cup of porridge added to the test meal decreased the glycemic index rather than increasing the AUC indicating its lack of sensitivity to changes in carbohydrate doses.

**Available carbohydrate calculations for GI**

Available carbohydrate for GI determination was calculated as sum of sugars plus starch in food tables, the sum of sugars plus starch- digestible under analytical conditions, or carbohydrate by difference. None of the above is same as glycemic carbohydrates.

Glycemic carbohydrates need to be defined more clearly-

Is it the carbohydrate that would be glycemic if digestive enzymes gain access to it or if digesta diffusion occurred properly? But then how can it be distinguished from dietary fiber. The use of the term glycemic carbohydrates may be considered as suboptimal because not all carbohydrates induce glycemia and some are only fermented in the colon (FAO1997). The compounded error involved in GI calculation i.e. glycemic carbohydrate and its blood glucose response and use of carbohydrate value with GI again for calculation of Glycemic Load (GL) often result in under or overestimation of available carbohydrate content of foods.
The analytically determined 50g available carbohydrate content used for GI is for raw food portions however, the bioavailability of that calculated amount of available carbohydrate when it undergoes processing such as cooking, boiling, etc is not taken into consideration.

For many years, controlling available carbohydrate has been the cornerstone of diabetes management. Often, the available carbohydrate analysed in lab is not the same as Cho available in the gut upon consuming food. The actual glycemic response depends not only on the amount of potentially available CHO consumed but also on how rapidly it is digested, absorbed and disposed in the body which in turn depends on a myriad of factors including food structure and influence of other food components that vary in importance from food to food.

GI is not very effective in comparing effect of serving of one food with a serving of another because most common servings do not contain same amount of available CHO (Monro 2002) so it is not simple for consumer to use. Jenkins et al (2002) admitted that with its limited application, GI can be assumed to be a tool to make the consumers aware of the starchy foods that they did not eat otherwise but over a period of time with new food choices, the concept may become redundant especially for low-energy foods with high carbohydrate content. For e.g foods like carrots has high available carbohydrate but is also rich in minerals, vitamins and fiber making it a very healthy food to be encouraged in the diet even though it is classified as high GI food.

Although GI is a good indicator of the glycemic nature of carbohydrates present in a particular food, factors such as dose response and co-existing intrinsic and extrinsic properties are not taken into account. GI reflects the glycemic potency of the carbohydrate
component in food only and not the direct response to finished food product consumed as a whole.

Therefore, it has been now suggested that the glycemic expressions be extended to include (Monro 2005)
- Quantity of food consumed
- Proportion of CHO in it
- Glycemic potency of the entire food
GI\textsubscript{food}: Relative Glycemic Impact & Relative Glycemic Potency

Definition of GI and GI\textsubscript{food}

According to the Oxford English Dictionary “Index” is defined as a number or a value that expresses physical property, etc in terms of a standard (Simpson & Weiner 1989). In the present context, the Glycemic Index is supposed to be the value that expresses the glycemic potency (physical property) of food in terms of a standard (glucose or bread).

Monro 2005 proposes that the GI is inadequately or inappropriately defined:

i) the definition does not provide the information that glycemic potency of the available CHO in food and not entire food is being expressed

ii) it does not indicate that glucose is being used as a reference

When people use the term “GI of a food” it leads to misunderstanding with regards to the utility of the Index as a value for practical dietary management tool.

A better representation of GI could be as

\[
\text{GI}_{\text{carb}} = \frac{\text{Incremental blood glucose response to glycemic carbohydrate in food}}{\text{Incremental response to glucose equal in weight to the food carbohydrate}} \times 100
\]

For expressing GI based on foods, we may rather use

\[
\text{GI}_{\text{food}} = \frac{\text{Incremental blood glucose response to a specified food}}{\text{x 100}}
\]
Incremental response to glucose equal in weight to the food

This $GI_{food}$ serves as a more accurate definition such that

i) the definition provide the information that glycemic potency of the entire food is being expressed

ii) it indicates that glucose is being used as a reference

**Relative Glycemic impact (RGI): $GI_{food}$ for specified amount (e.g.50g)**

The ad hoc committee established by American Association of Central Chemists (AACC) (2004) in defense of glycemic carbohydrates coined the term “glycemic impact” to provide a measurable definition that effectively conveys the glycemic response of whole foods (gm/serving) to the consumers. In 2005, Dietary Guidelines Advisory Committee also recognized the concept of Glycemic impact of foods, whereby impact on blood sugar could be determined by feeding identical portions of foods.

“Glycemic impact is the weight of glucose that will induce a response equivalent to that induced by a given amount of food” (Miller- Jones 2007).

The term impact emphasizes the two important properties:

- Effect of single intake
- Associated acute postprandial response

The relative postprandial glycemic response of given food quantity consumed in a single intake compared to standard (glucose or bread) will reflect its glycemic potency and help to rank food based on their relative glycemic impact (RGI) (Monro & Shaw 2008). People eat
foods and not just the CHO in it hence RGI is a more helpful way of communicating
glycemic impact of foods.

\[
\frac{\text{GI}_{\text{food}} \text{/ RGI}}{\text{IAUC response to glucose equal in weight to the food}} = \frac{\text{IAUC blood glucose response to a specified food}}{\text{IAUC response to glucose equal in weight to the food}} \times \text{Amt. of food}
\]

IAUC: Incremental Area Under the Curve

RGI = GGE intake (a single intake event)

**Relative glycemic potency (RGP): GI\text{food for 100g}**

The ‘RGI’ terminology is used to represent acute effects of single intake (Monro 2002). In
general, when describing the nature of foods at multiple intake levels we may use the term
‘Relative glycemic potency (RGP)’. Relative glycemic potency (RGP) of a food is the
glycemic response to 100g of food as a percentage of the effect of equal amount of reference
food, bread wherein the amount of bread that gives similar response as that of 100gm of test
food is expressed as GBE/100g.

RGP is defined as the glycemic impact of 100g food as a percentage of the effect of an equal
amount of glucose/bread.

\[
\frac{\text{GI}_{\text{food}} \text{/ RGP}}{\text{IAUC response to glucose equal in weight to the food}} = \frac{\text{IAUC blood glucose response to a specified food}}{\text{IAUC response to glucose equal in weight to the food}} \times 100
\]

IAUC: Incremental Area Under the Curve
It expresses the amount of glucose that could be equal to 100g food in its glycemic impact (Monro 2005). In other words, the amount of glucose (reference) required to give the same glycemic response as a relevant amount of food, thus providing an equi-glycemic measurement. Hence it is rightly termed as the Glycemic Glucose Equivalent (GGE) (Monro & Shaw 2008).

RGP is a way of comparing foods on equal (100g) food weight basis to allow relative glycemic potency of foods to be seen immediately. RGP can be used to obtain a value for the RGI of any weight of food as its content of GGE.

RGI is the number of GGE donated by a food item or by meal.

GGE derived from RGI/RGP is the weight of glucose that would induce the same glycemic response as a given weight of food (Monro & Williams 2000). Similarly, if bread is used as standard Glycemic Bread equivalent (GBE) can be calculated as the weight of bread that would induce the same glycemic response as a given amount of test food. Salmeron et al (1997) suggested the GGE serves as a measure of exposure to glycemia associated with a diet.

RGI= GGE/intake or GGE/amount of food

RGP = GGE/100g of food or GBE/100g of food

If a serving of food has glycemic impact equivalent to that of 15g glucose, then that food serving is said to have GGE content of 15g. Likewise, if the GBE value for a given quantity
of test food is 60, then it can be predicted that 60g of bread would induce glycemic response equivalent to that quantity of food.

![Figure 2.3: Glycemic impact expressed as glycemic glucose equivalent/ intake (Monro & Shaw 2008)](image)

The Figure 2.3 shows that sweet muffin has a glycemic impact equivalent to that of 46 g glucose, and the apple's glycemic impact is equivalent to that of 7.9 g glucose.

The GGE/ GBE values are expressed in gram units unlike GI which is an absolute value with no units. Representation in terms of grams makes it suitable for inclusion as a distinct nutritive value in the food exchange list along with other nutrients. This will enable a direct selection of the right quantities of whole foods to induce known response, which is an easier concept to understand.

GGE expresses glycemic potency per unit weight of foods rather than per unit weight of available carbohydrates hence it is capable of responding to effect of substituting non-glycemic or non-CHO ingredients for glycemic ones in food formulations. Though GGE, the
property of foods—glycemic potency is expressed without any assumptions as to the food component responsible

GGE values express glycemic potency as if it were a food component. It has been termed as a virtual food component (Monro 2004) because

- It is responsive to food intake
- Can be expressed per serving like other nutrients
- Used for weight-to-weight comparisons

GGE as a virtual food component expressing RGI in terms of weight of glucose causing a glycemic response equivalent to given food weights; data for muesli, serving size approx. 65g

Table 2.6: GGE for one serving and 100g of muesli

<table>
<thead>
<tr>
<th>Nutrient</th>
<th>Per 65g</th>
<th>Per 100g</th>
</tr>
</thead>
<tbody>
<tr>
<td>Energy (kJ)</td>
<td>1040</td>
<td>1600</td>
</tr>
<tr>
<td>Protein (g)</td>
<td>5.9</td>
<td>9.1</td>
</tr>
<tr>
<td>Fat (g)</td>
<td>5.1</td>
<td>7.8</td>
</tr>
<tr>
<td>Total carbohydrate (g)</td>
<td>46</td>
<td>70</td>
</tr>
<tr>
<td>GGE (g)</td>
<td>18</td>
<td>28</td>
</tr>
</tbody>
</table>

Source: Monro 2005
The Table 2.6 above shows that GGE values are consistent with other food values in the food exchange tables and can serve as a virtual food component. If a food label for 65g of muesli bar states that GGE is 8 then it clearly spells out that 65g of muesli bar will have the glycemic impact equivalent to 8g of glucose.

Monro (2006) emphasizes that “the AACC definition of glycemic impact of foods requires a change in thinking from:

- Food CHO to whole foods
- Static index values to intake sensitive values
- Unit-free values (% relative to glucose) to nutrient like values (in grams)”

The various properties of Glycemic Index and Glycemic index food with respect to their advantages and disadvantages have been presented in Table 2.7 below.

Table 2.7: Comparison of GI and GI<sub>food</sub> definition and application

<table>
<thead>
<tr>
<th>PARAMETER</th>
<th>GLYCEMIC INDEX</th>
<th>GI&lt;sub&gt;food&lt;/sub&gt; (GLYCEMIC GLUCOSE EQUIVALENT)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Predictive validity</td>
<td>Predicts the effects of individual food choices on postprandial glycemic response up until equal CHO portions are involved</td>
<td>Predicts the effects of individual food choices on postprandial glycemia under condition in which foods are selected and consumed</td>
</tr>
<tr>
<td>Reliability</td>
<td>If available carbohydrate is replaced by non available ingredient the GI will not change since it is based on available carbohydrate content only</td>
<td>If isomaltose or resistant starch (non available ingredient) replaces a proportion of available carbohydrate the glycemic potency will show a change because effect of whole foods are compared</td>
</tr>
<tr>
<td>Accuracy</td>
<td>With GI, Intra individual variation is high Determination of available CHO may be inaccurate, especially when CHO is calculated by difference</td>
<td>With GI&lt;sub&gt;food&lt;/sub&gt; No need for available CHO estimation</td>
</tr>
</tbody>
</table>

70
<table>
<thead>
<tr>
<th>PARAMETER</th>
<th>GLYCEMIC INDEX</th>
<th>$\text{GI}_{\text{food}}$ (GLYCEMIC GLUCOSE EQUIVALENT)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Safety</td>
<td>Even within food groups such as breads and breakfast cereals differences in CHO content and portion sizes cause overlap in high, medium and low GI categories. It is difficult to exchange a portion of high GI product for a low GI product and substantially increase glycemic impact.</td>
<td>To ensure safety, expression of glycemic potency should indicate effect of food rather than food component.</td>
</tr>
<tr>
<td>Ease of Use</td>
<td>With practical food intake differing in composition and serving sizes, consumer needs to first adjust GI for carbohydrate content.</td>
<td>Here, the carbohydrate content and food quantity are already taken into account. This makes food selection easier for the consumer.</td>
</tr>
<tr>
<td>Understandability</td>
<td>GI ranks foods based on glycemic impact of the available CHO in foods and not food per se. Hence, food choices based on GI can be incorrect in predicting glycemic responses.</td>
<td>$\text{GI}_{\text{food}}$ represents glycemic potency of given quantity of whole cooked food and shows effect of food intake. It is therefore, more understandable than GI.</td>
</tr>
<tr>
<td>Flexibility and Robustness</td>
<td>GI requires use of food quantities that provide 50g of available carbohydrates. It has been observed that taking into account the non-carbohydrate components including water the total amount of food required to provide 50g or 100g available carbohydrate is too large for consumption (Monro 1999). This level of intake is impractical as humans do not consume such large quantities at one time.</td>
<td>$\text{GI}_{\text{food}}$ involves weight-to-weight comparisons and can be used for prediction of glycemic responses for different serving sizes.</td>
</tr>
</tbody>
</table>
APPLICATIONS OF GI\textsubscript{food} (Monro & Shaw 2008)

- A separate column in food table or in nutrient information panel stating the glycemic effect of equal or different amounts of foods relative to glucose or bread

- Since GI\textsubscript{food} is a direct function of food quantity, the influence of changing intake levels can be understood

- GI\textsubscript{food} being representative of whole foods, these values can be summed to predict responses of combined foods for practical range of intake. Liu et al (2003) reported a proportionate increase in glycemic response with an increase in GGE upto 60g food intake. However intake of >100g carbohydrate in noodles suppressed the GGE by upto 10%

- Food exchanges can be established by grouping foods with similar glycemic responses together

- The GI\textsubscript{food} dose responses could be studied in conjunction with blood glucose monitoring and insulin-delivery in order to build database for nutrient management system.

- GGE allows RGP to be treated as a nutrient because it represents the role of all thee factors that govern glycemic responses to foods (Monro 2002)-
  
  (a) Quality (GI),
  
  (b) Content (GL) and
  
  (c) Intake
Dose response

There have been very few studies that reflect effect of changing level of food intake on glycemic response using GGE. Liu et al (2003) showed that foods with different GI and carbohydrate content fed at same GGE doses produced similar glycemic responses and the responses doubled with doubling of the GGE dose.

At food intake levels ≤60g, no marked association was observed in glycemic response based on GGE and carbohydrate intake. However, GGE response was suppressed by about 10% with intake >100g carbohydrate in noodles. Similar findings were made by Brand-Miller et al (2003) wherein glycemic response was found to be a linear function of the GL upto intake >100g carbohydrate indicating dose response relationship.

At levels >50g, a plateau is reached and linear relationship between food intake and glycemic response ceases (Wolever & Bolognesi 1996). However calculations have shown that linear relation exist form 25 to 100g carbohydrate intake for most samples, plateau may manifest due to truncation of IAUC at 2 h.
Calculation of Incremental Area Under the Curve (IAUC)

IAUC is only a crude method of correlating rate of physiological responses to glycemic impact. Measurements of glycemic impact are truncated at 2-3 h post meal and hence may not truly represent rate of carbohydrate digestion which may increase at a later stage (Monro 2005).

The best method for measuring IAUC is not known clearly, trapezoid method including calculation of all net increments over baseline at zero time by adding positive and negative trapezoids between time points until 2-3 h time limit seems to be the most accepted (Wolever 1991). The GI is measured as the incremental area under the blood glucose curve relative to fasting levels as distinct from the total area under the glycemic response curve (Wolever and Jenkins 1986). However a physiologically more complete measure of glycemic response should cover “rebound hypoglycemia”. This idea has been discarded considering the fact that summation of areas above and below the line may lead to ambiguities. Hence, the two could be calculated and reported separately.
**Insulin**

Insulin is a biosynthetic hormone involved in carbohydrate, protein and fat metabolism. Insulin increases glucose uptake into the cells, promotes glycolysis and glycogenesis. It promotes protein synthesis by increasing amino acid uptake by tissues. Insulin is involved in hepatic synthesis of lipids from carbohydrates and storage of triglycerides in adipose tissue and down regulates the receptors for fatty acid oxidation. Increased insulin concentrations following a meal strongly inhibit lipolysis and fatty acid release. Postprandial Hyperinsulinemia poses the risk of increased fat synthesis and deposition and may lead to development of insulin resistance (Haber et al 1977).

**PHASES OF INSULIN SECRETION**

Insulin secretion is mainly regulated by glucose concentrations. It has been reported that glucose not only stimulates release of newly synthesized insulin but also promotes synthesis of the hormone by increasing beta cell RNA content.

Insulin secretion occurs in a biphasic manner (Curry 1970)

1\(^{st}\) phase – It involves release of insulin granules which are already energized and docked inside the beta cells and lasts for 2 min (high insulin content bordering cell membrane). This phase of insulin secretion accounts for only about 5-10% of the total insulin released after a meal. It is comprised of pre-stored hormone only (Curry 1968)

2\(^{nd}\) phase- Docking and energizing of the granules are part of this slow second phase. There is a shift of granules to the readily released pool (insulin transported from Golgi apparatus and ER and then secreted). It is the major contributor to insulin release. Even with a steady
supply of glucose, the 2nd phase of insulin secretion is characterized by increasing rate of insulin secretion.

Maximum insulin levels are reached approximately 60 min after a mixed meal. Factors that inhibit protein synthesis may reduce the second phase insulin response indicating that this phase is related to release of stored hormone as well as newly synthesized hormone.

**Insulin and food intake**

Insulin at physiological levels has shown to reduce food intake (Schwartz 2000) and high GI diet is in fact associated with lower insulin resistance (Kiens & Richter 1996). Several studies have shown that feeding high carbohydrate diet resulted in increased insulin sensitivity as compared to low carbohydrate diets (Thompson, Hayford & Darney 1978, Fukagawa et al 1990, Chen, Bergman & Porte 1988) Beta cell exhaustion purported as a cause for type 2 diabetes (Ferrannini 1998) is considered to result from long standing insulin resistance (Martin et al 1992, Lillioja et al 1993). There is need to show more evidence regarding high carbohydrate diets leading to insulin resistance and subsequently to beta cell exhaustion. However, a lower incidence of diabetes has been reported amongst high carbohydrate consuming population in certain countries despite increased insulin demand (Pi-Sunyer 2002).

With an increase in amount of carbohydrate consumed from 0 to 100g the insulin responses increase at a greater rate than plasma glucose response indicating that insulin response is more sensitive to changing doses (Lee & Wolever 1998).
While glycemic response is responsible for only 23% variability in insulin response other factors such as osmolality, gastric emptying, gut hormone release, and viscosity of gut contents (Holt, Miller & Petocz 1997); antecedent diet and degree of obesity (Bagdade, Bierman & Porte 1967); age (Barrett-Connor et al 1996); and even sex (Nuttall et al 1985)

**Factors affecting insulin release**

Insulin responses have been assumed as being proportional to glycemic responses and hence have been less explored for its specific responses based on food composition. However, Service et al (1983) proposed that in order to plan treatment strategies for overall control of meal related glycemia knowledge of factors affecting both glycemic and insulin responses need to be studied.

Glucose stimulated release of insulin is potentiated by several insulinotropic factors such as fructose, certain amino acids, fatty acids, and GI hormones such as GIP, glucagon and cholecystokinin (Nutall & Gannon 1991, Morgan 1992). In a study by Holt, Miller & Petocz (1997) to determine insulin index of foods using 1000kJ portions of common foods it was observed that highly refined bakery products and snack foods which typically constitute the Western diet induced substantially more insulin secretion per kJ or per gram of food than more traditional diets based on less refined foods. Only 33% of the variation in insulin responses for the 38 foods examined could be explained. In addition to Glycemic responses which are significant predictors of insulin response accounting for 23% variability in insulinemia; other macronutrients such as protein or fat, water, sugar and starch were also found to be significant predictors, together accounting for 10% of the variability.
The degree of postprandial insulin secretion is also influenced by rate of starch digestion, the amount of rapidly available glucose and resistant starch, the degree of osmolality, viscosity of gut contents and rate of gastric emptying (Holt, Miller & Petocz 1997)

**Amino acids** - In the presence of amino acids, glucose-induced Insulin secretion is potentiated substantially in healthy (Rabinowitz et al 1966) and diabetic subjects (Berger & Vongraya 1966). The amino acids from various protein sources added to carbohydrates may induce variable glycemic and insulinemic response in healthy subjects by affecting insulin secretion or insulin extraction rates. Specifically leucine and arginine have shown to have stimulatory effects on insulin secretion, not synthesis. Amino acids such as alanine, glycine and arginine also activate insulin secretion by causing depolarization of the beta cells. However, it is postulated that glucose should be present in order to exert their effect. A protein rich meal has differential effect on metabolism by increasing insulin as well as glucagon secretion. The underlying reason for this phenomenon is that the excess amino acids remaining after fulfilling normal requirements are diverted to gluconeogenesis in the presence of glucagon to convert amino acids to glucose while insulin, which is also active in the postprandial period stimulates uptake of this glucose in muscle tissue and its storage as glycogen.

In a comparison of insulin responses to meals containing different sources of protein such as milk (cottage cheese), soya protein (soy protein isolate) or cod with same macronutrient composition, it was observed that meal providing milk protein produced the largest area under the curve (AUC) at 240 min, higher concentrations of insulin:C-peptide and increased insulin:glucose ratios (von Post-Skagegård et al 2006).
Milk products are generally considered as low GI, but milk has shown to produce higher Insulin Index in healthy subjects (Östman, Elmståhl & Björck 2001, Liljeberg & Bjorck 2001, Schezenmeir J et al 1989). Milk protein, especially the whey fraction seems to exert insulinotropic effects. Whey contains insulin secretagogue such as specific insulinergic acids and bioactive peptides either originally present or formed as products of digestion which are responsible for stimulating insulin secretion (Nilsson et al 2004).

Whey ingestion increases the plasma concentrations of incretin hormones such as Glucose dependent insulinotrophic peptide (GIP) and Glucagon like peptide 1 (GLP-1) which has insulinotropic properties (Nutrition Research Newsletter 2005). Nilsson et al (2004) observed that a mixture of leucine, isoleucine, valine, lysine, and theonine could mimic whey in producing similar glycemic and insulimetic responses even in the absence of an additional effect of GIP and glucagon-like peptide 1.

Legumes produce lower insulin response than glucose in normal control subjects compared to NIDDM patients wherein Insulin responses to legumes were found to be much higher than that produced by glucose (Vishwanathan et al 1989). “Legumes form an excellent supplement to cereal diets where they improve the biological value of protein by mutual supplementation of lysine from legumes and methionine from cereals” (Vishwanathan et al 1989).

**Fat-** Fat is considered to decrease glucose and insulin response in postprandial period due to reduced upper gastric motility (Welch et al 1987). However, Collier et al (1988) showed that fat may have an acute impact on increasing insulin secretion as it potentiates secretion of gastric inhibitory polypeptide.
**Degree of fat saturation**- With an increase in degree of saturation and chain length, the insulinotropic potency of individual fatty acids decreased. It is proposed that fatty acids may increase K⁺ flux out of the beta cell, increasing polarization and reducing insulin release. In type 2 diabetes patients, the postprandial rises in glucose, insulin, and GIP was attenuated while GLP-1 release was stimulated due to ingestion of fat before a carbohydrate meal (Gentilcore et al 2005). Fat induced release of GLP-1 slows down gastric emptying and stimulates insulin secretion which may be responsible for lower the glycemic response. MacIntosh, Holt & Brand-Miller (2003) observed that “substitution of unsaturated fats for saturated fatty acids had no acute benefits on postprandial glycemia, insulin demand or short-term satiety in young men”. In another study, palmitic and linoleic acid were shown to elicit significantly higher insulin responses compared to meals rich in oleic, DHA and EPA (Shah et al 2007).

**Carbohydrate**- Carbohydrates constitute 40-50% of the total energy intake in most Western countries. It is not known whether type of CHO affects insulin action at this level of CHO intake (Kiens & Richter 1996). At total CHO intake of 43-44% sucrose exerts deleterious influence on insulin sensitivity in healthy humans (Reiser et al 1979). At 46% CHO intake levels, slowly absorbable carbohydrates induce a lower whole body insulin action at high plasma insulin concentration. Kiens & Richter (1996) observed that initially low GI diets produced lower blood glucose and plasma insulin concentrations but after 30d of the diet, these differences ceased to exist.
**Fiber content**- Soluble fiber seems to have greater effect on reducing postprandial insulin response while resistant starch exerts greater effects on reduction of glucose response (Behall et al 2006). The effect of high fiber intake as breakfast cereals among normal and hyperinsulinemic men showed that lower glycemic responses in both groups was observed with high fiber cereal but reduction in insulin responses observed only in hyperinsulinemic subjects (Wolever et al 2004). In a study conducted on postprandial insulin, glucose and incretin responses to grain products such as rye breads, pasta, white wheat bread, it was concluded that different botanical and structural characteristics factors determined the postprandial insulin response rather than amount of fiber or the type of cereal. Incretin hormones such as GIP and GLP-1 may also get altered due to food form and structure (Juntunen et al 2002).

High carbohydrate high fiber diets have shown to lower fasting plasma glucose and insulin levels in healthy individuals; compared to usual western diets (Anderson & Chen 1979, Fukagawa et al 1990). In healthy young and older men, intake of High carbohydrate high fiber diets for 21-28 days increased peripheral insulin sensitivity compared to habitual diet (Fukagawa et al 1990).

**Cation concentration**- Extracellular calcium ion concentration is quantitatively related to insulin secretion (Grodsky & Bennet 1966). Potassium may also cause release of insulin in absence of normal stimulatory agent (Howell & Taylor 1968). High magnesium levels have an inhibitory effect on insulin release which may be due to competition between calcium and magnesium for common active site on beta cell membrane (Milner & Hales 1967).
Temperature- The beta cell membrane has an inherent property of suppressed metabolic activity when tissue is subjected to cold temperatures (Curry 1968).

GI hormones – Several gastrointestinal hormones tend to cause moderate increase insulin secretion such as gastrin, secretin, CCK, serotonin or enteroglucagon and gastric inhibitory peptide (Creutzfeldt 1973)

Age- Aging is accompanied by progressive impairment in glucose tolerance and thereby glucose induced insulin release. This may be attributed to decreased beta cell sensitivity to GIP which plays a role in stimulating insulin release from pancreas in state of hyperglycemia (Meneilly et al 1998).

Particle size- Finely ground flour produces a higher insulin response compared to whole or cracked grains as observed for isocaloric wheat based meals in normal healthy volunteers (Heaton et al 1988).

EFFECT OF INSULIN ON CARBOHYDRATE METABOLISM

Wolever & Bolognesi (1996) observed that 90% variances in insulin responses to mixed meals is mainly influenced by the amount of carbohydrate and the glycemic index of the meal in normal healthy subjects. Carbohydrate intake (as a percentage of total energy) has been found to be inversely correlated with insulin sensitivity (i.e. total carbohydrate and sucrose are positively correlated with insulin resistance) (Daly 2003). Persistent high glucose
levels stimulate high insulin secretion which can in turn cause deterioration of beta cell function and impaired insulin sensitivity (Wolever 2000).

Prolonged hyperglycemia and hyperinsulinemia results in decreased expression of the rate-limiting enzymes and alters the potential for fat oxidation in the long term. Reduced capacity to oxidize fatty acids is present in some obese human subjects (Simoneau et al 1999) and is linked with greater weight gain in several prospective studies (Zurlo et al 1990, Weyer et al 2000). In normal weight subjects, regular post meal elevation of insulin did not signal satiety (Woo, Kissileff & Pi-Sunyer 1984).
**Insulin resistance (IR) / decreased insulin sensitivity**

Organs such as muscles, liver and adipose tissue can absorb glucose only in the presence of insulin. Insulin secreted from the pancreas in response to increased blood glucose concentrations bind to the cell membranes at specific receptor site. This initiates series of post-receptor changes which make the cells permeable to glucose. In obese individuals, insulin resistance may develop due to

- Post receptor defects in glucose metabolism
- Abnormality in glucose transport systems (ex. Reduced GLUT4 mRNA) or
- Failure of Golgi body transporters to translocate glucose across plasma membrane

The insulin receptors or its functions are “down regulated” as a part of normal self regulation in response to high serum insulin levels in healthy individuals. In obese subjects, this capacity is impaired leading to persistently high circulating insulin levels increasing risk of diabetes and cardiovascular diseases. Hyperinsulinemia has been recognized as an independent risk factor for ischemic heart disease in men (Després et al 1996).

Bonora (2005) reported that 30-40% of normal subjects from general population are insulin resistant which acts as an independent risk factor for cardiovascular diseases. Insulin insensitivity is characterized by decreased insulin stimulated glucose transport and metabolism in muscle, adipose tissue and decreased suppression of hepatic glucose output due to impaired insulin signaling in the tissue.

At the level of adipose tissue, insulin has an anti-lipolytic effect however, in insulin resistance this effect is lost resulting in excessive release of free fatty acids and glycerol in
the blood which can have deleterious effect on glucose metabolism as well. The increased availability and utilization of FFA may lead to skeletal muscle insulin resistance. The intramuscular triacylglycerol concentrations are important determinants of muscle insulin sensitivity.

Genetic makeup and environmental influences play a large role in the manifestation of insulin sensitivity in healthy individuals. Insulin resistance and decreased pancreatic β-cell function caused by genetic or acquired abnormalities may result in development of Type 2 diabetes mellitus. IR or decreased insulin sensitivity is detected even before diabetes sets in and was found to cluster in some NIDDM families (Martin et al 1992). Thus it has been proposed that susceptibility to develop NIDDM may be partly inherited (Daly 1997).

Insulin action is affected by several environmental influences such as obesity (Bonadonna et al 1990), especially visceral obesity (Bjorntorp 1988) which are negatively correlated while weight loss and physical activity improve insulin sensitivity (Mikines et al 1988, Richter et al 1989). Himsworth (1935) first recognized the role of IR in development of type 2 diabetes and later it was known to be associated with hypertension and ischemic heart diseases as well (Avogaro & Crepladi 1965, Reaven 1988).

Several disorders associated with IR including type2 diabetes, cardiovascular disease and metabolic syndrome is attributed mainly to changes in dietary pattern and decreased physical activity. Higher consumption of fiber containing food such as whole grains, pulses and vegetables may help to reduced risk of developing insulin resistance. Lau et al (2005) noted that the incidence of IR was not directly associated with habitual intake of high glycemic
index and high glycemic load diet or with a high content of total carbohydrate (including simple sugars) but was inversely related to dietary fiber intake.

Insulin sensitivity increases with higher intakes of whole grains as dark breads, high fiber and cooked cereals as reported in the Insulin Resistance Atherosclerosis Study (IRAS Exam I, 1992-1994) (Liese et al 2003). Fasting insulin which is considered as a measure of insulin sensitivity has shown to have an inverse relation to whole-grain intake as observed by two epidemiologic studies (Pereira et al 1998, McKeown et al 2002).

Swinburn et al (1991) showed that high versus low carbohydrate diet led to marked and significant improvement in oral glucose tolerance and significant reduction in fasting plasma glucose. Although no significant change in insulin sensitivity was observed, significant improvements in glucose effectiveness (the ability of glucose to stimulate its own removal) and in pancreatic responsiveness (plasma insulin response after intravenous glucose injection) was noted suggesting that carbohydrate intake has more important effect on pancreatic function and other combined factors than directly on insulin sensitivity alone Storlien et al (1996) evaluated effects of dietary fats on insulin sensitivity and provided evidence based conclusion supporting importance of pattern of fatty acid consumption much as the quantity of fat.

Men who consume Western style diets which include intake of not only sweets and desserts, but also high saturated fat (red meat, processed meat, French fries, high-fat dairy products), known to be associated with decreased insulin sensitivity (Storlien et al 1997) and a high glycemic load (refined grains) have an increased risk of developing diabetes (van Dam et al
2002). Clearly sweets and desserts cannot be claimed to be the sole causative factors because fructose has a low GI, and sucrose has a lower GI than even Potatoes and white bread. Sevak et al (1994) have shown that greater intake of sucrose and not starch is associated with insulin resistance. Sucrose has shown to increase insulin by 20% compared to fructose containing meal alone.

The reports on effect of fructose and sucrose intake on insulin sensitivity have been debatable. Some controlled studies of specific carbohydrates showed that consumption of sucrose or fructose at high levels (> 15% of dietary energy from fructose and > 33% of dietary energy from sucrose) led to increased fasting or postprandial insulin concentrations (Reiser et al 1979, Reiser et al 1981, Beck-Nielsen, Pedersen & Lindskov 1980) while some others showed no effect (Dunnigan et al 1970, Mann & Truswell 1972, Thorburn et al 1990). Studies using the latest euglycemic clamp technique have not shown a negative effect of sucrose on Insulin sensitivity. Increased sucrose intake (>33% of total energy) (Reiser et al 1981) may alter insulin sensitivity but such high dietary doses are never consumed in the practical setting by humans due to poor palatability (Daly 2003).

Vinegar ingestion has shown to improved whole-body insulin sensitivity during the 60-min postmeal interval and reduced postprandial insulin fluxes significantly in insulin-resistant subjects and slight improvement in type 2 diabetic subjects as well. The underlying mechanism may be the suppression of disaccharidase activity and increased glucose-6-phosphate concentrations in skeletal muscle induced by acetic acid thus having physiological effects similar to acarbose or metformin (Johnston, Kim & Buller 2004).
In healthy humans, caffeine may result in elevated plasma epinephrine levels thereby decreasing insulin sensitivity (Keijzers et al 2002)

In a randomized cross over study by Farshchi, Taylor & Macdonald (2005) on effect of breakfast consumption on energy intake, energy expenditure, and circulating insulin, glucose, and lipid concentrations in healthy women, it was concluded that women who omitted breakfast showed higher total energy intake, significantly higher fasting total and LDL cholesterol and higher area under the curve of insulin response to the test meal compared to women who ate breakfast.
**Insulin index and Insulin score**

A U-shaped relationship exists between both fasting and 2 h postprandial insulin responses and all-cause mortality with low and high concentrations being associated with an increased risk, independent of other risk factors (Balkau & Eschwège 1999). Hence it is important to study insulin secretory response to foods and classify them accordingly.

It has been proven that the increase in plasma glucose concentration is attributed not only to the rate of disappearance of glucose but also on the postprandial hyperinsulinemia following the ingestion of carbohydrate containing foods. This was demonstrated by feeding corn flakes (CF) and bran cereals (BC) containing 50 grams of available carbohydrate in 6 healthy men and blood sugars were observed for 180 min. Results showed that the glycemic response of CF was more than twice that of BC since the postprandial hyperinsulinemia occurred earlier with BC than with CF (Schenk et al 2003).

Foods that stimulate excess insulin secretion pose the increased risk of reduced beta cell function due to amyloid deposition resulting in high blood glucose levels. Further deterioration of beta-cell function and insulin sensitivity via glucose toxicity occurs (Wolever 2000). There is substantial evidence to show that insulin responses may not always be parallel to glucose response. Insulin responses to protein rich foods or bakery products (rich in fat and refined carbohydrate) are disproportionately higher than their glycemic responses (Holt et al 1997). Several other studies also showed that addition of protein or fat to a carbohydrate rich meal increases insulin secretion without increasing blood glucose concentrations (Krezowski et al 1986, Collier, McLean, & O’Dea 1984).
Service et al (1983) proposed that a system for ranking or grouping of foods based on their insulin responses can be useful in discerning the etiology and treatment of NIDDM. Holt et al (1997) developed an insulin score calculated for 38 foods separated into 6 food categories (fruit, bakery products, snacks, carbohydrate rich foods, protein rich foods and breakfast cereals) by dividing the insulin AUC value for test food by the insulin AUC values for white bread (the reference food) and expressed as a percentage.

\[
\text{Insulin score} \, (\%) = \frac{\text{AUC} \, 120\text{-min insulin response for 1000kJ test food}}{\text{AUC the 120-min insulin response for 1000kJ white bread}} \times 100
\]

This equation is similar to that developed by Wolever & Jenkins (1986) for calculating GI but only difference is that test foods were not served based on 50g available carbohydrate portion. The postprandial insulin responses to isoenergetic portion (1000kJ) of range of common foods were compared and the insulin score was calculated as insulinemic effect relative reference food. The table 2.8 given below has been abstracted from their study.

**Table 2.8: Nutritional composition of test foods per 1000kJ serving from Australian food tables* or manufacturers data**

<table>
<thead>
<tr>
<th>Food</th>
<th>Serving weight</th>
<th>Fat</th>
<th>Protein</th>
<th>Sugar</th>
<th>Starch</th>
<th>Fiber</th>
<th>Water</th>
<th>Energy density</th>
<th>Insulin score</th>
</tr>
</thead>
<tbody>
<tr>
<td>White bread*</td>
<td>94</td>
<td>2.1</td>
<td>8.5</td>
<td>1.8</td>
<td>44.1</td>
<td>3.3</td>
<td>36.1</td>
<td>10.6</td>
<td>100±0</td>
</tr>
<tr>
<td>White Rice*</td>
<td>203</td>
<td>0.5</td>
<td>5.0</td>
<td>0.1</td>
<td>56.0</td>
<td>0.4</td>
<td>140.0</td>
<td>4.9</td>
<td>79±12</td>
</tr>
<tr>
<td>Potatoes#</td>
<td>368</td>
<td>1.0</td>
<td>10.0</td>
<td>3.1</td>
<td>45.9</td>
<td>9.2</td>
<td>290.8</td>
<td>2.7</td>
<td>121±11</td>
</tr>
</tbody>
</table>

The results show that energy density of Rice is low (Table 2.8) and consuming upto 200g of Rice would induce an insulin response of 79%. Similarly, since Potatoes provide only...
2.7kJ/kg, we need to consume 368g to attain 1000kJ. However, one may not consume that quantity of Rice or Potato at one time. Now, for determining the insulin score for a lesser quantity of Rice, iso-caloric comparison with bread would have to be made again.

In practical setting, it may not be possible each time to use iso-energetic food portion to calculate the insulin score. We suggest that the definition of GI<sub>food</sub> can be applied to insulin as well for comparing area under the 2 h (120 minute) insulin response curve of equal quantities of test foods and reference (white bread).

The Insulin Index<sub>food</sub> can be calculated by dividing the insulin response elicited by a specific quantity of test food to insulin response produced by the same quantity of standard such as glucose or bread. It can be expressed as the Insulinemic Bread Equivalent (IBE)

\[
\text{II}_{\text{food}} = \frac{\text{Incremental serum insulin response to a specified food}}{\text{Incremental response to glucose equal in weight to the food}} \times 100
\]

\[\text{II}_{\text{food}} \text{ value for 50g} = \text{Relative Insulinemic Impact (RII)} = \frac{\text{IBE}}{\text{amount of food (e.g.50g)}}\]

\[
\text{II}_{\text{food}}/\text{RII} = \frac{\text{IAUC serum insulin response to a specified food}}{\text{IAUC response to glucose equal in weight to the food}} \times \text{Amt. of food}
\]

IAUC- Incremental Area Under the Curve

\[\text{II}_{\text{food}} \text{ value for 100g} = \text{Relative Insulinemic Potency (RIP)} = \frac{\text{IBE}}{100\text{g of food}}\]
Hence IBE/100g food is the insulin response of an amount of glucose/bread equivalent to 100g test food. For example, the IBE content of 100g Potatoes is 29g; it means that 100g Potato would induce similar glycemic response as 29g of bread.
Common carbohydrate rich foods in Indian diet

Cereals are considered as rich and least expensive source of dietary carbohydrate. The CHO present is predominantly in the form of starch with small amounts of free reducing sugars. Starch is the most abundant polysaccharide in cereals and constitutes a bulk energy and nutrient resource in human diet at affordable price (Zobel & Stephen 1995). Cereal grains contain 40-90% starch on dry weight basis, while pulses contain 30-70% and tubers 65-85% (Shelton & Lee 2000).

Starch is composed of amylose and amylopectin units of which 98.5% is alpha-glucans. Amylose and amylopectin are glucose polymers linked together by alpha-1,4 linkages but amylopectin also contains 4-5% of alpha 1-6 linkages making it appear as a branched molecule. Cereal amylopectin have an average chain length of 20-26 glucose units (Cura & Krisman 1990)

Cereal starches have an amylose content of 20-30% varying with botanical structure, climatic and soil conditions during grain development (Champagne 1996). Cereals grains contain small amount of free reducing sugars about 1-2% only. The levels may increase upon subjecting the grain to different processing techniques such as germination to obtain malted cereals. The amylose and amylopectin ratios and starch granule size of some common carbohydrate containing foods are given in Table 2.9
Table 2.9: Starch composition and granule size of common carbohydrate food

<table>
<thead>
<tr>
<th>Food Product</th>
<th>Amylose (%)</th>
<th>Amylopectin (%)</th>
<th>Ratio</th>
<th>Granule size^</th>
<th>Range(µm)</th>
<th>Avg. size(µm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wheat flour, whole</td>
<td>25</td>
<td>75</td>
<td>1:30</td>
<td></td>
<td>3-35</td>
<td>25-40; 5-10</td>
</tr>
<tr>
<td>Rice, raw, milled</td>
<td>18.5</td>
<td>81.5</td>
<td>1:4.4</td>
<td></td>
<td>2-15</td>
<td>5</td>
</tr>
<tr>
<td>Potato, boiled</td>
<td>20</td>
<td>80</td>
<td>1:4.0</td>
<td></td>
<td>10-70</td>
<td>40</td>
</tr>
<tr>
<td>Sago*</td>
<td>24-31</td>
<td>76-69</td>
<td>1:3.7</td>
<td></td>
<td>30</td>
<td></td>
</tr>
</tbody>
</table>


Food grains are subjected to different processing treatments before being used for consumption. These techniques tend to alter both, the macro and micro nutrient bioavailability as well as phytochemicals of nutraceuticals value (Mahadevamma & Tharanathan 2007).

**WHEAT**

India is second largest producer of wheat in the world, averaging an annual production of 65,856 TMT. On average, India consumes 65,283 TMT of wheat, ranking them as the second largest consumer of wheat in the world. Wheat is the second – largest cereal crop cultivated globally after maize, third being Rice. The largest producer of wheat in India is Uttar Pradesh (35%) followed by Punjab (22%). Haryana, Rajasthan, Madhya Pradesh, Gujarat and Bihar are also considered as major wheat growing areas. Wheat forms the staple food for people staying in North.

Classes of wheat used in the United States ([http://www.ers.usda.gov/briefing/wheat/background.htm](http://www.ers.usda.gov/briefing/wheat/background.htm)) are

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• **Durum** - Very hard, translucent, light colored grain used to make semolina flour for pasta.

• **Hard Red Spring** - Hard, brownish, high protein wheat used for bread and hard baked goods.

• **Hard Red Winter** - Hard, brownish, very high protein wheat used for bread, hard baked goods and as an adjunct in other flours to increase protein.

• **Soft Red Winter** - Soft, brownish, medium protein wheat used for bread.

• **Hard White** - Hard, light colored, opaque, chalky, medium protein wheat planted in dry, temperate areas. Used for bread and brewing

• **Soft White** - Soft, light colored, very low protein wheat grown in temperate moist areas. Used for bread.

The starch content of Wheat is 63-72% (Pomeranz & MacMasters 1968) with starch being concentrated in the endosperm of wheat kernel (75-80% of endosperm on dry weight basis). The starch and protein content of wheat kernels seem to be inversely related with the hard wheat varieties (high protein content) having lesser starch than soft wheat varieties with low protein content (Shelton & Lee 2000). The different varieties such as Hard Red Spring (HS), Hard Red Winter (HW), durum, soft, etc demonstrate a narrow range of amylose content (23.4 – 27.6%) as noted by Medcalf & Gillis (1965). The amylose content of waxy wheat starch has been found to be negligible (Yasui et al 1996) ranging from 1.2 – 2.0% with biochemical features similar to those of waxy maize (Graybosch 1995).

Starch granules in wheat exhibit a bimodal size distribution with two basic forms: small spherical granules (B) and large lenticular granules (A). The B type small granules are
spherical and have a diameter of less than 10 µm with mean value of 4 µm. The large lenticular A type granules have a mean diameter of 14 µm.

Raw untreated wheat starch is crystalline with shorter chains to give an A pattern. However when starch is heated in excess water, the conformational changes result in amorphous and less ordered arrangement of atoms with diffraction of the x-ray pattern. During retrogradation the B pattern gains predominance. Hence gelatinised and stored starch contains the B pattern. These B type granules almost resemble Rice starch granules in size.

Wheat kernels contain free sugars in the form of monosaccharides (glucose, fructose and galactose), disaccharides (sucrose and maltose) and trisaccharides (glucodifructose and raffinose) (Saunders 1978). Wheat flour has 0.57-0.80% sucrose, 0.54-0.70% raffinose, 0.02-0.04% fructose and 0.02-0.03% glucose (Henry 1985).

Wheat grains are used to make flour, livestock feed and as an ingredient in the brewing of beer. The husk separated from wheat known as wheat bran is used as cattle feed but is now finding large application as a source of supplemental fiber. Application of wheat starch is mainly in baking industry and for manufacture of adhesives, confection and canning industries.
Wheat products

Chapatti-

Various studies and researches show that wheat and wheat flour play an increasingly important role in the management of India’s food economy (http://agro.indiamart.com/agricultural-commodities/wheat.html). Wheat kernels undergo milling and grinding to variable extent with production of wheat bran, wheat flour, broken wheat, semolina, durum wheat, etc. at every advanced step of processing more nutrients are lost, especially fiber. In India, Hard red, Hard white, and Durum are used to prepare wheat flour which are considered to have higher protein and lower starch content. The starchy Soft white variety of wheat is used to make refined wheat flour (Maida) and Pastry flour. (http://www.indiacurry.com/bread/br001 aboutwheat.htm).

A process for the production of wheat flour or semolina comprises of the following steps-

a) Wet the caryopses of wheat with an amount of water to bring their moisture content to at least 15 %, subjecting them to intense vibrations

b) Subject the wet caryopses to a conditioning step

c) Subject the conditioned caryopses to operations of decortication, to take off the outer layers of bran

d) Milling the conditioned and decorticated caryopses.

Other than the popular regular bread made with wheat flour, the varieties of bread prepared using other grains such as rye and oat breads also contain wheat flour in good amounts.
Many other popular foods are made from wheat flour as well, resulting in a large demand for the grain even in economies with a significant food surplus.

Whole wheat flour constitutes the wheat endosperm and retains more fiber compared to finely ground refined wheat flour (maida). In India, wheat flour is generally used for making Chapatti, roti or phulka. Chapatti dough is prepared using whole wheat flour and water with a pinch of salt added. The dough is then rolled out into Chapattis of required diameter and roasted on the open flame. These freshly made Chapattis (with or without ghee) are served with vegetables and/or dal. Wheat flour roti was classified as medium GI food with a GI value of 72± 6 (Widanagamage, Ekanayake & Welihinda 2009) and low GI food (GI=45.1) by Radhika et al (2010).

Thepla

Thepla, a variant of the Chapatti, is prepared by adding Bengal gram flour (25%) to whole wheat flour. Small amounts of fresh fenugreek leaves, curds and sometimes even jaggery is added making it a wholesome meal by itself. Thepla is a typical Gujarati specialty served with curds, pickle or chutney. The variety of ingredients used increase the fiber and protein content of Thepla thereby improving its nutritional value. They may also play a role in reducing the glycemic response. Fiber delays digestion of starch in the stomach producing a hypoglycemic effect (Nishimune et al 1991) while protein ingestion along with carbohydrate has an effect on lowering total postprandial blood glucose area (Spiller et al 1987).

In comparison to the plain Chapatti also made using whole wheat flour, the addition of Bengal gram flour, fenugreek and fat in Thepla create a balanced meal. In the present study,
Thepla has been selected as one of the test foods to understand whether it can be considered as a better substitute to plain whole wheat Chapatti in improving metabolic response.

In order to determine the effect of co-ingredients in mixed meals on the glycemic and insulin responses to foods, Chandalia et al (1992) fed two different iso-carbohydrate meals (50g available carbohydrate) containing whole wheat flour +Bengal gram flour bread (Chapatti) in ratio of 2:1 and plain Bengal gram flour bread against the standard white bread to 5 NIDDM patients. The GI of Bengal gram flour Chapatti was lowest at 48%, followed by wheat and gram flour mixed Chapatti (66.4%) and highest for plain wheat Chapatti (76%). However, the insulin response followed a reverse trend with lowest insulin response to plain wheat flour Chapatti, followed by wheat flour and gram flour mixed Chapatti (118.6%) and highest for the gram flour Chapatti (202.1). The study demonstrated a stimulatory effect of Bengal flour on insulin secretion of NIDDM patients which may appear to be an important mechanism responsible for the low GI of the gram flour.

**Marie biscuit**

Marie biscuit is prepared mainly by mixing refined wheat flour (maida), hydrogenated vegetable oil and sugar together kneaded into dough before being baked at high temperatures.

Low moisture dough and short baking time is required in the manufacture of biscuits. Starch granules tend to remain more intact due to low water availability inhibiting the swelling and subsequent gelatinization of starch granule which is less susceptible to the action of amylases. Out of the total CHO content of approx 77g%, upto 30% is in the form of free sugars (Table 2.10).
Marie biscuit is one of the most popular biscuits, being advised by Doctors and Nutritionists as a better substitute for glucose biscuits. It is considered as a light, non-sweet tasting biscuit which is consumed with tea/coffee by most individuals. The use of refined wheat flour reduces the fiber content of Marie and the fiber content is not mentioned on the pack either. Also the specific textural quality of biscuits (crispy and crunchy texture) is obtained using finely ground refined wheat flour and vanaspati which increases its saturated fat content. The consumer needs to be cautioned regarding overuse of this low fiber, high saturated fat and high sugar containing biscuit.

**Table 2.10: Nutrition facts per 100g Marie biscuit (as stated on pack)**

<table>
<thead>
<tr>
<th>Nutrient</th>
<th>g/100g</th>
<th>Nutrient</th>
<th>g/100g</th>
</tr>
</thead>
<tbody>
<tr>
<td>Energy</td>
<td>440kcal</td>
<td>Carbohydrates</td>
<td>77</td>
</tr>
<tr>
<td>Calcium</td>
<td>90mg</td>
<td>** Sugars</td>
<td>22</td>
</tr>
<tr>
<td>Iron</td>
<td>3</td>
<td>Proteins</td>
<td>8.5</td>
</tr>
<tr>
<td>Iodine</td>
<td>15mcg</td>
<td>Fat</td>
<td>10.9</td>
</tr>
<tr>
<td>Thiamine B1</td>
<td>0.12mg</td>
<td>Saturated fatty acid</td>
<td>5</td>
</tr>
<tr>
<td>Riboflavin B2</td>
<td>0.14mg</td>
<td>Monounsaturated fatty acid</td>
<td>4</td>
</tr>
<tr>
<td>Niacin B3</td>
<td>1.6mg</td>
<td>Polyunsaturated fatty acid</td>
<td>1</td>
</tr>
<tr>
<td>Pyridoxine B6</td>
<td>0.2mg</td>
<td>Trans fatty acid</td>
<td>0</td>
</tr>
<tr>
<td>Cyanacobalamin B12</td>
<td>0.1mcg</td>
<td>Cholesterol</td>
<td>0</td>
</tr>
<tr>
<td>Folic Acid</td>
<td>10mcg</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

The glycemic and insulin responses to specific amounts of Marie biscuit is compared with that of same amount of bread, Chapatti and Thepla to determine which amongst the thee is more beneficial in terms of overall metabolic response.
Vada Pav

The popular local snack of Mumbai- Vada Pav consists of *batata vada* (Potato cutlets coated in gram flour paste and deep fried) stuffed into Pav (burger buns) served with green coriander chutney and red imli chutney. Majority of the people dwelling in Mumbai throve on Vada Pav not only as a snack item but for some lower middle class group, it forms a meal often replacing lunch. The ingredients used in Vada Pav including chana flour (plant protein source), Potato, oil (fat), green coriander chutney (fiber, some vitamins) contribute some additional nutrients in comparison to plain white bread.

This savoury snack is sold on footpaths, small hotels, cafeterias and also in most restaurants in Mumbai. So widespread is its patronage that now there are specific outlets dedicated to Vada Pav such as Goli Vada Pav, Jumbo King, etc selling different variants even chhole Vada Pav, ragda Vada Pav, misal Vada Pav, cheese Vada Pav, brown bread Vada Pav, etc. The major concern in consuming Vada Pav from the streets or cafeterias is the use of low quality oil (which is reused), stale bread, unclean utensils, etc. however, the emergence of clean and hygienic franchises has helped to overcome these hurdles. If purchased from a hygienic outlet or prepared at home served with green coriander and red imli chutney, Vada Pav can indeed constitute a fairly satisfying mini-meal. In the present study, the glycemic and insulin response of Vada Pav as a combination of Potato and bread has been explored.

Rice

Rice (*oryza sativa*) is one of the most commonly consumed staple cereals in many countries including India where it constitutes 60% of the total calories consumed by the population (Mahadevamma & Tharanathan 2007). In Asia, average Rice consumption was reported to be
more than 80 kg/person per year with India alone showing consumption of 208g/day (Kennedy & Burlingame 2001).

Rice is prepared by milling of paddy (rough Rice as obtained from the harvest) as such or after preliminary hot water soaking followed by steaming, drying and milling. Generally, it is consumed as white, milled, polished Rice containing high amounts of starch but lacking in other nutrients and fiber. Several nutraceuticals present in bran layer of raw harvested Rice are lost depending upon degree of milling. However, Rice protein is considered to be of high quality due to presence of all eight essential amino acids in a well balanced proportion except lysine (Mahadevamma & Tharanathan 2007).

Rice embryo and endosperm mainly contain sucrose in addition to small amounts of Raffinose, glucose, and fructose. The total sugar in embryo varies from 8-25% while reducing sugar (primarily glucose) ranges from 1- 11%. With milling, the sugar content decreases being 0.22-0.45% for milled Rice and 6.4% in Rice bran (Pascual, Singh, & Juliano 1978).

Milled Rice has been reported to have particularly high starch content upto 77.6% (Juliano & Bechtel 1985). Maningat & Juliano (1979) have reported amylose content of Rice to be very low (0-9%), low (9-20%), intermediate (20-25%), or high (25-33%). Amylose content may vary between short grain (18-20%), medium grain (15-20%) and large grain (23-26%) (Juliano 1979). Rice starch has varied applications in food industry as a thickener for puddings and ice cream, in cosmetic dusting powders and as laundry starch.

Since Rice forms a part of staple diet in India, the post prandial response to Rice consumption has been studied extensively in normal as well as diabetic individuals. Rice, in
general (except high amylose-containing variety) has been classified as high Glycemic Index (GI) food (Miller, Pang & Bramall 1992). GI is expressed by comparing postprandial area under the curve (AUC) responses after feeding equal quantities of available carbohydrates from test foods and standard (glucose or white bread) (Jenkins, Wolever & Jenkins 1998). The GI of white Rice ranged from 54 to 121 when bread (GI=100) is used as reference (Jenkins et al 1981). The wide variation in values could be attributed to differences in cooking methods, amount of water added, variety of Rice, amylose content, temperature at which it is served, etc. But people consume Rice as a whole and not just the available carbohydrate in it. Therefore the response of Rice consumption will depend on the portion size inclusive of its other components such as moisture content in boiled/cooked Rice. In 1993, Trout, Behall & Osilesi had also suggested that the prediction of glycemic responses to starchy food should be based on method of cooking / processing and characteristics of starch and starch granules rather than total quantity of starch present alone.

The study of glycemic response to nine types of Rice showed that Rice noodles, long-grain Rice, easy cooking long-grain Rice and white basmati Rice are low GI foods while all other Rice products such as brown basmati, basmati and wild Rice, Thai red Rice, etc had moderate to high GI (Ranawana, et al 2009).

Rasmussen, Gregersen & Hermansen (1992a) compared responses of seven diabetics to 25 and 50 g carbohydrate from white Rice and white bread and found significantly lower glucose and insulin responses after the 50 g Rice compared to 50 g white bread, but no differences in the responses to 25 g of carbohydrate, indicating that amount of food consumed affects glucose and insulin responses. Similar reductions were found in responses to 100 g of Rice compared to white bread in men and women (Rasmussen et al 1992b).
Results for Rice are very inconsistent, but many foods appear to have higher glycemic indices than white bread, and brown Rice may have a lower glycemic index than white Rice (Miller 1995). Instant and parboiled Rice appear to be less likely to reduce glucose and insulin responses than standard Rice, and a higher level of amylose is beneficial.

These results show that the current method of carbohydrate exchange, which assumes that all carbohydrates are equal, can be a dangerous recommendation for the control of glucose in noninsulin-dependent diabetics. Many factors can affect the glucose and insulin responses resulting from consumption of grain-based carbohydrate-containing foods (Hallfrisch and Behall 2000).

**Rice products**

**Puffed rice**

Puffed Rice (*kadle puri*), one of the common Rice products, is traditionally prepared by popping unpolished raw Rice in the presence of moisture (13 - 17 %) at 220°C for 20 seconds (Mahadevamma & Tharanathan 2007). A newer method of puffing involves expansion of Rice kernels due to formation of high pressure steam with exposure to extremely high temperature for short time. This results in the formation of various stages of gelatinized starch cells as seen in transverse sections of Puffed Rice kernels. Puffed Rice form expanded porous structures with several void spaces and air vacuoles due to cellular expansion (Chandrasekhar & Chattopadhyay 1990). The structural modification due to processing of Rice alters the starch structure and may alter its functionality as well.
Table 2.11: Proximate composition of Puffed Rice

<table>
<thead>
<tr>
<th>Sample</th>
<th>Moisture</th>
<th>Protein</th>
<th>Total CHO</th>
<th>Amylose</th>
<th>RS</th>
<th>Damaged starch</th>
<th>In vitro digestibility</th>
<th>Insol DF</th>
<th>Sol DF</th>
</tr>
</thead>
<tbody>
<tr>
<td>Puffed Rice</td>
<td>8.2</td>
<td>6.6</td>
<td>74.2</td>
<td>22.0</td>
<td>2.4</td>
<td>82.0</td>
<td>83.0</td>
<td>3.0</td>
<td>1.0</td>
</tr>
</tbody>
</table>

Source: Mahadevamma & Tharanathan 2007

Puffed Rice starch has a flaky appearance and the morphology is totally lost with greater visibility of swollen aggregates of irregular sizes (Mahadevamma & Tharanathan 2007). The damaged starch (Table 2.11) inclusive of gelatinised starch is known to imbibe more water and is more susceptible to amylolysis. This leads to faster absorption and utilization of starch in Puffed Rice which may result in faster rise in blood sugars upon consuming Puffed Rice.

Rice Kheer

Rice Kheer, prepared from basmati Rice cooked in evaporated full fat milk, is a popular sweet dish in India where milk (protein) and sugar (sucrose) are the co-ingredients added to Rice. Full fat buffalo milk is boiled and reduced to half its quantity making it a dense, semi-solid sweet delicacy.

Lactose (milk) and sucrose (sugar) are the primary sugars present in Rice Kheer. In normal healthy individuals, there is immediate utilization of lactose resulting in no appreciable increase in postprandial blood glucose concentrations (Koehler, Rapp & Hill 1934). Sucrose is hydrolyzed rapidly to cause a sharp rise in blood sugars and its hydrolysis products are also rapidly absorbed in the body (Rabinowitch 1945). Studies have shown that sucrose added to foods does not affect the daily glycemic profile or the total calculated glycemic area under curves compared to refined starchy foods with no sucrose added (Louie et al 2008, Marchini
et al 1994) but the plasma insulin response curve was 20% higher than that of glucose
(Crapo, Reaven & Olefsky 1976). Milk products are generally considered as low GI but milk
have shown to produce higher Insulin Index in healthy subjects (Östman, Elmståhl & Björck
2001, Liljeberg & Bjorck 2001, Schezenmeir J, et al. 1989) which may be attributed to
insulinotropic effects of milk proteins. Whey in milk contains insulin secretagogue which is
responsible for increased insulin secretion in postprandial period (Nilsson et al 2004).
Effect of co-ingredients on postprandial glycemic and insulinemic responses to mixed meals
have been studied in Rice Kheer as a product of Rice.

**Potato**

Potato tuber is swollen stem storing energy mainly as starch which is almost completely
digestible when Potatoes are consumed freshly cooked. Hence the carbohydrate in cooked
Potato is considered as “available carbohydrate”. Approximately 80% of Potato tuber is
water and on dry weight basis, the carbohydrate is present in the form of starch with no lipid.

Potato starch has been reported to be sensitive to variety, climate and agricultural procedure
and hence may show large variations. Amylose amounts for 20–30%, but amylopectin is
typically the major component and is extensively branched containing short chains with an
average length of 14–17 glucosyl residues (Bertoft & Blennow 2009).

About 3% of the World crop of Potato is used for production of Potato starch which is
mainly used in food, paper, textile and adhesive industry. Tuber starch granules including
Potato and tapioca tend to be larger than seed starches and are less dense and easier to cook.
Typically they also have higher degree of polymerization and larger average number of chains (Hizukuri 1988).

Potato starch when subjected to moist heat undergoes gelatinization followed by retrogradation upon cooling. The amylose: amylopectin ratio and size of amylopectin side chains affect starch digestibility under conditions when gelatinised cooked Potato starch undergoes retrogradation (Jane et al 1999). Amylopectin chains vary widely in length and may affect rearrangement of starch molecules during retrogradation. The longer amylopectin chains seem to cause less structural impediment to alignment increasing the tendency to form slowly digestible starch thereby lowering the glycemic response. Amylopectin with high density of branching digests at a slower rate than amylopectin with longer internal chains but as the chain length increases the inhibition of digestion increases (Hamaker et al 2008). Starches with higher amylopectin content, which are known to release glucose faster, are encouraged during fasting, post-recovery and post-exercise period.

Ungelatinised Potato starch added to augment dietary fiber produces significantly higher proportion of butyric acid than commercially accepted resistant starch (Henningson et al 2003). Monro (2008) showed that propensity to form slowly digestible starch after cooking varies greatly amongst Potato genotypes. Native Potato starch has highly ordered, tightly packed structure rendering it highly resistant to enzymatic hydrolysis. Raw Potato starch is virtually resistant to enzyme activity but is rapidly digestible upon gelatinization. Raben (1994) reported no effect of native ungelatinized Potato starch on blood glucose levels. Gelatinised cooked Potato has been shown to have a high GI but the true glycemic impact in
postprandial period is dependent upon its concentration in food and amount of food consumed.

The water added for boiling or cooking acts as a diluent for all nutrients and also energy. Potato has intrinsically low energy density compared to many other carbohydrate staple foods. Hence, boiled Potato without any added fat can form a low calorie food product that can be used in blood glucose control and obesity management. When stating “GI of Potatoes” it must be pointed out that it is the “GI of the available carbohydrate in Potatoes” and not whole Potatoes per se (Monro 2003). Glycemic effect of foods depends upon all the amount of food consumed, the CHO in the food and GI of the CHO in it. A high GI is not necessarily synonymous with large glycemic effect either on an equal weight basis or common standard food measure basis, certainly not for high moisture containing foods such as Potatoes

Hence the concept of RGI as applied to whole foods can reflect the glycemic response better. RGI , and its close relation to GL, in contrast to GI, reflects glycemic effect of an entire food relative to effect of glucose and has been defined as “ weight of glucose that would induce a glycemic response equivalent to that induced by given amount of test food” (Miller Jones 2007). Therefore, it is expressed in gram units as the gram of glucose equivalent (Livesey 2005) or GGE (Monro 2002, Monro & Shaw 2008).

Relative glycemic Potency (RGP) is a calculated Relative glycemic impact (RGI) of 100g of food i.e. GGE/100g food (Monro 1999). These values can help to predict glycemic responses of Potatoes for 100g and also per serving like any other nutrient with weight units (grams). If the GGE of a serving of Potato is 20g then it shows that a serving of Potato has the same
glycemic effect as consuming 20g glucose. This forms a simple and easy to understand means of comparing food based on their glycemic potential (Monro 2004)
GI, referring to mainly available CHO alone (20%), provides an exaggerated idea of glycemic impact of Potato. Unlike RGI, GI does not directly indicate how glycemic impact is affected by the quantity of food consumed since it only an index without any units. Table 2.12 compares the glycemic potency of common standard measures of Potatoes cooked in different ways.

Table 2.12: Glycemic Potency of Potatoes and its nutritional composition

<table>
<thead>
<tr>
<th>Food</th>
<th>CSM</th>
<th>Amt (g)</th>
<th>RGI</th>
<th>GGP</th>
<th>GI</th>
<th>Energy (kJ)</th>
<th>Avg. CHO (g)</th>
<th>Total Fat (g)</th>
<th>Water (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Potato, Rua, flesh, boiled*</td>
<td>1 cup</td>
<td>164</td>
<td>17</td>
<td>10</td>
<td>56</td>
<td>559</td>
<td>30</td>
<td>0.3</td>
<td>126</td>
</tr>
<tr>
<td>Potato, Rua, flesh, boiled*</td>
<td>1</td>
<td>100</td>
<td>17</td>
<td>10</td>
<td>56</td>
<td>341</td>
<td>18</td>
<td>0.2</td>
<td>77</td>
</tr>
<tr>
<td>Potato, microwaved#</td>
<td>1</td>
<td>90</td>
<td>17.1</td>
<td></td>
<td>81</td>
<td>NA</td>
<td>21</td>
<td>NA</td>
<td>NA</td>
</tr>
</tbody>
</table>

Source: *Monro & Misha 2009, †Monro 2002, NA- not available

In foods like Potato which contain CHO mainly in the form of moderate to highly digestible form of starch (with little resistant starch), the GL and RGI vales can be used interchangeably because they are similar when applied to single food intake (Venn et al 2006). In a clinical study on 18 type 2 diabetic subjects meals containing 50g CHO either as starch (alone and as mixed meal) or as glucose were ingested to study impact on postprandial glycemia and insulinemia was studied. When fed alone, the glycemic index of Potato was higher than that of Rice whereas in mixed meals the responses were 20% lower. The insulin index was found to be similar for all test foods eaten alone in quantity that provided 50g available carbohydrate portion however, when eaten as mixed meals insulin secretion increased
significantly with a hierarchical pattern being highest for bread followed by Potato and Rice (Bornet et al. 1987).

For example, a recent study by Fernandes et al. (2005) examined the effect of cooking on the GI of Potatoes prepared in a variety of different ways including mashed; baked; reheated; boiled; boiled and cooled; and fried. The results indicated that the GI values of Potatoes varied significantly depending on both the variety and cooking method used, ranging from intermediate (boiled red Potatoes consumed cold: 56) to moderately high (roasted white Potatoes: 73; baked russet Potatoes: 72). Similarly, Kinnear et al. (2011) investigated the effects of cooking and cooling on the GI of four novel Potato varieties and found significant variability in the effects. Specifically, cooking and cooling reduced the GI of two Potato varieties by 40-50%, while it produced only a 8-10% reduction in the other two varieties.

**Sago**

Tapoica is recognized by several names as yuca, rumu or manioca in Latin America, manioc in French-speaking Africa and Madagascar, cassava in English-speaking Africa, Ceylon and Thailand, mandioca or aipim in Brazil, tapioca in India and Malaysia, and bi ketella or kaspe in Indonesia (FAO, 1998). Fresh roots contain about 60 - 70% moisture, 7 - 12% protein, 5 - 13% starch (32 - 35% total carbohydrate) and trace amounts of fat (Lancaster et al. 1982, Jackson 1990, FAO 1998). The high starch and moisture content render it extremely perishable (Hahn 1989, Mlingi et al. 1996). Processing is therefore indispensable to facilitate preservation, improve palatability and product quality as well as reduce cyanogenic glycoside toxicity (Jones 1998).
Tapioca was introduced in India during the later part of the 19th Century, but it was only in the 1940’s that Tapioca starch and sago were introduced in India. It is mainly grown in the States of Kerala, Andha-Pradesh, & Tamil Nadu. Sago and starch are prepared from Tapioca Root. Sago was produced first in Salem (Tamilnadu) and now it ranks first in respect of processing of tapioca into starch & sago, in India. Sago production started on a cottage scale basis in India in about 1943-44 by pulping the tapioca roots, filtering the milk-extract and after settling the milk, forming globules and roasting these globules. Generally, Indian tapioca root containes about 30% to 35% starch contents. Sago is considered to be a very nutritious product as it contains carbohydrates and appreciable amount of calcium and vitamin C (http://www.sabuindia.com/sago1.htm).

Sago (metroxylin sagu) is the commonest form of native starch tapioca consumed as sabudana in India. In West Bengal, it is termed as tapioca granules, JAVARISHI in Tamilnadu, SABUDANA or SABU in Maharastra- MadhyaPradesh, Bihar, Rajasthan, Uttar Pradesh & Gujarat. It is prepared from the milk of tapioca root. Boiled sago pearls can be mixed with other foods such as Rice, Potato or noodles and eaten as a direct source of carbohydrates (Ahmad, Singh & Ghosh 2009). SAGO is popularly used in KHICHADI in Maharastra & MadhyaPradesh, as baby food in West Bengal, KHIR (PAYASAM) in most states and FARIYALI Items in Gujarat, Rajasthan on Vrata-Upavas (Fasting Days). Generally, people regard as a food to be taken during illness for easy digestion & quick recovery.
Process of sago production

a. Large sized raw material (Tapioca Root) is washed
b. Skin from Tapioca Root is peeled
c. Root is crushed in rollers to make pulp
d. Pulp is sent to shifter for separating coarse particles (all fiber & impurities are retained)
e. Milk is allowed to settle in a tank for nearly 3 to 8 hs (all residual impurities float to the top of the tank & are drained out of the settled milk)
f. Settled milk cake is used for making globules by a very special & unique type system on very simple indigenous machine
g. Settled starch is passed through sieve and partially dried by roasting on hot plates or heating in steam
h. It is dried completely under direct sunlight in big platforms
i. Polished to be marketed as shiny sago pearls/ beads

Table 2.13 given below shows the special characteristics of Sago under AGMARK fixed by Department of Agriculture & Co-operation, Directorate of marketing and Inspection, Ministry of Agriculture
Table 2.13: Special characteristics of Sago under AGMARK

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Milkwhite</th>
<th>General</th>
<th>Special</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moisture percent by weight (Maximum)</td>
<td>12.0</td>
<td>12.0</td>
<td>12.0</td>
</tr>
<tr>
<td>Total ash (Maximum)</td>
<td>0.4</td>
<td>0.4</td>
<td>0.4</td>
</tr>
<tr>
<td>Acid insoluble ash (Max.)</td>
<td>0.1</td>
<td>0.1</td>
<td>0.1</td>
</tr>
<tr>
<td>Starch percentage (Minimum)</td>
<td>95.0</td>
<td>92.0</td>
<td>90.0</td>
</tr>
<tr>
<td>Protein percentage (Max.)</td>
<td>0.3</td>
<td>0.3</td>
<td>0.3</td>
</tr>
<tr>
<td>Sulphur dioxide PPM. (Max.)</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>Crudefibre percentage (Max.)</td>
<td>0.2</td>
<td>0.2</td>
<td>0.2</td>
</tr>
<tr>
<td>PH.of aqueous extract</td>
<td>4.5 to 7.0</td>
<td>4.5 to 7.0</td>
<td>4.5 to 7.0</td>
</tr>
<tr>
<td>Colour of gelatinised alkaline paste in the porcelain cuvetta on the lovibond scale not deeper than</td>
<td>0.2R + 1.0Y</td>
<td>0.3R+1.0Y</td>
<td>0.4R+1.5Y</td>
</tr>
</tbody>
</table>

Sago starch has been considered as an easily digestible, quickly absorbed, ready source of energy and hence is promoted for consumption on days of fasting. But like Rice, Sago is primarily rich in starch (30-35%) but lacking in other nutrients. It is soaked in twice the amount of water to allow it to swell to its maximum capacity and then used for cooking. With the growing emphasis on promoting high fiber nutrient dense foods, the functional and nutritional quality of sago and its products and their effect of metabolism need to be explored.

Hirao et al (2000) compared the effect of sago versus tapioca starch on blood lipid concentrations. Both raw and gelatinized Sago starch resulted in a decrease in serum total cholesterol levels and atherogenic indexes along with decrease in serum triacylglycerol
concentrations. This was attributed to the lower digestibility and higher amylose content of Sago starch compared to tapioca starch.