TO ESTABLISH CORRELATION BETWEEN GLYCEMIC INDEX AND IN-VITRO

CARBOHYDRATE DIGESTIBILITY OF BREAKFAST ITEMS-IDLI USING RICE RAWA VERSUS JOWAR RAWA

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B.Sc. (Home Science)

MASTER OF SCIENCE IN HOME SCIENCE

(FOODS AND NUTRITION)

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BY

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B.Sc (Home Science)

THESIS SUBMITTED TO THE ACHARYA N. G. RANGA AGRICULTURAL UNIVERSITY IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE AWARD OF THE

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2013

DECLARATION

I **AFIFA JAHAN**, hereby declare that the thesis entitled **"TO ESTABLISH CORRELATION BETWEEN GLYCEMIC INDEX AND INVITRO CARBOHYDRATE DIGESTIBILITY OF BREAKFAST ITEM-IDLI USING RICE RAWA VERSUS JOWAR RAWA"** Submitted to **Acharya N. G. Ranga Agricultural University** for the degree of **Master of Science in Home Science** is the result of original research work done by me. I also declare that no material contained in the thesis has been published earlier in any manner.

Place: Hyderabad

(AFIFA JAHAN)

I.D. NO. HHM2011-05

Date: 12-July- 2013

CERTIFICATE

This is to certify that the thesis entitled **"TO ESTABLISH CORRELATION BETWEEN GLYCEMIC INDEX AND INVITRO CARBOHYDRATE DIGESTIBILITY IN IDLI USING RICE RAWA VERSUS JOWAR RAWA"** submitted in partial fulfillment of the requirements for the degree of '**MASTER OF SCIENCE IN HOME SCIENCE**' of the **Acharya N. G. Ranga Agricultural University, Hyderabad** is a record of the bonafide original research work carried out by **Ms. AFIFA JAHAN** under our guidance and supervision.

No part of the thesis has been submitted by the student for any other degree or diploma. The published part and all assistance received during the course of the investigations have been duly acknowledged by the author of the thesis.

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(**DR. USHA RANI)**

Chairman of the advisory committee

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(AFIFA JAHAN)

LIST OF SYMBOLS AND ABBREVIATIONS

ADA	:	American Diabetic Association
CHO	:	Carbohydrate
GI	:	Glycemic Index
GR	:	Glycemic Response
IR	:	Insulinaemic Response
ICMR	:	Indian Council of Medical Research
IDDM	:	Insulin Dependent Diabetic Mellitus
IVSD	:	Invitro Starch Digestability
NIDDM	:	Non Insulin Dependent Diabetic Mellitus
OGTT	:	Oral Glucose Tolerance Test
PR	:	Protein
RAG	:	Rapidly Available Glucose
SAG	:	Slowly Available Glucose
SDF	:	Soluble Dietary Fiber
TDF	:	Total Dietary Fiber
WHO	:	World Health Organisation
g	:	Grams
mg	:	Milligrams

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ABSTRACT

Diabetes is a metabolic disorder with many potential complications over the long term. Therefore, any measure either to prevent precipitation of the disease or to alleviate the ailment is always a great help to Diabetics. Since it is not possible to prevent or cure diabetes completely, it can be kept under control through appropriate diet therapy.

It is always preferable to modify the diet based on individual life style considering the traditional eating pattern and food habits. Hence, commonly consumed south Indian breakfast product idli made with rice rawa and jowar rawa were selected for present study.

The raw materials were procured from local market in Hyderabad. Initially fifteen members were selected from Vasantha Nilayam Ladies hostel at ANGRAU campus with the age group of 22-23 years. Subjects were excluded if they reported a history of gastrointestinal disorders, suffered from diabetes, were taking medication for any chronic disease conditions, or intolerant or allergic to any of the foods. Finally, ten healthy subjects were identified for study from initial 15 members.

The glycemic index of the breakfast product-idli made with rice rawa and jowar rawa was assessed in non diabetic young subjects using blood sampling schedule and method of Incremental area under the curve (IAUC) using glucose as reference food. The IAUC of rice rawa idli was 592.5 mg/dl against the glucose value of 1110. mg/dl. The glycemic index of the rice rawa idli was 56.3, which is considered as medium glycemic index food. The 50g of carbohydrate can be taken in one serving i.e. five medium sized rice rawa idlis. The glycemic load of the experimental rice rawa idli was calculated as 26.65, which is considered as high glycemic load.

The IAUC of rice jowar idli was 562.5 mg/dl against the glucose value of 1110. mg/dl. The glycemic index of the jowar rawa idli was 51.2, which is considered as low glycemic index food. The 50g of carbohydrate can be taken in one serving i.e. six medium sized jowar rawa idlis. The glycemic load of the experimental jowar rawa idli was calculated as 25.3, which is consider as high glycemic load.

The IVSD value of rice rawa idli 18.9 and IVSD value of jowar rawa idli 20.92. IVSD value of rice rawa idli is lower than IVSD value of jowar rawa idli.

The correlation value of the GI and IVSD of the idli made with rice rawa is 0.899 which is found significant at 5 percent level and similarly the jowar rawa idli is calculated and the value is 0.849 which is found significant at 5 percent level.

Results obtained indicate that least glycemic response was observed with jowar rawa idli. Thus the inclusion of jowar based breakfast product-idli may be recommended to the habitual diet for achieving a good glycemic control in diabetics.

These findings should assist in development of high fiber and glycemic index sorghum products with good amount of IVSD, which will help in development of functional foods.

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Chapter I

INTRODUCTION

"Diabetes or Madhumeham" is known as a disease related to sweetness, which has been described in the literature as third world disease, or global disease due to its high prevalence rate. Diabetes affects more than 230 million people worldwide and is expected to affect 350 million by 2025. In 2003, the five countries with largest number of diabetic people were India(35.5 million),China(23.8 million),The United State (16 million),Russia(9.7 million) and Japan(6.7 million) (Diabetes Atlas,2003). The statistical data signifies diabetes as a major public health challenge.

Diabetes is estimated to be the fourth leading cause of death globally. Each year over three million deaths are directly related to diabetes. In every ten seconds, a person dies from diabetes related causes. It is also the leading cause with diabetes is two to four times are more likely to develop cardiovascular disease than people without diabetes. The devasting complications such as blindness, kidney failure and heart disease are imposing a high burden on health care services which accounts for 5-10 per cent of a nation's health budget. The human and economic costs of diabetes could be significantly reduced by investing in prevention, particular early detection in order to avoid the onset of diabetes. (Diabetes Atlas, 2003).

Clinically diabetes is manifested by overflow of sugar or glucose in the blood and urine instead of being converted into glycogen. The result is danger to the fundamental process of energy balance with stimulates the body to adopt some compensatory measures. Thus, diabetes is not a self-contained abnormality but a group of disorders with varying etiology and pathogenesis. It is characterized by insulin deficiency or decreased insulin action, abnormality of glucose, lipid and protein metabolism and the development of both acute and long term complications.

WHO study group (1985) on diabetes reported that if fasting plasma glucose level is greater than 140 mg/dl or the random plasma glucose is greater than 200 mg/dl on more than two occasions, a diagnosis of diabetes may be made. If the random blood glucose lies in the uncertain range (100-200 mg/dl)then oral glucose tolerance test (OGTT) must be done to establish diagnostic status. Recent diagnostic approach for diabetes is glycosylated haemoglobin (HbA_1C or HbA_1) test which provide a rapid assessment of the level of glycemic role in diabetic patients.

In 1997, American Diabetic Association (ADA) issued new diagnostic and classification criteria and the modification were made in 2003 regarding the impaired fasting glucose. Clinically diabetes is classified into:

- Type –I Insulin Dependent Diabetes Mellitus (IDDM) which results from b-cells destruction, usually leading to absolute insulin deficiency.
- Type –II Non Insulin Dependent Diabetes Mellitus (NIDDM), which results from progressive insulin secretary, defects on the background of insulin resistance.
- Gestational diabetes which occurs during pregnancy
- Other specific type of diabetes due to other cause eg; genetic defects in b cells function, gentic defect in insulin action, disease of the exocrine pancreas and drugs or chemical induced (ADA , 2005)

Type –II diabetes or NIDDM constitute nearly 95-97 per cent of all diabetic patients in most populations group. It is estimated that by the year 2010 A.D, 20 per ecnt of all TYPE-II diabetes in the world will be contributed from India (Premalata 1998)

The treatment stratergies to overcome these defects are modified meal plan, excersise,blood glucose lowering drugs and insulin (Sundaram 1998)among the package of strategies diet remain the corner stone especially for NIDDM subjects.

In diet therapy diet composition, amount, distribution, and the time of food intake are important factors. The diet must be acceptable, supply adequate amount of nutrient and be formulated in a way to normalize body weight (Khan et al 2003). The importance of diabetic diet has always been controversial and being continually evaluated.

The glycemic index (GI) is a concept introduced by Jenkins et al. (1981) to classify foods based on their immediate effect on blood glucose levels. The GI is defined as the postprandial incremental glycemic area after a test meal, expressed as the percentage of the corresponding area after an equicarbohydrate portion of a reference food (glucose or white bread). From a nutritional point of view, a low glycemic response is considered beneficial for prevention of common diseases such as coronary heart disease, diabetes and obesity. Therefore, there has been a recent interest in slow digesting starches due to their beneficial role in human health.

Cereals and pulses constitute the main source of food supply. The food grain output had increased from 50.8 million tonnes during 1951 to an impressive level of about 199 million tonnes in 2000. The per capita availability of food grains of India has increased from 394 gms during 1951 to 498 g in 1966 and 531 g in 2000 vijayalakshmi (2002)

Sorghum is an important source of dietary energy and a main food staple in semi-arid regions of Africa and Asia (Ezeogu *et al*. 2005). Due to its drought tolerance and adaptation attributes, this grain can be grown in those areas where agricultural and environmental conditions are unfavorable for the production of other crops. Sorghum is considered as the world's fifth most important cereal after wheat, rice, maize, and barley (Serna-Saldivar and Rooney, 1995). Worldwide, over 35% of sorghum is grown directly for human consumption, while the rest is used primarily for animal feed, alcohol, and industrial products (Rooney and Awika, 2004).

Sorhum is commonly served in form of chapatti,daliya,upma, idli and khichidi in rural and urban areas.

Sorghum is crucial to the world food economy because of its contributions to household food security in many of the world's poorest, most food – insecure regions (ICRISAT, 1996). Sorghum grains are important source of dietary proteins, carbohydrates, minerals, and B group vitamins particularly to the vegetarian diets in India (Salunkhe *et al*. 1984).

There is a considerable variation in sorghum in the levels of proteins, lysine, lipids, carbohydrates, fiber, calcium (Ca), phosphorus (P), iron (Fe), thiamine, and niacin. All these parameters impart sorghum grain quality (Hulse *et al*. 1980).

Several studies were conducted in order to improve the digestibility of sorghum, Invitro starch digestibility of sorghum in processed products depends on the type of processing. Popping of sorghum increases IVSD by 8 folds compared to whole grain (Phusphamma, 1993) but at the same time it has the negative effect on protein digestibility.

Rice is the staple food for 65% of the population in India. It is the largest consumed calorie source among the food grains. With a per capita availability of 73.8 kg it meets 31% of the total calorie requirement of the population. India is the second largest producer of rice in the world next to China.

Studies were conducted on different varieties of rice and results indicate that many varieties of rice, whether white, brown, or parboiled, should be classified as high GI foods. Only high-amylose varieties are potentially useful in low-GI diets.(Pang and Miller 1992)

Black gram originated in India, where it has been in cultivation from ancient times and is one of the most highly prized pulses of India. The coastal Andhra region in Andhra Pradesh is famous for black gram after paddy. The Guntur District ranks first in Andhra Pradesh for the production of black gram. Mainly Indian immigrants have also introduced black gram to other tropical areas.

After fermentation, in coarse rice and black gram, the Breakdown value was low compared with that in fine rice and black gram.(KOH 2009)

Idli, a very popular fermented breakfast food staple consumed in the Indian subcontinent, consists mainly of rice and black gram. Idli fermentation was carried out in the conventional way in a batter having rice to black gram in the ratios of 2:1.(Nagaraju 2000)

Idli is a fermented food of India, which is prepared by steaming a fermented black gram (Phaseolus mungo L.) and rice (Oryza sativa L.) batter. It makes an important contribution to the diet as a source of protein, calories and vitamins, especially B-complex vitamins, compared to the raw unfermented ingredients. It can be produced locally and used as a dietary supplement in developing countries to treat people suffering from protein calorie malnutrition and kwashiorkor. Other legumes such as soybeans and Great Northern beans could be substituted for black gram in preparation of a idli. Further research is needed regarding the increase of methionine content during idli fermentation, by which path way methionine is synthesized, and identification and isolation of microorganisms responsible for methionine production or synthesis.(Reddy 1982)

Hence considering the above aspects the present study was planned with following objectives

Objectives of Investigation:

- To prepare breakfast items-idli using rice rawa and jowar rawa
- To study the effect of selected food preparations on the area under curve of glucose levels and compare with the control in selected subjects.
- To study the in-vitro digestibility of carbohydrates in the selected food preparations
- To establish a correlation between carbohydrates digestibility and glycemic index of foods

Chapter II

REVIEW OF LITERATURE

Diabetes affects more than 230 million people worldwide and is expected to affect 350 million by 2025. In 2003, the five countries with largest number of diabetic people were India (35.3 million) China (23.8 million), the united states (16 million), Russia (9.7million) and Japan (6.7 million) (Diabetes Atlas,2003)

The prevalence of diabetes is increasing constantly and the WHO has estimated that diabetes will affect 221 million people worldwide by the year 2010. (Aslam, *et. al.*, 2007). Over weight and obesity are important risk factor for type11 diabetes. Obesity also complicates the management of type II diabetes by increasing insulin resistance and blood glucose concentration. Obesity is an independent risk factor for dyslipidaemia, hypertension, cardiovascular diseases and thus increasing the risk for cardiovascular complications and vascular mortality in patients with type II diabetes (Klein, *et. al.*, 2004).

The scientific evidence pertaing therapeutic dietary efforts from early historic approach to present are reviewed and briefed below under following order.

2.1 BREAKFAST CONSUMPTION AND ITS IMPORTANCE ON HEALTH OUTCOME.

2.2 IDLI AND ITS PROPERTIES

2.2.1 RICE A STAPLE FOOD GRAIN IN IDLI MAKING.

2.2.2 BLACK GRAM A PROTEIN SOURCE IN IDLI MAKING.

2.3 GLYCEMIC INDEX

2.4 SORGHUM A STAPLE FOOD GRAIN IN DIABETES

2.5 IN-VITRO STARCH DIGESTIBILTY

2.1 BREAKFAST CONSUMPTION AND ITS IMPORTANCE ON HEALTH OUTCOME.

Studies done by Agostoni, *et al.*, (2010) disclose that breakfast represents a healthy habit and association with positive health outcomes proves breakfast should be consistent with local and family dietary behaviours. According to neurobehavioral data, the good example of parents and access to a variety of palatable and pleasant breakfast foods should drive children to choose self select breakfast models with balanced composition, while respecting recommended dietary allowances. A balanced macronutrient composition, the proposition of a variety of models leading to a total energy density preferably within lower ranges (< 1 to 1.5), as well as glycemic indices in the lower range for the same food class, could emphasize the positive short and long term health outcomes which is now attributable to breakfast.

Regular breakfast consumption can have a multitude of positive health benefits, yet young people are more likely to skip breakfast than any other meal. Given the evidence that dietary behaviors established in childhood and adolescence track into adulthood along with evidence that breakfast skipping increases with age, identifying correlates of children's and adolescent's breakfast behavior is imperative. Few studies have examined the same specific family correlates of breakfast consumption, limiting the possibilities of drawing strong or consistent conclusions. Parental breakfast eating and living in two parent families were the correlates supported by the greatest amount of evidence in association with adolescent's breakfast consumption. The results suggest that parents should be encouraged to be positive role models to their children by targeting their own dietary behaviors and that family structure should be considered when designing programmers to promote healthy breakfast behaviors (Pearson *et al.,* 2009).

Eating breakfast is important for the health and development of children and adolescents. Reports on the findings of an Australian survey of 699 thirteen year old concerning the extent of skipping breakfasts, indicated approximately 12 percent of the sample skipped breakfast. Gender was the only statistically significant socio demographic variable, with females skipping at over three times the rate of males. Skippers were more likely to be dissatisfied with their body shape and to have been on a diet to lose weight than were those who ate breakfast (Shaw, 1998).

Wesnes *et al.,* (2003) reported in their study that a typical breakfast of cereal rich in complex carbohydrates can help maintain mental performance over the morning. Frequency of breakfast and cereal consumption decreased with age. Days eating breakfast were associated with higher calcium and fiber intake in all models, regardless of adjustment variable. After adjusting for energy intake, cereal consumption was related to increased intake of fiber, calcium, iron, folic acid, vitamin C, zinc, and decreased intake of fat and cholesterol. Cereal consumption as part of an overall healthful lifestyle may play a role in maintaining a healthful Body Mass Index (BMI) and adequate nutrient intake among adolescent girls (Barton *et al.*, 2005).

2.2 IDLI AND ITS PROPERTIES

Idli, a very popular fermented breakfast food consumed in the Indian subcontinent, consists mainly of rice and black gram. Idli fermentation was carried out in the conventional way in a batter having rice to black gram in the ratios of 2:1, 3:1 and 4:1 at room temperature. It makes an important contribution to the diet as a source of protein, calories and vitamins, especially B-complex vitamins, compared to the raw unfermented ingredients. It can be produced locally and used as a dietary supplement in developing countries to treat people suffering from protein calorie malnutrition and kwashiorkor (Nagaraju and Manohar, 2000).

Adding Saccharomyces cerevisiae, along with natural bacterial flora of the ingredients, was the best method for standardizing idli fermentation in terms of improved organoleptic characteristics, leavening and nutritional constituents. Traditional idli fermentation involves several bacteria and yeasts, contributed by the ingredients rice (Oryza sativa), black gram (Phaseolus mungo) and the environment, with overall dominance of the former in bringing about various changes. Idli fermentation is accompanied by an increase in total acids, batter volume, soluble solids, reducing sugars, non protein nitrogen, free amino acids, amylases, proteinases and water soluble vitamins B1, B2 and B12 contents, thus accounting for improved digestibility and nutritional value of the staples. Novel idli batter prepared by replacing conventional black gram with other legumes, revealed significant change but with difference in the levels of some biochemical constituents (Soni and Sandhu, 1989).

Idli, Dhokla, Nan, Kulcha, Bread, Jalebi, Bhatura, Bhalla, Dosa, Gulgule and Wadian were prepared in the laboratory using traditional fermentation techniques. The fermented batter of idli and dosa contained higher amount of available lysine, cystine and methionine. After processing, maximum retention of lysine, methionine and cystine was observed in steamed idli (Riat and Sadana, 2009).

2.2.1 RICE A STAPLE FOOD GRAIN IN IDLI MAKING.

Cereal grains particularly rice, form a major source of dietary nutrients for all people, particularly those in the developing countries. However, compared with animal foods, nutritional quality of cereal grains is inferior due to lower protein content, deficiency of certain essential amino acids, lower protein and starch availabilities, and the presence of some antinutritional factors. Fermentation of cereals for a limited period of time improves amino acid composition and vitamin content, increases protein and starch availabilities, and lowers the levels of antinutrients.

Cereals are deficient in lysine, but are rich in cysteine and methionine. Legumes, on the other hand, are rich in lysine but deficient in sulphur containing amino acids. Thus, by combining cereal with legumes, the overall protein quality is improved (Camphell-Platt, 1994).

Fermented foods prepared from cereals and legumes are an important part of the human diet in Southeast Asia and parts of East Africa. The popularity of legume based fermented foods is due to desirable changes including texture and organoleptic characteristics. Improvement in digestibility and enhancement of keeping quality, partial or complete elimination of anti-nutritional factors or natural toxins, increased nutritive value, and reduced cooking time (Joseph, 1994).

Cereal grains constitute a major source of dietary nutrients all over the world. Although cereals are deficient in some basic components, fermentation may be the most simple and 10 economical way of improving their nutritional value, sensory properties, and functional qualities. Products produced from different cereal substrates (sometimes mixed with other pulses) fermented by lactic acid bacteria, yeast and/or fungi (Blandino, *et al.*, 2003).

Rice colour changes from white to amber during parboiling (soaking and steaming). Colour parameters indicated that, during soaking, yellow bran pigments leaches out in the water. The levels of the Maillard precursors (i.e., reducing sugars and free α-amino nitrogen (FAN)) depends on soaking temperature and time: leaching of RS was compensated by enzymatic formation for long soaking times (>60 min), while proteolytic activity was too low to compensate for FAN leaching. Parboiled rice soaking under nitrogen, oxygen, or ambient conditions and determination of polyphenol oxidase activity allowed to conclude that the effect of enzymatic colour changes on the soaked rice colourwas rather small. Colour measurements of brown and milled mildly, intermediately, and severely parboiled rice samples showed that both brown and milled rice samples were darker and more red and yellow after parboiling and that the effect depended on the severity of parboiling conditions. Furthermore, steaming affected the rice colour more and in a way opposite to that observed in soaking (Lamberts, *et al.*, 2006).

Parboiled brown rice contained considerably more Reducing Sugars (RS) but less sucrose and Free Amino Acids (FAA) than raw brown rice. On milling, there was considerable loss of sucrose and FAA from raw rice, but very little from parboiled rice; reducing sugars changed little in either. Processing conditions affected the contents of sugars and FAA. Maximum increase in RS and decrease in sucrose content occurred after soaking at 60^0C. Controlled incubation of rice flour, intact grain, separated germ and deemed rice in water showed that considerable changes in sugars and FAA occurred in all cases, the magnitude depending on the circumstances, but a greater part of the sugars leached out into the water during soaking (Ali and Bhattacharya, 1980).

Grinding characteristics of raw and parboiled rice were evaluated in various wet grinding systems like, mixer grinder, stone grinder and colloid mill. The duration of grinding had inverse effect on the particle size and direct impact on the starch damage as well as energy consumption in batch grinders. Stone grinder was the least energy efficient and specific energy consumption for grinding raw rice (160.6 kJ/kg) was nearly twice as that 11 of mixer grinder (74.9 kJ/kg). Parboiled rice required longer duration of grinding compared to raw rice, consequently specific energy consumption was higher (~220 kJ/kg) (Sharma et al., 2008).

The nutritional quality of wild rice tends to be comparable with other cereals characterized by a high content of starch and protein and a low fat content. As a whole grain, wild rice is also a good source of dietary fibre (Qiu, *et al.*, 2010).

Parboiled brown rice contained considerably more (RS) but less sucrose and FAA than raw brown rice. On milling, there was considerably loss of sucrose and FAA from raw rice, but very little from parboiled rice; reducing sugars and FAA. Maximum increase in RS and decrease in sucrose content occurred after soaking at 60^0 C (Ali *et al.*, 2007).

A study showed (Mohan *et al.,* 2010) there was an effect of severely pressure parboiled rice reduced the glycaemic index.Brown rice malt from Indica and Japonica type rice were prepared and their nutrient composition as well as Non-Starch Polysaccharide (NSP) contents and also some of the 12 physicochemical characteristics were determined. The activity of α- and β-amylases in the un-germinated (native) rice was negligible but increased considerably on germination. Malting altered the chemical composition of both Indica and Japonica rice to a small extent but caused noticeable changes in the pasting characteristics. Controlled germination or malting causes considerable changes in the physicochemical and biochemical properties of both Indica and Japonica rice.

2.2.2 BLACK GRAM A PROTEIN SOURCE IN IDLI MAKING.

Blackgram (Phaseolus mungo) is a pulse traditionally used in the preparation of South Indian breakfast foods, such as idli, which is relished for its soft and spongy texture (Susheelamma and Rao, 1979a). The components responsible for these properties are the surface active proteins that generate a foam and as a result impart a porous structure to the food, and the viscogenic mucilaginous polysaccharide (~6%) that stabilizes the porous structure against thermal disruption during steaming.

During fermentation of black gram, for the preparation of leavened foods, it was found that the mucilaginous polysaccharide undergoes compositional and rheological changes (Muralikrishna, *et al.,*1987). Here, the fermentation is due to the activities of endogenous microflora (endophytes) in blackgram, in particular Leuconostoc mesenteroides, yeasts, lactic acid bacteria and coliforms. More than one oligosaccharide was observed as in green gram (stachyose, maltohexaose), sorghum (stachyose, maltotriose), barley (stachyose, raffinose), wheat (stachyose, raffinose) and black gram (stachyose, raffinose). In ragi, bajra and rice malt oligosaccharides were absent. Germination of seeds for 48 h resulted in complete loss of 13 stachyose and raffinose in cereals and pulses. The maltotriose content in pulses completely disappeared on germination but among cereals, 45.1 and 57.3 percent loss was observed in sorghum and maize, respectively (Sampath, *et al.*, 2008). In black gram after fermentation, apparent viscosity of cold paste increased. Some of the properties such as intrinsic viscosity, swelling and solubility after fermentation were reported by them.

Fermentation and steaming approximately 40 per cent reduction in oligosaccharides resulting in reduced flatulence in the body (Koh and Singh, 2009).

2.3. GLYCEMIC INDEX

The concept of GI was developed by Jenkins *et al* at university of Toroto more than a quarter century ago as a physiologic rather than a structural approach to classifying carbohydrates (Pereira, 2006). The GI is defined as the area under the 2hours postprandial blood glucose concentration curve per 50g available carbohydrates consumed from the test food relative to a reference food either white bread or pure glucose.

The low GI foods are kwon to produce less postprandial hyperglycemia and hyperinsulinemia than high GI foods but still the role of low GI foods in the prevention and treatment remains unclear (Miles 2008).

Miller *et al.,* (2003) studied whether low GI diets compare with conventional or high GI diets improved glycemic control in individuals with diabeties as assessed by reduced Hb AIC or fructosamine data together,glycated protein were reduced 7.4 percent more on low GI than on high GI diet. Thus, the author concluded that choosing low GI foods in place of conventional or high GI foods has a small but clinically useful effect on medium term glycemic control in patients with diabetic and the incremental benefit is similar to that offered by pharmacological agents that also target postprandial hyperglycemia.

<u>Role of nutrients on glycemic response:</u>

A delicate balance between the process of addition and removal of blood glucose into and from the blood helps to maintain the blood glucose concentration within an individual (Schenk *et al.,* 2003). It has been firmly established that among various influential factors food has a dramatic impact on blood glucose concentration. Therefore it has been a urge of interest in how foods and its nutrients specifically the major nutrients such as carbohydrate (CHO), Protein (PRO), fat affect the blood glucose. The effect of these nutrients is briefly underneath.

Carbohydrate:

In diabetic diet, effect of the CHO intake both in terms of amount and type has always been emphasized in various scientific studies carried out on diabetics.

Amount of carbohydrate:

The amount of total CHO recommended for the diabetic diet has varied significantly over the year. A low CHO were widely used for the management of diabetes until 1970s. Later a liberalization of CHO content was allowed in the diet. This was due to assumption of high CHO intake led to improvement in glucose tolerance and insulin sensitivity.

American diabetic association (ADA, 1986) suggested that amount of CHO should be liberalized, ideally upto 55-60 percent of total calories and individualized with the amount dependent on blood glucose and lipid levels and individual eating patterns. However, it was recommended by Institute of medicine (2002) that CHO intake should be in range of 45-60 per cent of total calories. However, the amount of carbohydrate in the diabetic diet remains controversial.

Type of carbohydrate (CHO):

It has been established that any change in CHO component of food will produce the reciprocal change in glycemic response (G.R).

EFFECT OF SUGAR:

The variations in the blood glucose response to different kinds of CHO led to new hypothesis that the glucose component of food but not the total CHO is the main determinant of G.R.

Hughes *et al.,* (1989) tested the above hypothesis in IDDM patients. The patients were given glucose alone, fructose alone, glucose plus fructose /lactose or glucose plus protein/fat. They observed that fructose given alone with glucose had no greater effect than glucose. Similarly, galactose contributed only slightly to G.R. when given as lactose whereas protein and fat had no additional effect.

Dukar *et al.*, (1990) reported similar findings by comparing a cholesterol free tofu based frozen diet (TFD) containing high fructose corn syrups with a dairy-based sucrose sweetened ice cream (IC) in NIDDM subjects. The TDF elicited a higher G.R. than IC inspite of high sucrose content of IC. Thus explained that this unexpected response was related to the substantial amount of total glucose in the fructose dessert.

Brand and Lobbezoo (1994) studied the effect of glycaemic and insulinemic response (IR) to a equi CHO meals based on popular puffed rice cereal containing sucrose 0,21,43 g. The results showed that G.R. and I.R was significantly lower after the meal containing the highest amount of sugar compared with non-sweetened cereal. Thus it was concluded that replacing starch with sugar (sucrose) in a high glycaemic index (G.I) food causes a significant decrease in G.R and I.R.

Englyst *et al.*, (1999) established the relationship between rapidly available glucose (RAG) and glycaemic response in healthy subjects. Eight separate meals containing RAG from 11-49 g were tested. Results indicated that a significant correlation exist between GR and RAG i-e. a given percent increase in RAG was associated with the same percentage increase in G.R.

In another study Englyst *et al.*, (2003) proceeded further to know the effect of slowly available glucose (SAG) along with rapidly available glucose (RAG) on G.R. and I.R. They tested 23 products (5 breakfast cereals,6 bakery products and crakers,12 biscuits). The finding was the biscuits group, which had the highest SAG due to presence of ungelatinised starch, was found to be lowest G.I. It was also concluded that there was no significant association between GI and either starch and sugar while RAG was positively and SAG was negatively correlated with G.I.

Heacock *et al.*, (2002) studied the effect of G.R to small doses of fructose administered before or simultaneously with a high G.I. starchy food. Treatment consisted of (50 g of available CHO) instant mashed potato fed alone or with 10g fructose fed at 60, 30 or 0 minutes before the meal. The results indicated that blood glucose area under curve (AUC) for fru (O) and non-fru control.thus concluded that fructose pre feeding reduces the G.R to a high G.I. starchy foods.

Lau *et al.,* (2005) found that lactose was positively associated with HOMA-IR (An index for insulin sensitivity) where as daily G.I and intake of glucose, fructose and dietary fiber , total CHO, fruits were inversely associated. It was also observed that there is no significant difference for sucrose.

Effect of CHO from different foods:

Das *et al*., (1991) studied the G.R and I.R to 3 products (parboiled rice ,flaked rice,suji) in six patients with mal-nutrition related diabetes (MRDM), six under nourished patients and ten healthy controls. Result indicated that serum insulin both basal and in response were lowest in MRDM patients. In under nourished patients G.I for all 3 foods was higher than control.

Vishwanathan *et al* (1991) invested the G.R and IR to 4 common breakfast items (idly, pongal, upumma and bread) in NIDDM subjects. The breakfast provided 350k.cal of which 68-81 per cent of calories contributed from complex CHO.the results indicated that idly, pongal, upumma were more suitable than bread as their G.R are low. It wa also concluded that a number of parameters like mode of processing ,cooking,the form of food and the differences in food constituents also digestion, absorption and metabolism seem to influence the glycaemic and insulin response.

Kavita and Prema (1997) studied G.R to four lunches (Isocaloric and of 75g CHO) in the form of rice, wheat, ragi and tapioca in 20 NIDDM subjects. This study confirmed that wheat has the least G.R followed by ragi, rice, and tapioca. It was concluded that the high amylase and protein content of wheat were responsible for its low G.I.

Gannon *et al.,* (1998) studied the impact of mixed meal on G.R in NIDDM subjects. The test were first meal contained 43 per cent CHO, 22 per cent PRO,34 per cent Fat compared to a second meal of 55 per cent CHO,15 per cent PRO,30 per cent Fat and third meal of 40 per cent CHO, 20 per cent PRO, 40 per cent Fat. The findings was the plasma glucose area response was smaller after ingestion of low-starch meals compared with high starch high CHO meals.

Buyken *et al.,* (2000) examined the relation of CHO intake (cereal,fruits vegetable, milk and potato) to HbA1C concentration in NIDDM subjects. The result showed that increased intake of total CHO (% of energy) and higher consumption of potato (g) were associated with higher levels of HbA1C. Whereas intake of vegetables (CHO) inversely related to HbA1C. Consumption of cereal and fruits (CHO) was not related o HbA1C.

Rendell *et al*., (2005) compared the G.R and I.R of prowash, a hull less barley with low starch , high fiber , high protein and high concentration of free sugar to oatmeal and a commercial liquid meal replacer (LMR) in diabetic and non diabetic subjects.

The results showed that a substantial reduction of postprandial glycemic peak following ingestion of powarsh was observed as compared to LMR and oatmeal.

Thus various findings have shown that equivalent amount of CHO from different food source do not have an identical effect on blood glucose levels. The emphasis is also laid on the selection of food with low G.I and low glucose content for diabetic.

Protein:

In diabetic dietary management, the protein allowance is essentially the same as that of normal individuals and may vary from 0.8 to 1.4 g of PRO /kg of desirable body weight. However, in the presence of renal failure, a protein intake should be restricted to 0.8 /kg body weight. Thus, the recommended protein intake for people with diabetes is 10-35 per cent of total daily energy undermost circumstance (Institute of Medicine, 2002)

Gannon, *et al.*, (2003)examined the effectiveness of dietary protein on blood G.R twelve subjects with untreated type -2 diabetes were served with a high protein diet (CHO:PRO:FAT in 40:30:30) to control of (55:15:30). The observation was that the high protein diet results a 40 per cent decrease in the mean 24 hour integrated G.R Glycated Haemoglobin also decreased 0.8 per cent and 0.3 per cent after 5 weeks of high protein and control diet respectively. The difference was significant, thus concluded that a high protein diet lowers postprandial blood glucose in type 2 diabetes and improves overall glucose control.

Abdel, *et al.,* (2004) compared the effect of different processed chickpea (germination, soaking and cooking) on blood glucose level with unprocessed chickpea. The findings were the processed chickpea meal had decreased serum glucose as compared to control. So results suggested that soaked, germinated or cooked chickpea could correct metabolic response in diabetic patients

Nicsson, *et al.*, (2004) evaluated the effect of common dietary sources of animal or vegetable protein (reconstituted milk powder, cheese, whey, cod and wheat gluten with lactose) on concentration of postprandial blood glucose, insulin amino acids and incretin hormones (GIP AND GLP-1) in healthy subjects. The finding was the reconstituted milk powder and whey had substantially lower postprandial glucose area than reference bread and whey meal was accompanied with higher AUC for insulin and GIP. A correlation was found between postprandial IR and early increment in plasma amino acids; the strongest correlation were seen for leucine ,valine ,lysine and isoleucine. Thus suggesting that milk proten have insulinotropic properties; the whey fraction containing the predominating insulin secretagogue.

Frid, *et al.,* (2005) further studied the effect of whey on blood glucose and insulin responses to composite breakfast and lunch meals in type 2 daibetic subjects. They were provided with high G.I breakfast (white bread) and subsequent high G.I lunch (Mashed Potatoes with Meatballs) in which whey was supplemented. It was found that insulin response were higher after both breakfast (31%) and lunch (57%) when whey was included with no whey included. After lunch blood glucose response was significantly reduced (21%) after whey ingestion. Postprandial GIP response were higher after whey ingestion whereas no differences in GLP-1 between the reference and test meals.

Manders, *et al.*, (2005) investigated insulin responses and the subsequent plasma glucose disposal rates after the ingestion of CHO alone (0.7g CHO/KG/HT) and with a protein hydrosylated and aminoacid mixture of lysine and phenyl alanine (0.35g/kg/ht) in type-2 diabetes mellitus subjects. They found that plasma glucose response were reduced in CHO+PRO trial than CHO trial. So it was concluded that combined ingestion of CHO with protein hydrosylated and aminoacid mixture significantly increase denovo insulin production in patients with long term diagnosis of type-2 diabetes mellitus. The increased I.R stimulates plasma glucose disposal and reduces postprandial glucose concentration.

Thus, various studies showed that protein content of the diet may blunt the rate of increase in blood glucose by augmenting the plasma insulin levels.

Fat:

Fat is also important in determining the glycemic response like CHO and PRO as it affects the rate of gastric emptying and glucose absorption.

Fukagawa, *et al*., (1990) compared an high CHO-low fat diet (CHO:PRO:Fat in 68:18:14) with a low CHO and high fat diet (43:18:42) in twelve healthy subjects. The results showed that HCLF diet significantly reduced fasting glucose and insulin concentration and increased glucose disposal rates than low CHO and high fat diet. In addition, the HCLF diet enhanced peripheral insulin sensitivity. Similarly, Rajeha

(1991) reported that a high fat diet induces insulin resistance and as a result, glucose uptake by the tissue is impaired and leads to hyperglycemia and hyperinsulinemia.

Milne, *et al*., (1994) compared three diets –a weight maintenance diet, a high CHO diet and high fibre diet, a modified lipid diet on glycemic control. The study confirmed that both the high –CHO and high fibre diet and the modified lipid diet in which monosaturated fatty acids (MUFA) were substituted for saturated fatty acids resulted in similar improvement in LDL cholesterol and glycemic control as measured HbA1C concentration.

Luscombe, *et al*., (1999) similarly compared three diets. A high G.I diet (53 percent CHO, 21 percent fat, 63 G.I unit in reference to glucose=100), Low –G.I diet (51percent CHO, 23percent fat, 43 G.I unit) and a high mono and high G.I diet (42 percent CHO,35 percent fat,59 G.I unit). It was noted that HDL cholesterol was higher on both G.I and high G.I compared to high G.I but no marked effect on glucose metabolism in controlled NIDDM.

Strazinsky, *et al.,* (1999) compared an high CHO low fat diet (CHO: PRO: FAT: in 54:21:25) with high fat diet (36:19:45) in fourteen healthy subjects. It was marked that insulin sensitivity was significantly higher on the HCLF as compared with HF diet.

Stene, *et al.,* (2003) studied the effect of cod liver oil in the onset of childhood typr1 diabetes. It was concluded that frequent use of cod liver oil might reduce the risk of type1 diabetes through the anti-inflammatory effect of long chain (n-3) fatty acids.

Wang, *et al.,* (2003) investigated the relationship of plasma cholesterol (CE) and phospholipid (PL) of fatty acid compositions with the incidence of diabetes mellitus. They concluded that diabetes incidence was significantly and positively associated with the portion of total SFAs in plasma CE and PL. the incidence of diabetes was also positively associated with proportion of palmitic (16:0) palmitoleic (16:1 n-7) and dimono-y-linolenic (20:3 n-6)acids and inversely associated with the proportions of (16:0) and stearic acid.

Since diabetes is associated with a 3-4 fold increase in the prevalence of cardio vascular diseases, the intake of dietary fat both interms of quantity and quality has its own significance. Thus, institute of medicine (2002) recommended that fat content of a diabetic diet should provide 25-30 percent of total calories and saturated fatty acid should not be more than 7 per cent of total calories.

Hence all these studies illustrated that in major nutrients not only CHO but also PRO and fat affect the blood response to a food.

Fiber:

The health related importance of fibre as part of balanced diet has been recognised for decades. The inclusion of dietary fibre in the diet improves many clinical complications including diabetes

Reports of Trowel (1972) and Burkitt *et al.,* (1974) suggested that dietary fiber might prevent hyperlipidemia. These reports have not only stimulated a great deal of research efforts but have also resulted in substantial controversy and speculation.

Jenkins, *et al.,* (1980) compared the effect of high CHO intake in diabetic to see the impact of fibre supplementation on the level of dietary carbohydrate. It was noted that high carbohydrate (> 40percent calorie intake) led to a mean 64 percent reduction in glycosuria than low carbohydrate intake.

Numerous studies have been reported that whole grain breads which contain a high propotion of whole cereal grain may be useful in reducing the post prandial blood glucose profile in diabetics as they are slowly digested (Heinonen, *et al.*,1985, Jenkins, *et al.*,1988 Rasmussen, *et al.,* 1991)

Butchilakshmi (1992) studied the consumption of whole and dehulled jowar recipes in NIDDM subjects.glycemic response of whole jowar recipe was found to be significantly lower than that of jowar recipe. Thus, conclude that loss of fibre during dehulling may be result of this effect. Similar findings were reported with whole bajra and dehulled bajra recipes (Neelima 1999).

Granfelst, *et al.,* (1995) concluded that neither incomplete gelatinisation in rolled oats nor naturally viscous dietary fibre in oats affect postprandial glycemia where as enclosure of intact kernels significantly blunt metabolic responses in healthy subjects.

Lu, *et a*l., (2000) studied the effect of a major component of cereal dietary fibre arabinoxylan (which was extracted from byproduct of wheat flour processing) on postprandial glucose response in normoglycemic subjects. It was found that postprandial glucose and insulin responses were improved by ingestion of such fibre.

Gunde and Plemnitas (2001) reported that oyster mushrooms have hypoglycaemic activity due to their high protein and dietary fibre content.

A followup study conducted by Fung *et al.,* (2002) in men stated that the relative risk of type II diabetes was reduced by increasing the intake of whole grain. Thus concluded that the effect may be mediated by cereal fiber

Juntunen *et al.,* (2003) conducted a 8 week cross over study on high fibre rye bread and white bread in twenty post menopausal women. It was observed that rye bread is effective in enhancing insulin secretion without any effect on insulin sensitivity suggesting an improvement in B cell function.

It has been stated that neutral detergent fibre isolated from blackgram possess a significant hypoglycaemia , hypolipidemic and anti-colon cancer properties (Indria and Kurup,2003)

Scafer, *et al.,* (2003) compared the glycemic response and insulinmeic response to three different meals based on dried peas,potatoes or both in type II diabetic subjects. It was found that postprandial glucose and insulin concentration were delayed and significantly lowered after pea meal followed by combined meal (potatoes and pea) and potato meal. This effect may be due to low glycemic and high fibre content of peas.

Nestel, *et al.,* (2004) compared the effect on insulin sensitivity of chickpeas based meal with wheat based foods and white bread when eaten as single meal or over six weeks. It was found that after single meal the plasma glucose was substantially lower at 30, 60 minutes after chickpea meal than after the other two meals. The plasma insulin and insulin sensitivity index were also lower after 120 minutes. Despite this long term study (over six weeks) failed to show any significant difference between plasma glucose, insulin either in the fasting state or after a glucose load.

Weickert, *et al.,* (2006) reported that insoluble dietary fiber for three days significantly improved whole body insulin sensitivity. Thus, data suggested a potential mechanism between cereal fiber intake and reduced risk of type II diabetes.

A lowering of intestinal reabsorption of endogenous amino acids secreted into the gut has been observed with a fiber –supplemented diet. Any reduction in the intestinal transit time associated with fiber containing diets would allow less time for digestion and absorption of dietary protein.

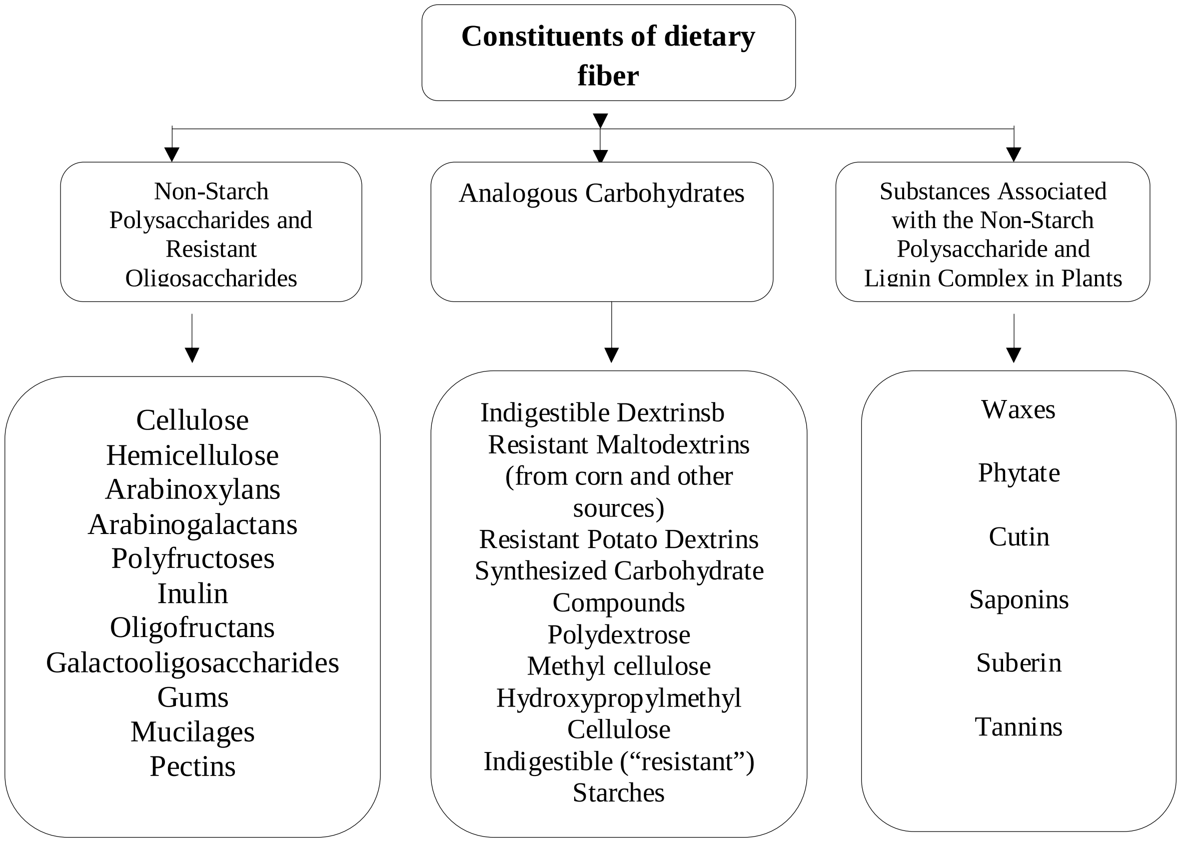

Fig 2.1. Constituents of dietary fiber

Sorghum contained 2.5 to 9.0% total dietary fiber and the soluble fraction was about 0.5% of all flours with different extraction rates. The increase in the insoluble fraction was almost proportional to the extraction rate. The dietary fiber in sorghum flour of low extraction consisted mainly glucans.

Several studies reported processing effects on fiber content of millets and sorghum. Phusphamma (1993) found that when sorghum grains were subjected to different processing conditions Crude fiber content decreased significantly on parching, malting, and parboiling, the lowest being in malted grain, where the crude fiber in grain i.e., 2.0% was decreased to 1.4% after parboiling.

Ramulu and Rao (1997) reported that processing of cereals had no effect on their dietary fiber. Before processing dietary fiber content of cereals ranges from 12.5(wheat) to 4.1% (rice), when they were processed into product i.e., chapathi no significant difference was observed in fiber content of products.

2.4. SORGHUM A STAPLE GRAIN FOR DIABETIC

Sorghum bicolor (L.) Moench is a cereal in the grasses (Poaceae) family. The grain is native to the tropical areas of Africa, and was first domesticated around 3,000 to 5,000 years ago (U.S. Grains Council, 2008). Today, the cultivated species, *S. bicolor*, has been further divided into four classifications of sorghum by intended purposes: grain, sweet, broom and grass. Sweet sorghum is used for producing sweetener syrup; broom sorghum is used for making brooms. Grass sorghum's purpose is for feed and forage use, while grain sorghum is mainly used as a human food source, as well as a raw material for alcoholic beverages, sweets, and glucose production (U.S. Grains Council, 2008).

Sorghum production:

In India, sorghum grains are mostly used for the *roti* preparation. M 35-1 (*Maldhandi*) is a sorghum cultivar known for its good quality of *roti* due to pearly white grain color, its flour having higher water holding capacity, and good organoleptic taste (Chavan, *et al*., 2009).

Limitation for the use of sorghum is poor digestibility compared to other cereals. Sorghum generally has the lowest starch digestibility compared to other cereals (Zhang and Hamaker, 1998). The lower digestibility of cooked sorghum starch has been shown to affect the feeding value in livestock (Rowe *et al*., 1999), and to cause a higher loss of energy in humans (MacLean *et al.,* 1981). Sorghum endosperm proteins play an important role in restricting the availability of starch (Rooney and Pflugfelder, 1986). Lower protein digestibility by pepsin is due to the resistance of Kafirins, the major storage protein constituting the protein body and matrix with many intra-and iner-disulfide bonds.

Protein quality of sorghum can be up graded to meet human physiological requirements by compositing with legume flours using the principle of complementation (Young and Pellet, 1994). Formulation of foods from low lysine staples fortified with legumes has been proposed as most practical and sustainable approach to improving the protein nutritional value of foods for young children in developing countries (FAO/WHO, 1994).

The production of sorghum – legume composite biscuits has been reported by Hikeczi (1994) who supplemented sorghum with peanut or sunflower flours raising the protein content to 16%. Mridula *et al*., (2007) showed that biscuits of acceptable quality can be made using wheat – sorghum composites with 10 to 50% sorghum and 5% defatted soy flour.

Sorghum utilization:

Many populations in Africa and India have consumed sorghum for thousands of years, sorghum is used in many traditional foods, including porridges, fermented and non leavened breads, rice like products(ex: couscous), alcohol and non alcoholic beverages, snakes, and many bread products.

Industrial production of sorghum beer, non-alcoholic beverages and porridges occurs in Africa (especially Nigeria), South Africa and Botswana. Malt extracts are used extensively or non-alcoholic beverages and commercial products in Africa. African and Indian cultures generally prefer white kernals with a tan plant and glume color. Soft white sorghums grown under dry conditions during grain maturation and harvesting often are preferred. In India, rabbi (dry season) sorghum of the M 35 -1 types often are preferred over wheat for milling into whole grain flour for roti or chapattis (flat bread). They are used in many foods, including rice substitutes, thick and thin porridges, snacks, and other products (Roony and Awika, 2005).

Composition

Starch is the main component of sorghum grain, followed by proteins, non-starch polysaccharides (NSP) and fat the average energetic value of whole sorghum grain flour is 356 kcal/100g (BSTID-NRC, 1996). Sorghum has a macromolecular composition similar to that of maize and wheat (BSTID-NRC, 1996). Sorghum contains non-starch polysaccharides (NSP), mainly located in the pericarp and endosperm cell walls, with proportions in the kernel ranging from 2 to 7% depending on variety (Knudsen and Munck, 1985., Verbruggen *et al*., 1993).

The NSP in sorghum grain are essentially constituted of arabinoxylans and other beta glucans representing 55% and 40% of the total NSP (Verbruggen *et al.*, 1993., Hatfield *et al.*, 1999).

Verbruggen and co-workers (1993, 1998) found arabinoxylans from sorghum to be glucuronoarabinoxylans containing ferulic acid and *p*-coumaric acid. Arabinoxylans, being one of the major NSP present in sorghum cell walls, play an important role in the processing of sorghum for baking and brewing (Verbruggen *et al.,* 1998).

The other b-glucans comprise cellulose (1, 4-b-D-glucans), curdlan-type glucans (1, 3-b-D-glucans), and lichenantype glucans (1, 3; 1, 4-b-D-glucans) (Knudsen and Munck, 1985., Verbruggen *et al*., 1993, 1998).

These b-glucans are predominantly water-unextractable, and form viscous and sticky solutions. In brewing, together with arabinoxylans, they are associated with processing problems like poor wort and beer filtration rates and the occurrence of haze (Aisien and Muts, 1987).

Sorghum also contains non carbohydrate cell-wall polymers such as lignin's with proportions constituting up to 20% of the total cell wall materials (Hatfied, *et al.,* 1999).

The protein content in whole sorghum grain is in the range of 7 to 15% (FAO, 1995., Beta *et al* 1995). Using the solubility-based classification (Jambunatan, *et al*., 1975), sorghum proteins have been divided into albumins, globulins, kafirins (aqueous alcohol-soluble prolamins), cross-linked kafirins and glutelins. The kafirins comprise about 50-70% of the proteins (Hamaker, *et al.,* 1995., Oria, *et al,.,* 1995., Duodu, *et al*., 2003). a- Kafirins (23 and 25 kDa) make up about 80% of the total kafirins and are considered the principal storage proteins of sorghum, whereas b-kafirins (16, 18, and 20 kDa), and g-kafirin (28 kDa) comprise about 5% and 15% of total kafirins, respectively. The nutritional quality of sorghum proteins is poor because these kafirins are protease resistant (Badi, *et al.,* 1990., Oria *et al.* 1995., Anglani, 1998). However, a wide variability according to variety has been observed with respect to the levels of proteins in sorghum (Reddy and Eswara, 2002). The protein digestibility of sorghum may decrease upon cooking (Axtell *et al.* 1981. Taylor and Taylor, 2002), but pre fermentation may increase the digestibility (Taylor and Taylor, 2002). The low digestibility is due to protein-protein, protein-carbohydrate, protein-(poly) phenol and carbohydrate-(poly) phenol interactions (Knudsen, *et al.,* 1988., Axtell, 1981., Cherney, *et al.,* 1992., Taylor and Taylor, 2002).

The fat in sorghum grain (mainly present in the germ) is rich in polyunsaturated fatty acids (Glew, *et al.,* 1997). The fatty acid composition of sorghum fat (linoleic acid 49%, oleic 31%, palmitic 14%, linolenic 2.7%, stearic 2.1%, etc.) is similar in content to that of corn fat, but it is more unsaturated (Knudsen, *et al.,* 1988., Adeyeye and Ajewole, 1992., FAO, 1995)

Sorghum is a good source of vitamins, notably the B vitamins (thiamin, riboflavin, pyridoxine, etc.), and the liposoluble vitamins A, D, E and K. Sorghum is reported to be a good source of more than 20 minerals (BSTID-NRC, 1996). Sorghum is also rich in phosphorus, potassium, iron and zinc (Glew *et al.* 1997., Anglani, 1998).

Zinc (an important metal for pregnant women) deficiency is more common in corn and wheat than in sorghum (Hopkins *et al*. 1998).

Table 2.1. Proximate composition of sorghum grain

Macro-components (g/ 100g f. m.)		**Vitamins (mg/100g d. m.)**		**Minerals (mg/ 100g d. m.)**	
Carbohydrates	65-80	Vit A	21RE	Ca	21
Starch	60-75	Thiamin	0.35	Cl	57
Amylase	12-22	Riboflavin	0.14	Cu	1.8
Amylopectin	45-55	Niacin	2.8	Iodine	0.029
Non starch	2-7	Pyridoxine	0.5	Fe	5.7
Low Mw CHO	2-4	Biotin	0.007	Mg	140
Proteins	7-15	Pantothenat	1.0	P	368
Alpha kefrins	4-8	Vit C	<0.007	K	220
Beta kefrins	0.2-0.5			Na	19
Gama kefrins	0.7-1.6			Zn	2.5
Other proteins	2-5				
Fat	1.5-6				
Ash	1-4				
Moisture	8-12				

Sources: Verbruggen, *et al.*, (1993), FAO (1995), Hamaker, *et al.*, (1995), BSTID-NRC (1996), Glew, *et al.*, (1997), Duodu, *et al.*, (2003), Dicko, *et al.*, (2006).

*Not strictly essential amino-acids, **RE = retinol equivalent; f.m. = fresh matter, d. m. = dry matter; NSP = non starch polysaccharides

Kafrins:

Kafirins, comprising 70–80% of the protein in whole grain sorghum flour (Hamaker *et al.,* 1995), are synthesized and translocated into the lumen of the endoplasmic reticulum where they form protein bodies (Taylor *et al.*, 1985). They are sub classified as: a- (23 and 25 kDa), b- (20 kDa) and g- (28 kDa) types based on molecular weight, extractability, structure and cross-reactivity with sera against analogous maize zeins (Belton, *et al.,* 2006., Mazhar, *et al.,* 1993., Shull *et al.,* 1991).

Comprising 80% of total kafirins, the a-type is considered the principal storage protein, followed by the g- (w15%) and b- (w5%) members. Recently, a DNA-derived sequence for a d-kafirin was shown to have high homology with the Mr 10,000 d-zein, except for absence of part of the methionine-rich region (Belton *et al.* 2006., Izquierdo and Godwin, 2005). a-Kafirins have low levels of cysteine relative to the b- and g-types (5 and 7 mol%, respectively), whereas d-kafirins are rich in methionine (16–18 mol%) but lack cysteine.

Electron microscopy of internal protein body structure reveals that g-kafirins, and to a lesser extent b-kafirins, encapsulate the more digestible a-kafirins in a disulfidebond polymer network (Shull *et al.,* 1992), there by impeding exposure to proteases.

Amino acid composition of sorghum protein:

The composition of the indispensable amino acids of a protein is an important indicator of its protein nutritional value. Like other cereals, lysine is the first limiting amino acid in sorghum relative to the WHO (2007) reference pattern and is much compared to egg protein. Sorghum has the lowest lysine content of approximately 2.1% when compared to the other cereals rice, wheat and maize with lysine contents of approximately 3.5%, 3.0% and 3.4% respectively (Young and Pellet, 1985).

Further in the sorghum kernel, the lysine rich and globulin predominate in the germ pericarp and debranning of sorghum reduces a substantial amount of the lysine content. (Taylor and Schussler, 1986)

2.5. IN-VITRO STARCH DIGESTIBILTY

Starch is the primary source of stored energy in cereal grains. Starch is deposited as granules in the endosperm cells, being the main constituent of the endosperm. Sorghum starch granules have diameters ranging from 5 to 25 µm (average 15 µm). Sorghum starch has a specific particularity because of its high gelatinization temperature (70-75ºC), which decreases its industrial application (Dufour, *et al.,* 1992., Taylor, 1992). Native starch granules are essentially insoluble in cold water. The term "gelatinization" is used to describe the swelling and hydration of granular starches (Zobel, 1984). Starch gelatinization is the disruption of molecular orders within the starch granule manifested in irreversible changes in properties such as granular swelling, native crystalline melting, loss of birefringence, and starch solubilization. These changes render all or part of the material in granules soluble and consequently enable to contribute to food properties such as texture, viscosity, and moisture retention (Whistler and BeMiller, 1997).

The point of initial gelatinization and the range over which it occurs is governed by the starch structure. Sorghum starch is classified as type-B, e.g. a moderate swelling starch compared to type-A starches (potato, tapioca, waxy sorghum, etc.), which are high swelling starches (Beta and Corke, 2001). The retrogradation involves re association of the molecules and occurs when the starch is cooled, and this is dependent on the ratio of amylose and amylopectin. Enzymatic sorghum starch hydrolysis or chemical treatment can improve its technological properties (Zhang and Hamaker, 1999).

Regardless of the botanical source, starch is structurally composed of two high molecular weight homopolysaccharides known as amylose and amylopectin. Amylose content in mature sorghum grain is varietal dependent. While waxy sorghums do not contain amylose (level < 1%), the content of amylose in normal sorghums is ranging from 10 to 17% (w/w, fresh weight basis), constituting approximately 20-30% of starch. There is no significant difference between red and white sorghum grains in their starch contents (Dicko, *et al.*, 2006). The screening of starch content in 50 sorghum varieties before and after germination showed that there is an inter-varietal difference of content in these compounds (Dicko, *et al.*, 2006).

Amylose is composed of essentially homogenous linear units of a-(1_4)-D-glucopyranose, which can form helicoidal structures in solution (Manners, 1974., Jarvis and Walker, 1993). The interior of the helix is hydrophobic, allowing amylose to form a complex with free fatty acids, iodine, etc. (Fennema, 1985). There is a significant inter-varietal difference of content of amylase among sorghum varieties (Beta and Corke , 2001., Dicko, *et al.*, 2006).

Although, some varieties contained relatively little amylose, waxy sorghum were not found among cultivated sorghums in West Africa (Dicko, *et al.,* 2006). This is probably because cultivated varieties were primarily selected and bred for tô, for which high amylase content is required (Bello, *et al.,* 1990).

Amylopectin is constituted of short chains of a-(1_4)- D-glucopyranose (majority 10-20 units in sorghum starch) branched to a-(1_6)-D-glucopyranoses to form a highly ramified structure (Blennow *et al.* 2001). The content of amylopectin is varietal dependent. The content of amylopectin in sorghum is ranging from 45 to 54% (w/w, fresh weight basis). Contrary to amylose the levels of amylopectin in sorghum varieties are not significantly different (Dicko *et al.* 2006).

There are relationships between the levels of starch component and sorghum utilization for several foods. Content of starch and starch components such as amylose and amylopectin may give directions for the selection of sorghum varieties for specific foods (Dicko, *et al.,* 2006). For instance, in to preparation, the formation of a thick paste linked to high amylose content is necessary. On the other hand, sorghum varieties with low viscosity are desired in the formulation of weaning foods with high energy density (WHO, 1998).

In that case low amylose content and high amylase activities are determinant. Since amylose has a higher gelatinization temperature than amylopectin (Whistler, *et al.,* 1984), sorghum with low amylose content could be targeted for industrial brewing.

Amylose is more susceptible to retrogradation than amylopectin and waxy sorghum is less viscous than normal sorghum (FAO, 1995). Low amylose-containing sorghum varieties are also preferred for extrusion-cooking because they give better functional characteristics of the extrudates, such as enzyme susceptibility and solubility (Gomez *et al.,* 1988). Sorghum varieties with low amylose content may be recommended for infant porridges preparation.

Starch digestibility:

In the corneous endosperm, non-kafirins (albumins, globulins, glutelins) form around protein bodies, effectively ‘‘gluing’’ the bodies into a matrix surrounding the starch granules (Hamaker and Bugusu, 2003., Shull *et al.* 1990., Taylor *et al.* 1984). This protein matrix appears to act as a barrier to starch gelatinization and digestibility

(Chandrashekar and Kirleis, 1988., Duodu *et al.* 2002., Ezeogu *et al.* 2005, 2008) due to cross-linking between g- and b-kafirins and matrix proteins (Duodu *et al.* 2001., Hamaker and Bugusu, 2003).

Cooking reduces digestibility by effecting a conformational change in proteins that could facilitate formation of disulfide-linked polymers (Axtell *et al.* 1981., Duodu *et al.* 2002, 2003., Hamaker *et al.* 1987., Oria *et al.* 1995b). The negative impact of cooking on protein digestibility was mitigated by addition of 2-mercaptoethanol (ME) or other reducing agents (Elkhalifa *et al.* 1999., Hamaker *et al.* 1987).

Sorghum grains rich in kafirin-containing protein bodies also have a lower capacity for starch gelatinization (Chandrashekar and Kirleis, 1988., Ezeogu *et al.* 2005, 2008) an observation consistent with the finding that adding ME during cooking to cleave disulfide bonds within the protein matrix increased the degree of starch gelatinization and digestion (Elkhalifa *et al.* 1999., Ezeogu *et al.* 2005, 2008., Zhang and Hamaker, 1998).

The protein barrier surrounding the starch granule may also reduce the hydrolysis of native and processed starch by amylolytic enzymes (Rooney and Pflugfelder, 1986). Thus, the addition of pronase to hydrolyze the protein matrix significantly enhanced in vitro rates of starch hydrolysis by increasing surface area and enabling starch to interact with a-amylase and amyloglucosidase (Rooney and Pflugfelder, 1986). Similar effects were observed when sorghum flour was either treated with pepsin before cooking or cooked in the presence of dithiothreitol (DTT) or other reductants (Zhang and Hamaker, 1998). Abundance of starch granules may also decrease proteolysis by limiting accessibility of proteolytic enzymes, especially when gelatinized during cooking (Duodu *et al.* 2002). Uniqueness of the protein matrix and its interaction with starch that affect the rate of starch digestion are key differences between the feed quality of sorghum and corn (Rooney and Miller, 1982., Rooney and Pflugfelder, 1986). In addition, endosperm texture and cooking conditions have been shown to have a significant effect on in vitro digestibility of starch and protein in these two cereals (Duodu *et al.* 2002, 2003., Ezeogu *et al.* 2005, 2008).

Sorghum displays significant variation in rates of starch disappearance (Wester *et al.* 1992). Based on a 12-h incubation in the in vitro dry matter disappearance (IVDMD) assay, loss of dry matter in sorghum correlated closely with rates of starch, but not necessarily protein digestion (Pedersen *et al.* 2000).

Effect of processing on in vitro starch digestibility:

Processing increases the enzymatic availability of starch at different extents. An increased availability may be attributed to several factors such as gelatinization, disruption of the protein structure and cell walls encapsulating starch, expansion and physical disruption of the samples. Starch digestibility was increased when foods subjected to conditions that increased the accessibility to amylase. The percent starch digested varied depending on the method of cooking. The IVSD after processing was increased to different extents by extrusion cooking, popping and flaking of cereals. (Malleshi *et al.* 1996).

Germination of sorghum significantly improved the IVSD (Bhise *et al.* 1998) increase in the IVSD on germination was probably due to increase in the enzymatic activity. (Kshirsagar, 1994).

Osman *et al.* (1996) reported flaking affects on digestibility of starch in sorghum and barley. Steaming or pressure cooking at 1.4 kg/cm2 for 1 minute without flaking decreased in vitro starch digestibility compared to flaked grains after steaming or pressure cooking.

Starch digestibility and health:

General recommendations for carbohydrate sources are to consume whole-grain cereal products and products rich in dietary fiber (Dietary Guidelines for Americans, Anonymous 2005). Among the health benefits from wholegrain/ high-fiber foods is their slower digestion and absorption compared to refined products.

Dietary carbohydrates can be classified in many different ways; simple or complex, sugars or starches, available or unavailable. However, besides the classification on the basis of their chemical characteristics, the FAO/WHO has classified carbohydrates considering their physiological properties. Therefore, carbohydrates can be classified according to their potential impact on blood glucose, which can be measured as a “glycemic response” or glycemic index.

For nutritional purposes, Englyst *et al.* (1992) proposed a classification of starch based on the rate and extent of the starch digestion and developed an in vitro enzymatic method to measure rapidly digestible starch (RDS), slowly digestible starch (SDS), and resistant starch (RS). They concluded that the RDS has a large impact in the glycemic response in humans, while the impact from SDS is small.

The RS escapes digestion in the small intestine (may be digested in the large intestine), without causing a glycemic response.

Chapter III

MATERIAL AND METHODS

The present research study was designed to assess the glycemic response to breakfast product idli made with rice rawa and jowar rawa in young adult subjects. The details of the methodology are given under the following heads.

3.1 LOCATION OF STUDY

3.2 PROCUREMENT OF RAW MATERIAL

3.3 DEVELOPMENT OF THE PRODUCT

3.4 STUDY OF GLYCEMIC INDEX

3.4.1 SELECTION OF SUBJECTS

3.4.2 METHOD OF GLYCEMIC INDEX

3.5 DETERMINATION OF GLYCEMICINDEX

3.6 ESTIMATION OF INVITRO STARCH DIGESTIBILITY

3.7 STATISTICAL ANALYSIS

3.1 LOCATION OF STUDY:

The product was developed and analysis of the selected product were planned and conducted in the department of Foods And Nutrition, Post Graduate and Research Centre, Acharya N. G. Ranga Agricultural University, Rajendranagar, Hydrerabad. The Glycemic index trials were conducted in Vasantha nilayam ladies hostel, ANGRAU, Rajendranagar, Hyderabad.

3.2 PROCUREMENT OF RAW MATERIAL

The ingredients required for the preparation of idli were obtained from the local market in bulk from Hyderabad. The jowar rawa for making idli was procured from DIRECTORATE OF SORGHUM RESEARCH (DSR) Rajendranagar Hyderabad. Non-dehulled jowar rawa was used for the present study.

3.3 DEVELOPMENT OF THE PRODUCT

TABLE 3.1 DEVELOPMENT OF IDLI USING RICE RAWA AND JOWAR RAWA

Type of idli	Rice rawa	Jowar rawa	Black gram dal	Total Grams	Available CHO in Grams
Rice rawa idli	49 gm	-	24.5 gm	73.5 gm	50gm
Jowar rawa idli	-	47gm	23.5 gm	70.5 gm	50gm

Method of preparation

Black garm dal was soaked in water for four hours and then made into a smooth paste. The test rawa (rice rawa or jowar rawa) and salt were mixed with the paste and then made into a thin batter. The batter was left overnight to ferment. Then idlis were prepared with the batter by following ususal method of preparation. The idlis were seved with tomato chutney.

3.4 STUDY GLYCEMIC INDEX

Glycemic index of IDLI was estimated using glucose as reference food.

3.4.1 Selection of subjects

Initially fifteen members were selected from Vasantha Nilayam Ladies hostel at ANGRAU campus with the age group of 22-23 years. Subjects were excluded if they reported a history of gastrointestinal disorders, suffered from diabetes, were taking medication for any chronic disease conditions, or intolerant or allergic to any of the foods.

Finally, ten healthy subjects were identified for study from initial 15 members. Before starting the glycemic index trial, subjects were explained about the study and few instructions were given to follow.

3.4.2. Method for Glycemic Index

The method used for measuring and calculating the GI of the idli was in accordance with WHO/FAO recommendations (FAO/WHO, 1998). Subjects attended each testing session after 10hrs overnight fast but not exceeding 16 hrs and had been instructed not to consume unusually large meals and exercise vigorously on the previous day. On the first six occasions, the subjects were given the standard reference food (Glucose). The 50g glucose was made up with 250 ml water, and subjects were given 250 ml of water to drink.

Again on the next six occasions, 73.5gm of experimental rice rawa idlis (seven idlis) was given to provide 50 g of carbohydrates for each subject.

Again, on the next six occasions, 70.5gm of experimental jowar rawa idlis (six idlis) was given to provide 50 g of carbohydrates for each subject.

Blood glucose levels were measured by using Horizon one touch Glucometer in capillary whole blood obtained by finger prick in the fasted state and at 30, 60, 90 120 mins after the consumption of the idli.

3.5 DETERMINATION OF GLYCEMIC INDEX

The incremental area under two hour glucose response curve (IAUC) was calculated according to the formula used by Wolever et al (1991). Glycemic index of the test idlis (made with rice rawa and jowar rawa) were calculated by applying the following formula.

$$\text{Glycemic index} = \frac{\text{IAUC of test food}}{\text{IAUC of reference food}} \times 100$$

Glycemic load = **GI**/100 $\times$ dietary carbohydrate content of serving

3.6 ESTIMATION OF INVITRO STARCH DIGESTIBILITY

In vivo methods which involve studies of postprandial glucose and insulin response to starchy foods are in many aspects laborious and demand several motivated subjects during a long period of time. (Gramfeldt *et al.*, 1992) .Strict regulations are required when using human subjects and this makes the situation difficult as some people may decide not to adhere to the regulations such as diet and medication.

There is also a common problem of lack of facilities for most of the laboratories involved in food research.

In vivo digestibility is automatically modified by numerous stimuli for eg; Food itself. This means that enzymes which are with protein, carbohydrate and lipid digestion adapt to any changes in substate intake (Corring *et al.*, 1989). Also antinutritional factors and dietary fiber affect enzyme secretions; it is on these grounds that several scientists have concluded that the in vivo conditions can be completely stimulated under in vitro conditions (Boisen and Eggum, 1991., Granfeldt *et al.*, 1992).

Advantages of in vitro techniques are; they are simple techniques, can be designed to use specific enzymes either to give maximal digestibility values or to measure the initial rate of hydrolysis. In both cases, the enzymes used should have specificities similar to those which are present in the digestive tract (Boisen and Eggum, 1991).

IVSD: In vitro starch digestibility was estimated as mg of maltose released per gram sample according to the procedure of Singh and Jambunathan (1975).

Reagents

1. 0.2 M phosphate buffer pH 6.9:

   A = 0.2 M Na_2HP0_4, $2H_20$

   B = 0.2 M $NaH2P0_4$, $2H_20$

   45 ml A and 55 ml of B were diluted to 200 ml and pH was adjusted.

2. 3-5 dinitro salicylic acid (DNSA) reagents: 2g of DNSA was dissolved in 40 ml of water. A solution of 3.2g of sodium hydroxide in 30 ml of water was added

drop wise while stirring and gently heated on water bath until a clear solution was obtained. 60g of potassium sodium tartarate was added in small portions and water was added to the final volume of 200 ml. The reagent is stored in dark.

3. Enzyme buffer: 20mg of pancreatic amylase was dissolved in 50 ml of the phosphate buffer and filtered through No 41 filter paper.

4. Standard maltose: 100mg of maltose was dissolved in 100ml of water.

Procedure

25 mg of sample was dispersed in 1.0 ml of 0.2 M phosphate buffer. pH 6.9. 0.5 ml of enzyme buffer was added to the sample suspension and incubated at 37^{o}C for 2 hrs. After the incubation period, 2 ml of 3-5 DNSA reagent was quickly added and the mixture was heated for 5 min in a boiling water bath and after cooling the solution was made up to 25 ml with distilled water and filtered. The absorbance was measured at 550 nm. A blank was run simultaneously. 4ml of maltose standard was taken and the reaction from the addition of 3-5 DNSA standards was followed. The values were expressed as mg of maltose released /g sample.

3.7 Statistical analysis

The results of the study were subjected to statistical analysis with the following statistical tests.

- Means and Standard deviation was used for blood glucose levels of the subjects.
- Pearson correlation was used for IVSD and GI.

Fig 3.1 Experimental Design of The Study

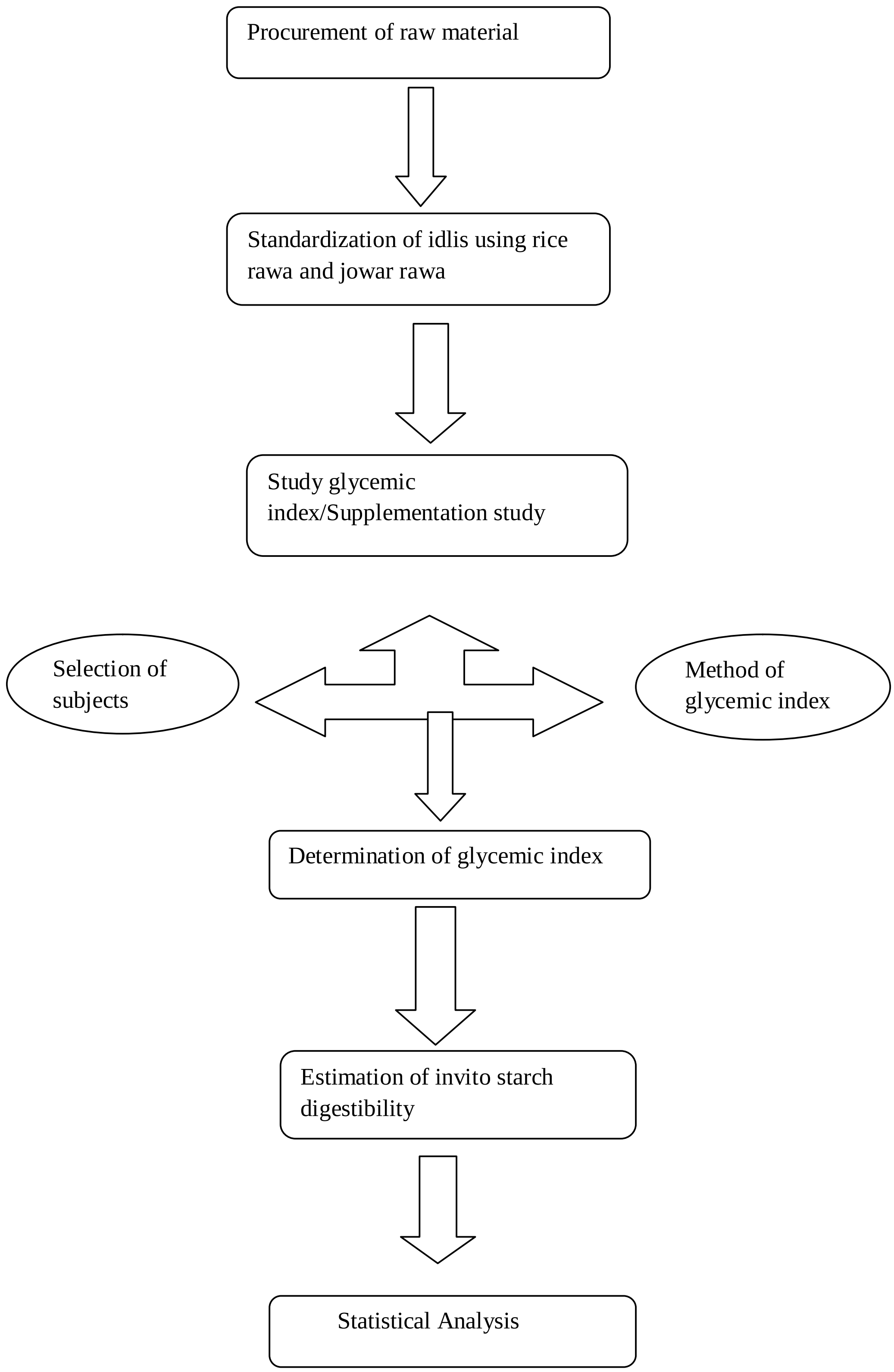

Fig. 3.2 Glucometer

Fig. 3.3 Blood Glucose Estimation

Fig. 3.4 Rice Rawa Idli Served With Tomato Chutney

Fig. 3.5 Jowar Rawa Idli Served With Tomato Chutney

Chapter IV

RESULTS AND DISCUSSION

The present investigation was undertaken to study correlation between the glycemic index and invitro starch digestibility of idli a breakfast item. The results obtained in the present study are presented in this chapter under the following sub heads.

4.1 DEVLOPMENT OF THE IDLI A BREAKFAST PRODUCT

4.2 EFFECT OF IDLI A BREAKFAST PRODUCT

4.3 GLUCOSE RESPONSE OF SUBJECTS

4.3.1 GLUCOSE TOLERANCE TEST (GTT) /REFERENCE TEST

4.3.2 GLYCEMIC RESPONSES OF SUBJECTS FED ON IDLI MADE WITH RICE RAWA

4.3.3GLYCEMIC RESPONSES OF SUBJECTS FED ON IDLI MADE WITH JOWAR RAWA

4.4 DETERMINATION OF GLYCEMIC INDEX AND GLYCEMIC LOAD ON EXPERIMENTAL IDLI

4.5 INVITRO STARCH DIGESTIBILTY

4.6 CORRELATION OF GLYCEMIC INDEX AND INVITRO STARCH DIGESTIBILTY

4.7 EFFECT OF IVSD ON GI FOR IDLI MADE WITH RICE RAWA

4.8 EFFECT OF IVSD ON GI FOR IDLI MADE WITH J0WAR RAWA

4.1 DEVLOPMENT OF THE IDLI A BREAKFAST PRODUCTS

Two types of idlis (idli with rice rawa and idli with jowar rawa) were selected. The products were standardized.

Table 4.1: STANDARDIZATION OF THE PRODUCT

Type of idli	Rice rawa	Jowar rawa	Black gram dal	Total Grams	Available CHO in Grams
Rice rawa idli	49 gm	-	24.5 gm	73.5 gm	50gm
Jowar rawa idli	-	47gm	23.5 gm	70.5 gm	50gm

Method of preparation

Black garm dal was soaked in water for four hours and then made into a smooth paste. The experimental rawa (rice rawa or jowar rawa) and salt were mixed with the paste and then made into a idli batter. The batter was left overnight to ferment. Then idlis were prepared with the batter by following usual traditional method of preparation (steaming).idlis are served with small amount of tomato chutney.

4.2 EFFECT OF IDLI A BREAKFAST PRODUCT.

During supplementation phase the idli made with rice rawa (five in number) and jowar rawa (six in number) were given to subjects and the glycemic response was tested. It was observed that all the test products were well accepted and consumed by all the subjects within the allotted time (10-15 min). No discomforts were reported following the consumption of any of the test idlis. The quantity served to the subjects was found to be sufficient and same was reported by subjects.

4.3 GLUCOSE RESPONSE OF SUBJECTS: The glucose responses of subjects were explained under different heads.

4.3.1 GLUCOSE TOLERANCE TEST (GTT)

An oral glucose test (OGTT) was conducted for all the selected subjects (ten) to assess the metabolic response to glucose load and for comparison with G.R to breakfast products. (This was done prior to actual treatment) Reference test with glucose was conducted for all selected 10 healthy subjects on six occasions to assess the glycemic response of glucose that was administered. This was used as a tool to compare the individual differences in the glycemic response to experimental product. The average blood glucose levels of six occasions for 50g glucose administration are presented in Table 4.2.

TABLE 4.2: PLASMA GLUCOSE VALUES OF THE SUBJECTS ON OGTT

Subjects	**Blood Glucose levels (mg/dl)**				
	Time Intervals (mins)				
	Fasting	**30**	**60**	**90**	**120**
1	107	220	146	138	136
2	99	186	166	160	111
3	81	187	212	146	135
4	95	195	120	223	146
5	98	185	140	124	77
6	78	125	162	148	129
7	96	152	132	117	105
8	89	131	119	70	92
9	94	190	185	140	102
10	98	185	140	124	77
Mean +_SD	**93.5 +_ 8.68**	**157.6 +_ 57.9**	**152.2 +_ 29.4**	**139 +_ 38.4**	**111 +_ 24.8**

From the table 4.2 indicates that the ranges of fasting plasma glucose levels are from 78 to 107 mg/dl. At half an hour interval the plasma glucose levels were increasing in an average of 64.1 mg/dl than that of fasting levels. The levels were increased after 30 mins. of consumption of glucose.

At 60 mins. the glucose levels were decreased in all subjects except one. At 90 mins the levels were decreased in all subjects except one.

At 120 min, there was a decrease in blood sugar levels in all the subjects and the levels were more than fasting levels except in two subjects. The blood glucose levels of all subjects reached its peak at 30 mins.

4.3.2 GLYCEMIC RESPONSES OF SUBJECTS FED ON IDLI MADE WITH RICE RAWA

The blood glucose levels of the subjects on one of six occasions at five different intervals after consumption of experimental product were presented in Table 4.3. The subjects were given five idlis made with rice rawa to provide 50g of carbohydrate during the GI trial.

TABLE 4.3: PLASMA GLUCOSE VALUES OF THE SUBJECTS AFTER CONSUMING IDLI WITH RICE RAWA

Subjects	**Blood Glucose levels (mg/dl)**				
	Time Intervals (mins)				
	Fasting	**30**	**60**	**90**	**120**
1	108	157	147	136	133
2	90	140	114	108	102
3	92	139	136	129	124
4	105	115	113	132	102
5	77	87	129	107	123
6	82	127	134	111	82
7	101	120	140	112	125
8	90	133	92	101	91
9	92	139	136	129	124
10	77	87	129	107	123
Mean +_SD	**91.4 +_ 10.8**	**124.4 +_ 22.8**	**127 +_ 16.2**	**117.2 +_ 12.3**	**112.9 +_ 17.2**

The table 3 indicates that the ranges of fasting glucose levels are from 77- 108 mg/dl. At 30 mins the glucose levels of the subjects were increasing in an average of 33mg/dl and at 60 mins the levels were at the peak with an increase with an average of 35.6 mg/dl.

At 90 mins the levels were decreased in subjects except in one subjects.

At 120 mins there was a decrease in blood glucose levels except in three subject. The blood glucose levels of six subjects reached its peak at 30 mins where as for other subject the levels reached at 60 mins.

4.3.3 GLYCEMIC RESPONSES OF SUBJECTS FED ON IDLI MADE WITH JOWAR RAWA

The blood glucose levels of the subjects on one of six occasions at five different intervals after consumption of experimental product (idli made with jowar rawa) were presented in Table 4. The subjects were given six idlis made with jowar rawa to provide 50g of carbohydrate during the GI trial.

TABLE 4.4: PLASMA GLUCOSE VALUES OF THE SUBJECTS AFTER CONSUMING IDLI WITH JOWAR RAWA

Subjects	**Blood Glucose levels (mg/dl)**				
	Time Intervals (mins)				
	Fasting	**30**	**60**	**90**	**120**
1	113	146	121	109	108
2	92	129	107	105	95
3	82	105	104	100	94
4	94	135	113	92	91
5	84	115	119	119	118
6	89	137	146	120	72
7	92	122	123	105	110
8	83	116	94	105	96
9	82	105	104	100	94
10	84	115	119	119	116
Mean +_SD	**89.5 +_ 9.4**	**122.5 +_ 22.8**	**117 +_ 15.3**	**107.4 +_ 9.3**	**99.4 +_ 17.2**

The table 4 indicates that the ranges of fasting glucose levels are from 82- 113 mg/dl. At 30 mins the glucose levels of the subjects were increasing in an average of 33mg/dl and at 60 mins the levels were increased with an average of 27.5mg/dl.At 90 mins the levels were decreased in subjects except in one subjects.

At 120 mins there was a decrease in blood glucose levels except in one subject. The blood glucose levels of six subjects reached its peak at 30 mins where as for other subject the levels reached at 60 mins.

4.3.4 Mean IAUC of the IDLI'S

The incremental area under the curve (IAUC) was calculated according to the formula used by Wolever *et al* (1986). The IAUC values for the idlis and glucose are given in Table 4.5

Table 4.5. MEAN INCREMENTAL AREA UNDER CURVE OF THE EXPERIMENTAL IDLIS (IDLI MADE WITH RICE RAWA AND IDLI MADE WITH JOWAR RAWA)

Product	**Mean IAUC (mg/dl)**
Glucose	1110
Idli made with rice rawa	592.5
Idli made with jowar rawa	562.5

It can be seen from Table 4.5 that the mean total IAUC of experimental idli with jowar rawa is least when compared to idli made with rice rawa and glucose.

4.4 DETERMINATION OF GLYCEMIC INDEX AND GLYCEMIC LOAD ON EXPERIMENTAL IDLI

The glycemic index ranks carbohydrates on a scale of 0 to 100 based on changes in blood sugar levels after eating. Foods with a high GI are thought to be rapidly digested and absorbed, thereby leading to marked fluctuations in blood sugar levels. Low glycemic index values are thought to produce more gradual rises, and thus flattened fluctuations, in blood sugar and insulin levels due to their slower digestion and absorption by the body. Generally, low glycemic index foods are defined as having a glycemic index of 55 or less, medium glycemic index foods as having a glycemic index of 56 to 69, and high glycemic index foods as having a glycemic index of 70 or higher.

4.4.1 The glycemic index of the idli made with rice rawa was calculated by the following formula.

$$\text{Glycemic index} = \frac{\text{IAUC of test food}}{\text{IAUC of reference food}} \times 100$$

$$\text{Glycemic index} = \frac{592.5}{1110} \times 100 = 53.3$$

The glycaemic index was calculated as **53.3.**

Glycemic index of all the six days were calculated and are given in table 4.6

TABLE 4.6 GLYCEMIC INDEX OF RICE RAWA IDLI

DAYS	GLYCEMIC INDEX OF RICE RAWA IDLI
1	53.3
2	55.2
3	54
4	57.5
5	58
6	59.9
Mean	56.3
S.D	2.56

The average of six days of GI of rice rawa idli is 56.3 which is near to the glycemic index of idli given in international table 2002 (60)

4.4.2 The glycemic index of the idli made with jowar rawa was calculated by the following formula.

$$\text{Glycemic index} = \frac{\text{IAUC of test food}}{\text{IAUC of reference food}} \times 100$$

$$\text{Glycemic index} = \frac{562.5}{1110} \times 100 = 50.6$$

The glycaemic index was calculated as**50.6.** Glycemic index of all the six days were calculated and are given in table 4.7

TABLE 4.7 GLYCEMIC INDEX OF JOWAR RAWA IDLI

DAYS	GLYCEMIC INDEX OF JOWAR RAWA IDLI
1	50.6
2	52.3
3	50.4
4	51.2
5	50.8
6	51.9
Mean	51.2
S.D	0.75

The average GI of jowar rawa idli is 51.2

4.4.3 Glycemic loads of experimental idlis

The glycemic load (GL) is a ranking system for carbohydrate content in food portions based on their glycemic index (GI) and the portion size. Glycemic load is the product of the GI and the amount of available carbohydrate. The result combines quality and quantity factors of carbohydrate in a food, whereas the GI does not account for quantity. There is a scale to classify glycemic load similar to that of GI as shown in table 4.8

Table 4.8 classification of Glycemic Load

Category	**Glycemic Load**
Low	≤10
Medium	11-19
High	≥20

The glycemic load does present a limitation in that when converting from GI to glycemic load, the foods category may change depending on the serving size. This is positive when addressing healthy foods like watermelon with a GI of 72 (high), but when adjusted for serving size has a GL of 4 (low) [GL= .72 GI X 6g available CHO]. This can also be misleading in a food such as mashed potatoes with a GI of 74 (high) and a GL of 15 (med) [GL= .74 GI X 20g available CHO] (Aziz 2009).

Another concept developed was the glycemic glucose equivalent (GGE). The glycemic glucose equivalent gives the theoretical weight of glucose that would induce the glycemic response equivalent to that induced by a given amount of food. This more refined measurement takes into account the GI, food quantity and food composition (Miller-Jones 2007). Another constructive aspect of the glycemic glucose equivalent is that it is presented as g/serving which is familiar to consumers since nutrition labels are presented in this fashion (Aziz 2009). A limitation of the glycemic glucose equivalent is the fact that a large amount of time consuming testing would be required to generate proper tables which would be expensive. Also, this measure doesn’t account for high levels of other nutrients. A high fat product may result in a positive glycemic glucose equivalent, yet may not be an overall healthy choice (Aziz 2009).

4.4.4 Glycemic load of experimental idli made with rice rawa was calculated by applying the following formula.

$$\text{Glycemic load} = \text{GI}/100 \times \text{dietary carbohydrate content of serving}$$

Glycemic load made with rice rawa= 26.65

4.4.5 Glycemic load of experimental idli made with jowr rawa was calculated by applying the following formula.

$$\text{Glycemic load} = \text{GI}/100 \times \text{dietary carbohydrate content of serving}$$

Glycemic load made with jowr rawa =25.3

There is much debate on the digestibility of sorghum in terms of glycemic response. Powell *et al.* (2002) reported roasted jowar bread, or sorghum bread as having a GI of 77 ± 8 in the 2002 edition of the International Table of Glycemic Index and Glycemic Load. Mani *et al.* (1993) also found sorghum to have an identical GI after testing six commonly consumed sorghum foods of India. In both cases sorghum is classified as having a high GI. Abdelgadir *et al.* (2004) compared the glycemic effect on individuals with diabetes of a sorghum based flatbread and porridge against wheat pancakes, millet porridge, and maize porridge. Results indicated the wheat pancakes and millet porridge had significantly lower glycemic effect than the maize and sorghum foods.

One mechanism suggesting that sorghum is slowly digested due to its tannins. Dyke and Rooney, (2007) reported that tannins bind to protein, carbohydrates, and minerals which decrease the digestibility of these nutrients.

Thompson *et al.* (1983) found a negative correlation between GI and concentration of polyphenols after testing thirteen foods The mechanism of the starch-protein interaction can be explained by protein disulfide bond cross linking involving the kafirin prolamins in the protein matrix around the starch granules which reduces starch digestibility (Taylor and Emmambux, 2010).

4.5 INVITRO STARCH DIGESTIBILTY

Gelatinization or any other treatment that destroy the granular structure of starch increases the digestibility of starch to enzyme action. The in vitro methods of starch hydrolysis provide useful means of assessing the degree of gelatinization of starch in a product, in predicting bio availability of starch in vivo and selecting starchy foods according to the specific needs of different groups of consumers. Several workers have shown significant correlation in starch availability in vitro and in vivo. Hence in vitro method of starch digestibility based on the availability of maltose was used in the present study. The results of IVSD of idli made with rice rawa and jowar rawa are given in table 4.9 and 4.10

Table 4.9: Invitro Starch Digestibility of Rice Rawa Idli.

Days	Invitro starch digestibility of rice rawa idli
1	18.6
2	18.5
3	18.2
4	18.9
5	19.7
6	19.9
Mean	18.9
S.D	0.68

Table 4.10: Invitro Starch Digestibility of Jowar Rawa Idli.

Days	Invitro starch digestibility of jowar rawa idli
1	19.62
2	22.5
3	20.4
4	20.0
5	21.0
6	22.0
Mean	20.92
S.D	1.13

IVSD of whole sorghum flours was ranged from 36.4 to 37.1 mg maltose released/g sample, similar results were reported by Rachid Souliah *et al.* (2012) found IVSD in milled whole sorghum flour as 36.4. There was significant difference ($P<0.05$) among the IVSD contents of sorghum flour (50.1 mg maltose released/g sample), wheat flour (85 mg maltose released/g sample) and soy flour (74 mg maltose released/g sample).

There was significant increase in the IVSD content of sorghum products (59 to 70.5 mg maltose released/g sample) when compared to their flours (36.4 to 52.5 mg maltose released/g sample), In general, cooking of sorghum by any method significantly improved IVSD, similar findings were reported by Phusphamma (1993), when sorghum grains were subjected to different processing conditions like popping, germination etc., IVSD was increased compared to whole grain, which was maximum in popped followed by parched. (Whole grain – 11.2%, parched – 55.7%, popped – 83.2%, malted – 46.8%, parboiled – 25.4%, boiled – 46%) and similar results were found when different traditional processed products of sorghum were evaluated for IVSD, significant increase was observed in IVSD content of processed products compared to whole grain (11.2 mg maltose released /g sample), where there was a 5 fold increase in

IVSD content of roti and biscuits, for idli and dosa 8 fold increase was observed. Rao, (1983) reported that in the peripheral endosperm of sorghum, the starch granules are embedded in a dense mixture of protein bodies (Kafirins) and mixture protein making it difficult for the enzyme to hydrolyze starch. The difference in the susceptibility of starch to digestion could be due to variations in gelatinization during various processing methods.

The digestibility of starch in sorghum grain deserves special attention in the diets of dry land areas where sorghum is a more productive and reliable crop than other cereals and constitutes the principal dietary source of energy.

4.6 CORRELATION OF GLYCEMIC INDEX AND INVITRO STARCH DIGESTIBILTY

The correlation coefficient worked out between the six days of glycemic index values of breakfast item idli made with rice rawa with invitro-starch digestibility values of rice rawa idli. Similarly, the correlation coefficient worked out between the six days of glycemic index values of jowar rawa idli with invitro-starch digestibility values jowar rawa idli.

Correlation value jowar idli is **0.8499** which is found significant at 5 percent level. Similarly, Correlation value of rice rawa idli is **0.899** which is found significant at 5 percent level.

4.7 EFFECT OF IVSD ON GI FOR IDLI MADE WITH RICE RAWA

The regression equation was fitted by taking in-vitro starch as independent variable and GI as dependent variable. The fitted equation was Y= 3.358x - 7.380.

The regression coefficient, b= 3.358, which indicate that for every 1mg increase in-vitro starch digestibility, there will be an increase of 3.358 mmol/l in glycemic index.

4.8 EFFECT OF IVSD ON GI FOR IDLI MADE WITH JOWAR RAWA

The regression equation was fitted by taking in-vitro starch as independent variable and GI as dependent variable. The fitted equation was Y= 0.564x + 39.38.

The regression coefficient, b= 0.564, which indicate that for every 1mg increase in-vitro starch digestibility, there will be an increase of 0.564 mmol/l in glycemic index.

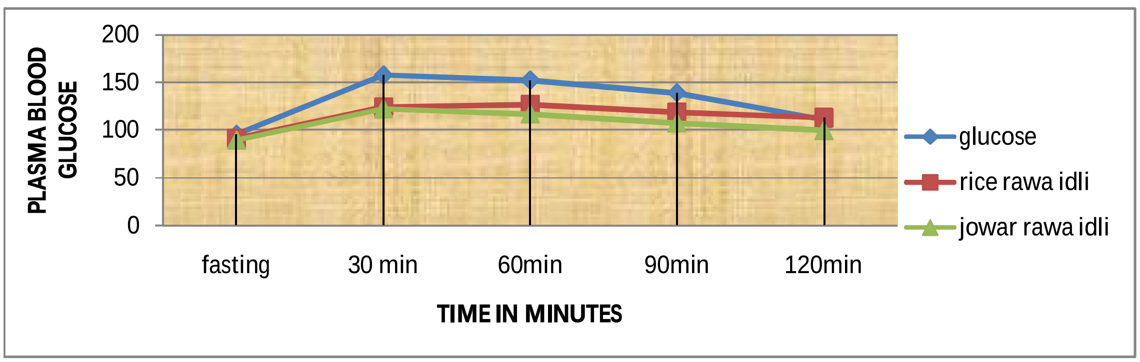

Fig. 4.1 Plasma Glucose Concentration of subjects after consumption of Glucose , Rice Rawa Idli and Jowar Rawa Idli

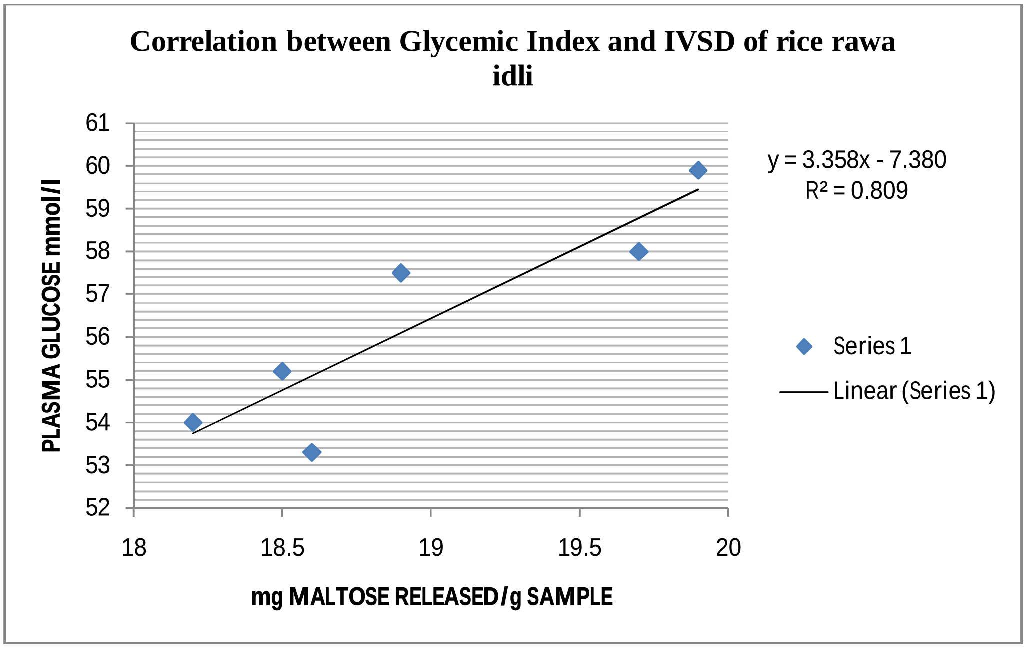

Fig. 4.2 Correlation between Glycemic index and IVSD of Rice rawa idli

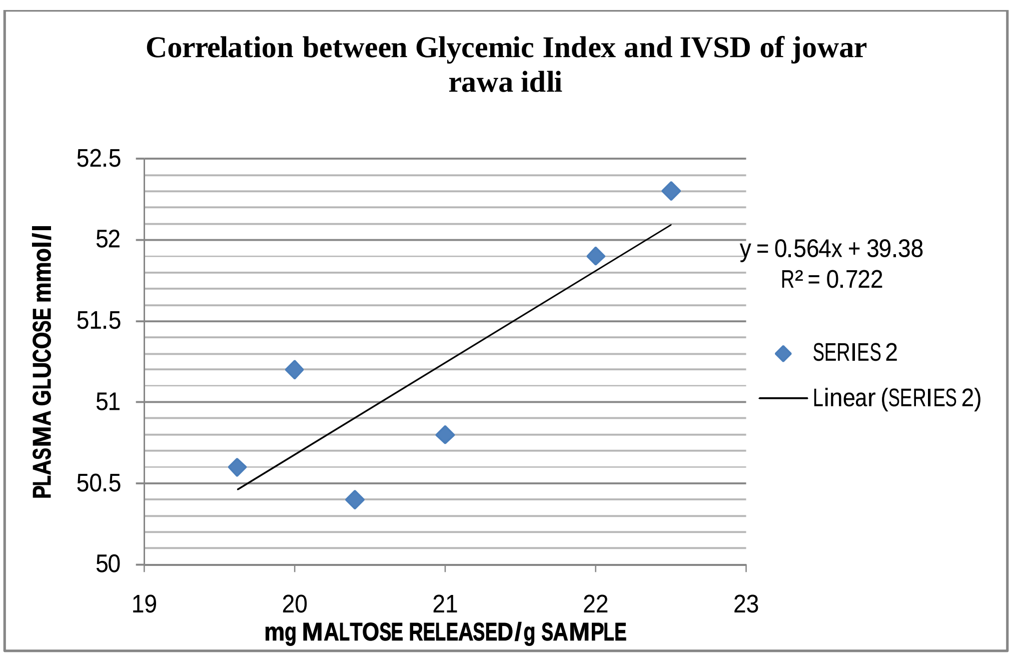

Fig 4.3 Correlation between Glycemic index and IVSD of jowar rawa idli

Chapter V

SUMMARY AND CONCLUSIONS

The global epidemic of diabetes has always been considered as a key issue in public health challenges. Its prevalence rate is rapidly increasing specifically in developed and developing countries such as India. Today India is recognized as diabetic capital of the world with 40 million diabetic people. Therefore, the emphasis is laid on the treatment strategies of the diabetes as the disease remains throughout the life of an individual. Although many changes have been brought about by its discovery, insulin has by no means solved all the problems. Therefore, there is a need to study on dietary treatment and its application on diabetes.

For an effective dietary management of diabetes, it is better to modify the diet appropriate to the individual life style considering the traditional eating patterns and food habits. Hence, idli a common breakfast item consumed in south India was selected for present study. The present study was designed to develop idli with rice rawa and idli with jowar rawa and to determine the glycemic response of these items.

In order to study the effectiveness of the idli made with rice rawa and idli made with jowar rawa on glycemic response, ten healthy subjects were selected. The diets were planned to be isocaloric and contain same amount of carbohydrate (50 gms). Initially an OGTT was conducted for all the subjects to assess their capacity to metabolize glucose and for comparison with G.R to idlis. Idlis made with rice rawa and jowar rawa were given separately to the subjects after overnight fast. An adaptation of six days was maintained for each type of idlis. A gap of three days was given between the different types of idlis.

The fasting and postprandial blood samples at half an hour (30 mins.),one hour (60 mins), one and half hour (90 mins) and two hour (120 mins) after consumption of both type of idlis were collected. Blood glucose levels were measured by using Horizon one touch Glucometer in capillary whole blood obtained by finger prick in the fasted state and at 30, 60, 90 120 mins after the consumption of the idli. The glycemic responses to the idlis (rice rawa and jowar rawa) were determined by comparing the area under curve (AUC) of breakfast items to that of OGTT.

Glycemic index of breakfast product idli was estimated with the help of ten subjects using glucose as reference food. The glycemic index of jowar idli was found to be 51.2, which was considered as low glycemic index food. The 50g of carbohydrate can be taken in one serving i.e. six medium size sorghum idli. The glycemic load of the experimental sorghum idlis was calculated as25.3.Jowar idlis have high glycemic load.

The glycemic index of rice rawa idli was found to be 56.2, which was considered as medium glycemic index food. The 50g of carbohydrate can be taken in one serving i.e.five medium size sorghum idlis. The glycemic load of the experimental sorghum idlis was calculated as 26.65. Rice rawa idlis have high glycemic load.

In vitro starch digestibility was estimated as mg of maltose released per gram sample according to the procedure of Singh and Jambunathan (1982). IVSD content of jowar rawa idli was higher than the rice rawa idli. Among breakfast product idli, the highest IVSD values were observed from jowar rawa idli 20.92 and IVSD value of rice rawa idli were lower 18.9.

To establish correation between glycemic index and invitro starch digestibility Pearson correlation is used. The correlation value of the GI and IVSD of the idli made with rice rawa is 0.899, at 5 percent level, it is significant and the jowar rawa idli is 0.849, at 5 percent level, it is significant. Idlis made with rice rawa and jowar rawa are highly correlated.

The results indicated that consumption of jowar idli lowers plasma glucose than rice rawa idli. The lower glycemic response shown by jowar idli may be due to higher content of dietary fiber and protein, which are known to affect carbohydrate digestion and absorption. In addition processing like fermentation also affects the G.R

Hence from the above study it may be inferred that the millets and legume blends especially high fiber and protein source such as jowar idli a breakfast product should be encouraged for achieving a good glycemic control in diabetes.

The development of this type of products with high yield potentials will be of immense value in alleviating the nutritional problems of the population, which can be formulated in therapeutic diets.

Suggestions for future research:

- To Develop multi grain idliand other breakfast or snack items.
- The effect of addition of hypoglycemic agents like fenugreek,isabgol to these breakfast items need to be investigated.
- The efficacy of jowar based breakfast products can be studied in the management of hypercholesterolemic subjects.
- It is recommended that further study can be carried out to determine the nutritional quality of sorghum products using a small animal assay as this is the standard method of determining nutritional quality in foods.
- Incorporation of vegetables in idli using rice rawa and jowar rawa
- Longitudinal study can also be done for same research.

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