

**EFFECTS OF WHITE YAM PROCESSING METHODS ON GLYCEMIC PROFILE IN
TYPE-2 DIABETIC NIGERIANS AND BIOCHEMICAL VARIABLES IN
ALLOXAN-INDUCED DIABETIC RATS**

BY

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CERTIFICATION

I certify that Dr. ANYAKUDO MICHAEL MAGNUS CHUKWUDIKE carried out this research study in the Physiology department of College of Medicine, Faculty of Basic Medical Sciences, University of Ibadan, Ibadan. Oyo State.

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DEDICATION

This work is dedicated to God Almighty, the author and the finisher of our faith who in His time made all things good and beautiful,

To my wife – Regina OlaideAnyakudo, and children: Michael-Benedict, Cynthia, Dominic,Anthoniaand Esther;

And to all people living with Diabetes Mellitus in Nigeria.

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ABSTRACT

Food processing methods demonstrated using apparently healthy subjects have been reported to affect Glycemic Index (GI) and Glycemic Responses (GR) in different ways with either favourable or adverse effects. However, there are limited data on the effects of food processing methods on GR and GI in diabetics. This study was carried out to determine the effects of boiling, frying, pounding, roasting and flouring (parboiling, sun-drying and grinding) processes on the Proximate of white yam (*Dioscorea rotundata*), GI and GR in male type-2 diabetic Nigerians. The effects of low- and high-GI diets on organ weights, pancreas histoarchitecture, Glycemic Tolerance (GT) and Lipid Profile (LP) in alloxan-induced male diabetic rats (DR) were also investigated.

Proximate of white yam-based meals (raw and processed) and 50g digestible carbohydrates of served portions were determined using Platt and Southgate's standard methods of food analysis while the lipid profile of the experimental rats was analysed using a dry-chemical auto-analyser. The GI was determined in 10 type-2 diabetic and 10 apparently healthy subjects (control) who consumed served portions of each processed meals containing 50g digestible carbohydrates. Postprandial capillary blood glucose concentrations taken half hourly for two hours using glucometer were used to construct the mean incremental GR curve from which the GI was calculated. The product of each processing method was classified into high or low GI diet based on international GI classification of foods. Effects of low-GI (fried yam) and high-GI (roasted yam) diets on organ weights, pancreas histology, GT and LP were respectively determined and compared in the six grouped (n = 8) adult experimental alloxan-induced (150mg/kg BW intraperitoneal) diabetic and control rats fed with test and standard diets for six weeks. Data were analysed using ANOVA and Student's t test at $p \leq 0.05$.

Fiber, protein and calorie contents of the processed meals were significantly increased compared with the unprocessed raw yam, while the ash and moisture contents of the processed

meals were affected by the degree and extent of exposure to heat of processing. Roasting of yam gave the highest GI (93.34±4.04%) frying gave the lowest GI (36.16±2.71%). Postprandial GR to low-GI diet showed significant improved glycemic tolerance over that of High-GI diet (3082.5mg/dl.min vs 8332.0mg/dl.min). An increase in body weight of rats was observed in all groups after six weeks of feeding with highest increase (24.8%) observed in DR on high-GI diet (251.00±1.61g vs 312.33±5.85g) and the lowest increase (9.2%) in DR on Low-GI diet (250.67±1.12g vs 273.17±1.72g). No significant change was observed in organ weights. Pancreas histology of diabetic rats on high-GI diet displayed degeneration with degranulation and vacuolation of the islet β cells while regeneration of some β cells was observed in rats fed with low-GI diet. Triacylglycerides (52.2±1.0mg/dL) and cholesterol (60.5±1.5mg/dL) increased in DR fed with fried yam compared to those fed with roasted yam.

White yam processing methods increased glycemic index with quick glycemic response and decreased glycemic index with delayed glycemic response. Although the fried yam with low glycemic index improved glycemic profile, its recommendation for dietary control of diabetes mellitus may be considered with caution due to its associated dyslipidaemia – a known cardiovascular risk factor.

Keywords: Food processing methods, type-2 diabetes mellitus. Glycemic profile, white yam

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CHAPTER ONE

INTRODUCTION

Processing of foods affects carbohydrate micronutrient content and bioavailability in different ways with either desirable or adverse effects on the nutritional values (Foster- Powel et al, 2002). The bioavailability of starch is affected dramatically through processing regarding both rate and extent of small intestinal digestibility. This permits optimizing the digestion of starch by choice of raw materials and processing conditions. Many metabolic studies have shown that method of food processing vary greatly in their effect on blood glucose and insulin concentrations (Jenkins et al., 2002 and Foster- Powel et al, 2002).

Studies have shown that processing of foods with different methods results in significant differences in the rate and extent of carbohydrate digestion thus affecting the glycemic response (GR) and glycemic index (GI) (Brand Miller et al., 2003). Two different brands of the same type of food such as a plain cookie may look and taste almost the same, but difference in the type of flour used, moisture content and the cooking time can result in differences in the degree of gelatinization and consequently the GI values (Wolever et al, 2002). Boiling, cooking and heating of carbohydrates results in alteration of their physical properties through gelatinization and retrogradation. Altering the physical form of a complex carbohydrate changes the postprandial glucose and insulin response to it (O'Dea et al, 1980).

Many Nigerian foods are prepared under factory processing conditions which is very different from the conventional cooking methods. The food industry has developed a wide range of convenient and novel snack products which are ready-to-eat or minimize preparation in the home with increase storage life. New processes/methods such as extrusion cooking, explosion puffing and instantization which make use of extreme temperature and pressure or repeated

wetting and drying may affect the digestibility of starch as they make starch readily available for digestion. Cooking not only increases the viscosity but also cleaves the starch granules, thereby increasing the starch availability to amylase. Conditions which are known to increase the digestibility of starches are those which produce obvious hydration of the granules (gelatinization), distinct changes in chemical nature or disruption in the organized structures (Boohher et. al, 1951)

Such conditions increase the availability of starch to amylase and are more likely to occur during factory processing because of the higher temperature and pressure involved. In extrusion puffing or cooking, enough energy and heat must be applied to thoroughly gelatinize or cook the ingredients (Kent et. al, 1987). In contrast, conventional cooking methods such as boiling involves less physical disruption in moderate heat, and are therefore less likely to cause starch damage or complete gelatinization. Difference in fiber content, trace minerals concentrations and other nutrient could contribute to difference in the glycemic response to ingested food.

The human diet contains many types of carbohydrates, each of which contributes to different physiologic responses. Diets rich in rapidly digested carbohydrates (High-GI diets) have been suggested to have detrimental effect on health due to high level of postprandial blood glucose and insulin responses associated with them (Salmeron et al, 1997) while slowly digested carbohydrates (Low-GI diets) may protect against chronic diseases (FAO/WHO, 1998).

Dietary carbohydrates have an essential physiological role in the body. The rate of digestion and absorption of carbohydrates can be a determinant factor for the metabolic control of some human chronic non-infectious diseases (Jenkins et al, 2002 and FAO/WHO, 1998). For this reason, there has been a growing interest in the biological utilization of carbohydrates by human body, especially referring to starch and dietary fiber and their effects on the glycemic

response and index and on the large bowel physiology (Jenkins et al, 2002, Lajolo et al, 2001, Danone Vitapole/FAO, 2001 and FAO/WHO, 1998).

The glycemic index (GI), which quantifies the glycemic response produced by foods, evaluates indirectly the in-vivo availability of the carbohydrates (FAO/WHO, 1998 and Wolever et al., 1991). It is an important parameter of food quality that compares the hyperglycemic effect of a tested meal with pure glucose (or of another defined standard food). The GI is a measure of the food power to raise blood-glucose concentration after a meal (Chlup et al, 2004). It is defined as the relation of the incremental area under the blood-glucose (glycemic) response curve (IAUC) of a tested meal containing 50g of digestible carbohydrates and the incremental area under the Blood-glucose (glycemic) response curve of the standard food, i.e. 50g pure glucose (IAUCS).

In principle, the glycemic index is calculated as the measured glycemic response to a portion of a test food containing 50g “available” carbohydrate expressed as a percentage to the same amount of “available” carbohydrate from a standard food eaten by the same subjects (Wolever et al, 1991; 1996 and Jenkins et al, 1981). Glycemic index values have been published for a wide range of foods (Foster -Powel et al, 2002) and have been used in several studies to design low-glycemic load diets for diabetic subjects.

Currently, the importance of glycemic index studies is linked to the possible therapeutical and physiological effects of diets with low GI on healthy, obese, diabetic and hyperlipidemic subjects. The GI has also been related to colon diseases and physical activity (FAO/WHO, 1998; Danone Vitapole/FAO, 2001; Jenkins et al., 2002).

Diabetic patients have been encouraged to increase their carbohydrate intake (FNB/IOM, 2002) but exact details of which foods to use are lacking. In spite of the evidenced positive

physiological effects of low GI foods, there is little information available about the GI of Nigerian foods (Anyakudo and Fasanmade, 2007).

Yams are a primary agricultural commodity cultivated for their starchy tubers consumption in West Africa and New Guinea. Cultivated first in Africa and Asia about 8000BC (Kay, 1987), several species have been identified. Yams of African species are prepared and processed in various ways including boiling, pounding into paste, roasting, frying, leaching, drying and grounding into flour for consumption as foods. Yam is the common name for members of the genus *Dioscorea* (family Dioscoreaceae). More than 600 species of yam have been identified. The word yam comes from Portuguese *inhame* or Spanish *ñame*, which both ultimately derive from the Wolof word *nyam*, meaning “to sample or taste.” Yam tubers can grow up to seven feet (approx. two meters) in length and weigh up to 150 pounds (68kg). (CGIAR, 2006)

Scientifically, the taxonomy of yam has been defined under the following classification: Kingdom – Plantae; Division – Magnoliophyta; Class – Liliopsida; Order – Dioscoreales; Family – Dioscoreaceae; Genus – *Dioscorea* (D). Among several species identified, the major cultivated yam species include: *D. rotundata*, *D. cayenensis*, *D. alata*, *D. opposita*, *D. bulbifera*, *D. esculenta*, *D. trifida* and *D. dumetorum*.

D. rotundata, the white yam, and *D. cayenensis* the yellow yam, are native to Africa. They are the most important cultivated yams. In the past, they were considered two species but most taxonomists now regard them as the same species. There are over 200 cultivated species between them. They are large plants; the vine can be as long as 10-12 meters (35-40 feet). The tubers most often weigh about 2.5 to 5 kg (6-12 lbs) each but can weigh as much as 25kg (60

lbs). After 7-12 months growth the tubers are harvested. In Africa most are pounded into a paste to make the traditional dish “fufu or Iyan” (Kay, 1987).

The nutritional quality of the carbohydrates and the effects of processing on that quality is a major concern when considering foods to be incorporated into diabetic diets because processing in a number of ways can alter both the content and the nutritional quality of food carbohydrates (Jenkins et al, 2003).

Clinical studies of persons with diabetes have shown a good positive correlation between improved glycemic control and diets with low glycemic index (Brand et al, 1991; Wolever et al, 1992; Fontvielle et al, 1992 and Frost et al, 1994). Dietary advice based on the glycemic index improves dietary profile and metabolic control in type 2 diabetes mellitus (Frost et al, 1994). Thus, food processing favouring low glycemic index should be advocated. Wolever et al (1992) studied the beneficial effects of a low-glycemic index diets in type 2 diabetes mellitus. Studies on the control of diabetic complications have shown a decrease in progression of diabetic complications as a result of euglycemic control from dietary therapy (Hodges et al, 2004).

The use of animals as diabetic model has provided useful information and knowledge on the pathogenesis and management of diabetes mellitus. It has also made provision for researchers to perform various manipulative interventions in finding solutions to various problems associated with metabolic disorders. The second phase of this study which involved the use of animal (rat) model allowed the comparative study of the effects of Low-GI and High-GI diets on certain body parameters and tissue histology thus providing rational basis and caution for dietary selection, advice and recommendations.

Nigeria as an African country with large population and various ethnic groups has various eating habits and foods. With rapid development of economy, lifestyle of Nigerians is

changing also and a cluster of chronic diseases is emerging such as diabetes mellitus. Some of these emerging chronic diseases have become health threat to the people as seen in the western countries. Therefore, links between dietary habits and chronic diseases should be investigated and strategies should be developed for prevention as well as treatment of such diseases.

Monotonous consumptions of certain food such as unripe plantain, beans and beans-based products with associated fear of eating other food types either due to lack of adequate dietary advice or fear of unknown among Nigerian diabetics leads to poor glycemic compliance/response with subsequent poor glycemic control. It is therefore necessarily important to determine the GI and the effect of processing on the glycemic response and glycemic index values of other food types suitable to be incorporated into dietary menu for diabetics and also to compare the effects of low-GI and high-GI diets on glucose tolerance, body and organ weights, pancreas histology and lipid profile thus providing rational basis and caution for dietary selection, advice and recommendations.

It is urgent to determine the GI values of Nigerian foods and factors affecting it in an effort to establish the database of GI of Nigerian foods bearing in mind the effect of processing on the glycemic response and the glycemic index values among other factors. Moreover, the effects of low and high glycemic index foods on total body and organ weights, lipid profile and pancreas should be examined to provide rational basis for dietary selection, advice and recommendations.

Few studies however, have reported the effect of processing on the glycemic response and glycemic index of various foods in our environment in both healthy subjects and type 2 diabetics. Also there is paucity of studies comparing the effect of low- and high-GI diets on certain body parameters such as lipid and glycemic profile in healthy and diabetic states using

animal models in our environment. Recently, diabetic patients have been encouraged to increase their carbohydrate intake but exact details of which foods to use are lacking. For many foods in Nigeria, the glycemic indices and the effect of processing on the glycemic responses and indices are yet to be determined. Hence this study is designed to provide answers to these important questions:

- i. What conventional or local processing method(s) will favour low glycemic index value of various prepared diets in our environment?
- ii. Is there any indication for dietary selection and caution for diabetic persons compared to non-diabetic healthy subjects when recommending dietary pattern?
- iii. Which food type based on GI classification should be considered when recommending dietary menu to diabetics – Low-GI or High-GI menu?
- iv. What is the rational basis for selection and recommendation of Low-GI diets to diabetics?

Therefore, any effort to enable the determination and practical use of GI (bearing in mind the effect of food processing/preparation on the glycemic response and index values) and comparison of Low- and High-GI diets may support establishing optimum dietary recommendations and good eating habits in diabetic and healthy subjects in our environment.

The rationale of this study therefore is to identify processing method(s) that favour prepared diets with Low-GI values which are recommended in dietary menu by WHO through epidemiological and dietary intervention studies to promote good glycemic response and overall euglycemic control in diabetics (Neuhouser et al, 2006 and Brand et al., 1990) and also to provide rational basis and caution for dietary selection, advice and recommendations.

AIM

This study carried out in two phases using both human subjects (Phase I) and animal models (Phase II) was designed to determine the effect of processing/preparation of food on the glycemic responses and the glycemic index values of prepared food using a white yam (Dioscorea rotundata) species and to compare the effects of Low-GI and High-GI diets on body and organ weights, pancreas histoarchitecture, glycemic tolerance and lipid profile.

OBJECTIVES

The objectives of this study include:

1. To determine the nutrients composition (proximate) of white yam before and after processing.
2. To determine the effect of boiling, sun-drying/flouring, pounding, frying and roasting processing methods on the GR to ingested meals in diabetic and healthy subjects.
3. To determine the glycemic indices (GI) of test meals (Amala - yam flour paste, Boiled, Pounded, Fried and Roasted yams) and to compare the GR and GI between Diabetic and Non- diabetic healthy subjects.
4. To assess the variability of the GI of the test meals and correlate it with the GR in diabetic and non-diabetic healthy subjects.
5. To determine and compare the effects of low-GI and high-GI diets on glycemic tolerance, body and organ weights, pancreas histology and lipid profile in alloxan-induced diabetic and non-diabetic healthy adult rats.

CHAPTER TWO

LITERATURE REVIEW

2.1 YAM

2.1.1 INTRODUCTION

Yam is the common name for members of the genus *Dioscorea* (family *Dioscoreaceae*). More than 600 species of yam have been identified and out of the estimated species available, over half dozen principal species are grown for the consumption of their starchy tubers, while others are grown for medicinal purposes. Among the cultivated species, there are hundreds of cultivars. The word yam comes from Portuguese 'inhame' or Spanish 'ñame'; both ultimately derived from Wolof word nyam, meaning "to sample or taste" (Kay, 1987).

Yams are a primary agricultural commodity in West Africa and New Guinea and are originated in the Far East and spread westwards. They were first cultivated in Africa and Asia about 8000B.C. To this day, yams are important for survival in these regions. Independently evolved in the eastern and western hemispheres, today, yams are grown widely throughout the tropics. In the West African yam zone, which is the principal producer on a global basis, *Dioscorea rotundata*, *D. alata* and *D. esculenta* are the most common species. However, consumer's preferences might account for some of the predominance of certain cultivars in some regions in addition to agro-climatological impacts on the growing attributes (Opara, 2001).

Yam tubers can grow up to seven feet (approx. two meters) in length and weigh up to 150 pounds (68kg) CGIAR, 2006). The yam has a rough skin which is difficult to peel, but softens after heating. Yam skins vary in colour from dark brown to light pink. The majority of the yam is composed of much softer substance known as the "meat". This substance ranges in colour from white to bright orange in ripe yams. Yam tubers can be stored for four to six months without

refrigeration, which makes them a valuable resource for the yearly period of food scarcity at the beginning of the wet season.

Most of the world production of yams comes from Africa (about 96%) with Nigeria alone accounting for nearly 75% of the total world production (FAO 1991). World annual production of yam has increased with time but however, the importance of yam in the economy of the main producing areas appears to be declining partly due to competition with other crops like Cassava in Nigeria and Taro in south Pacific (Dorosh, 1988).

2.1.2 TAXONOMY

Scientific classification (taxonomy) of yam according to O’Hair (1954) include: Kingdom – Plantae; Division – Magnoliophyta; Class – Liliopsida; Order – Dioscoreales; Family – Dioscoreaceae; Genus – Dioscorea (D) and Species. Among several species identified, the major cultivated yam species include: *D. rotundata* (white yam), *D. cayenensis* (yellow yam), *D. alata* (water yam), *D. opposita* (Chinese yam), *D. bulbifera* (potato yam), *D. esculenta* (lesser yam), *D. trifida* (cush cush yam) and *D. dumetorum* (bitter yam).

2.1.2.1 Dioscorea rotundata (White Yam)

D. rotundata, the white yam, and *D. cayenensis* the yellow yam, are native to Africa. They are the most important cultivated yams. In the past, they were considered two species but most taxonomists now regard them as the same species. There are over 200 cultivated species between them. They are large plants; the vine can be as long as 10-12 meters (35-40 feet). The tubers most often weigh about 2.5 to 5 kg (6-12 lbs) each but can weigh as much as 25kg (60 lbs). After 7-12 months growth the tubers are harvested. In Africa most are pounded into a paste to make the traditional dish “fufu or Iyan” (Kay, 1987).

2.1.3 ECONOMIC AND SOCIAL IMPACT

Yam is a tropical root crop. Second to cassava, yam is a staple crop in many parts of Africa and south East Asia. In south pacific, yam is a significant food crop accounting for more than 20%, 8.1% and 4.6% of the total dietary calorie intake in the kingdom of Tonga, Solomon Islands and the Papua Guinea respectively (Dorosh, 1988).

Yam plays significant role in the socio-cultural lives of some yam producing regions like the celebrated New Yam Festival in West Africa, a practice that has extended overseas where there is a significant population of the tribes that observe it. In some part of South Eastern Nigeria, the meals offered to gods consist principally of mashed yam.

Yam store relatively longer in comparison with other tropical fresh produce and therefore stored yam represent wealth which can be sold all year round by the farmer or the marketer. In part of Igbo land in South Eastern Nigeria, it is customary for the parents of a bride to offer her yams for planting as a resource to assist both couple in raising a family (Dorosh, 1988).

2.1.4 PHYSICO-CHEMICAL PROPERTIES

Few studies have been conducted on the physicochemical variation of yam tubers. Using *D. alata* tubers, Martin (1974) observed in Puerto Rico that high dry weights are associated with fine structure, dense feel, high quality, and concluded that high density is a varietal character that is not changed much by environmental influences. In the Pacific, although some preliminary work has been done at the inter-specific level (Bradbury and Holloway 1988), the lack of information on the variation within *D. alata* hinders its prospective utilisation as a high quality exportable vegetable. Egesi et al. (2003) studied the extent of genetic diversity existing for organoleptic properties in 40 water yam varieties cultivated in Nigeria. Two thirds of their accessions were identified as being suitable for boiled yam, while more than half of these

accessions were good for pounded yam. Their results were, however, based on the respective quality attributes evaluated but the physicochemical characteristics of the tubers were not quantified. Furthermore, *D. alata* is thought to have been introduced clonally in Africa and its genetic base is narrow as demonstrated by the limited isozyme variation detected between African cultivars (Lebot et al. 1998).

Indigenous knowledge claims that there is tremendous variation between the culinary and palatability properties of yam varieties, some being suitable for certain types of preparation, while others are not, and some being cooked much faster than others (Bourrieau 2000). When the tuber of some varieties is cut open, the colour of the surface begins to change rapidly with the oxidation of polyphenolic compounds and become yellowish or brown. Polyphenolic oxidation is also associated with off-flavours and bitterness. In some cases, it deserves special preparations. In most islands of Vanuatu, only certain varieties are suitable for the preparation of the national dish, *laplap*, a pudding made from freshly and finely grounded tubers steam cooked in *Heliconia indica* leaves. Others have to be boiled or roasted in order to be palatable (Bourrieau 2000).

Previous work conducted on the physico-chemical characteristics variation of yam tubers focused mostly on differences existing between species measured on samples collected in different growing environment (Bradburry and Holloway 1988; Treche 1998). Lebot et al. (1998) analysed 131 cultivars of *D. alata* from New Caledonia for dry matter, starch, proteins and minerals but amylase and total sugars were not measured and oxidation and eating quality ratings were not available. Overall, less variation (expressed in CV%) was found in New Caledonia (i.e., 11.8 vs. 17.2% for dry matter, 5.6 vs. 9.1 for starch, 11.4 vs. 15.2 for minerals and 16.8 vs. 17.8 for proteins) which is consistent with the greater genetic diversity found in Vanuatu (Malapa et al. 2005). Egesi et al. (2003) evaluated 40 accessions for the suitability of their tubers for the preparation of boiled and pounded yams. Ratings were based on hedonic scales and the mean

scores for general preference were regressed on individual attribute scores. Mealiness, colour and taste were found to be important for boiled yam, while consistency, colour and stickiness determined the general preference for pounded yam.

2.1.5 PREPARATION/PROCESSING

Yams of African species are prepared and processed in various ways including boiling, pounding into paste, roasting or baking, frying, leaching, drying and grinding into flour for consumption as foods. Yams of African species must be cooked or processed to be safely eaten because various natural substances in raw yams can cause illness if consumed; the most common cooking method in Western and Central Africa is pounding of yam into a paste called “fufu”. Preparing some species of yam is a time-consuming process, involving days of pounding, leaching and boiling to remove the toxins. Yams may be served fried, boiled or pounded into paste. In the Western Nigeria, it may be sun-dried and grounded into yam flour to be consumed as yam flour meal called “Amala”. In the Eastern Nigeria, it may be pounded with ‘Eba’ to make ‘Utaraji’ (Opara, 2001).

Boiled and baked (roasted) yam can be eating with vegetable sauce or palm oil. It could also be pounded or mashed in the mortar with pestle and eaten as ‘fufu’, ‘iyan’ or ‘utara’. Yam cultivars which contain toxic substances such as dioscorene are first sliced and soaked in salt water for several hours before further processing for consumption as food (Opara 2001).

Yams are mainly grown for direct human consumption and marketed as fresh produce in all the growing areas. Industrial processing and utilization of yam includes starch, poultry and livestock feeds and production of yam flour.

One of the major disadvantages of processing yam for food is that nutrient losses in these products can be high particularly minerals and vitamins. In products obtained from secondary

processing as biscuits and fufu, the amount of loss depends principally on the amount of edible surface area exposed during processing operations. Primary unit operations such as milling affect the thiamine and the riboflavin contents of the *D. rotundata* with average losses of 22% and 27% respectively (Opara, 2001).

Sun drying results in high losses of vitamins B with little change in mineral contents. Pounding yam in a traditional wooden mortar or grinding in an electric mixer had similar effects (Bencini, 1991).

Nearly every food preparation process reduces the amount of nutrients in food. In particular, processes that expose foods to high level of heat, light and or oxygen cause the greatest nutrients loss. Nutrients can also be washed out of food by the fluids that are introduced during a cooking process. Boiling of potato and yam can cause much of the vitamins B and C to migrate into the boiling water. These vitamins can be salvaged if the water and yam/potato are been turned to yam/potato soup. Similar losses occur during broiling, roasting or frying in oil when the drippings are drained off (USDA, 2003).

2.2 CARBOHYDRATES

2.2.1 Sources

Carbohydrates are one of the three main nutrients in the foods, the others being fats and proteins. Several sources of carbohydrates have long been established. Carbohydrates come from a wide array of foods including yam, bread, beans, milk, popcorn, potatoes, cookies, spaghetti, corn, yam, cassava cherry pie, wheat and so on. The best sources of carbohydrates especially fruits, vegetables and whole grains deliver essential vitamins and minerals, fiber and a host of important phytonutrients (Franz, 2001). Carbohydrate is the most important source of energy in

food, representing between 40% -70% of total calorie supply in developed (at lower limit of normal) and developing (at upper limit of normal) countries (Wursch, 1990).

In a variety of forms, most common and abundant carbohydrates are sugars, fibers and starches. Dietary carbohydrates are in most part polymers of hexoses such as galactose, fructose and glucose. Very few foods contain significant amounts of free glucose. Significant quantities of glucose are present in the form of disaccharides (especially sucrose and lactose) and polysaccharides (mainly starch in plant foods and some glycogen in animal foods).

The basic building block of carbohydrates is a sugar molecule with a chemical formula of $C_6H_{12}O_6$ consisting of union of carbon, hydrogen and oxygen atoms held together by chemical bonds. Starches and fiber are essential polymers of sugar molecules existing in either straight or branched chains form. Carbohydrates provide energy to body cells, particularly the brain, which is the only carbohydrate-dependent organ in the body. The cells absorb glucose and convert it into energy for cellular activities.



Figure 1: Basic structure of building block of carbohydrates.

2.2.2 Types (Classification) of Carbohydrates

Many foods generally contain a mixture of different types of carbohydrates. Based on certain factors, carbohydrates have been classified in various ways. According to molecular and biological structures, carbohydrates are classified into two main types namely simple carbohydrates (monosaccharides and disaccharides) and complex carbohydrates (oligosaccharides and polysaccharides).

Jenkins et al (2003), classified dietary carbohydrates into high, medium or low glycemic index foods based on how quickly they raise the blood sugar after a meal. Jenkins and his pioneers have long established this concept of glycaemic index since 1981.

In the past, carbohydrates were sometimes classified into sugars (simple carbohydrates) and dietary fibers and starches (complex carbohydrates). Recently, based on the degree of processing by the food manufacturers, carbohydrates can also be classified into refined and unrefined carbohydrates.

2.2.2.1 Simple Carbohydrates and Composition

Simple carbohydrates, otherwise known as simple sugars, are made up of one or two units of sugar (“saccharide”). Carbohydrates containing only one unit of sugar are called monosaccharides while those with two sugar units, disaccharides. Simple sugars, which are typically sweet tasting, are rapidly absorbed into the blood stream, metabolized and converted by the body into energy. Common examples of monosaccharide simple sugars are glucose (also called dextrose) and fructose while a less common example is galactose. Simple sugars (except fructose) are typically high on the glycemic index, so they tend to cause a rapid rise in blood sugar.

Glucose is the primary form of sugar stored in the human body for energy. Some, while present in the blood (as blood glucose), others are stored as glycogen in the skeletal muscles and liver. There are three sources of blood glucose. The first two major sources include the gastrointestinal tract and the liver while the third source: the kidney, does not become significant unless in cases of prolonged starvation (Owen et al, 1969).

The most common dietary sources of glucose in our environment are the table sugar (sucrose) and starch (which is the major storage form of carbohydrate in plant). Fructose is the main sugar found in most fruits, honey and high-fructose corn syrup. Before it can be utilized as

a source of energy in the body, it must first be converted to glucose (although structurally, both fructose and glucose have the same chemical formula). Galactose is obtained from lactose in milk. Like fructose, it is also converted into glucose before being utilized as energy source.

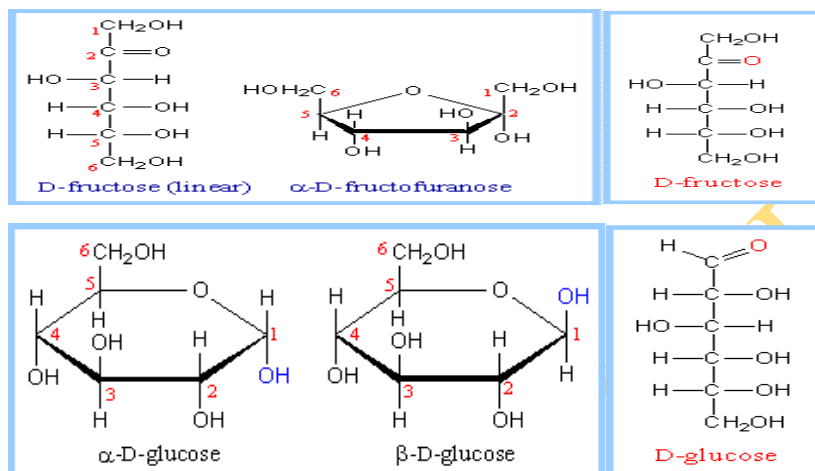


Figure 2: Chemical structures of monosaccharides

(From Robert K.M et al: Harper's Biochemistry 1990.Courtesy of the Appleton and Lange Co.)

Disaccharides, meaning “two sugars”, are commonly found in nature as sucrose, lactose and maltose. They are typically high on the glycemic index. Sucrose (formed as a result of photosynthesis in plants) is found in table sugar and is made up of glucose and fructose.

Lactose, composed of glucose and galactose, comes from milk. Due to its complex molecular structure, some individual are lactose intolerant. Like galactose, Maltose is not commonly found in nature.

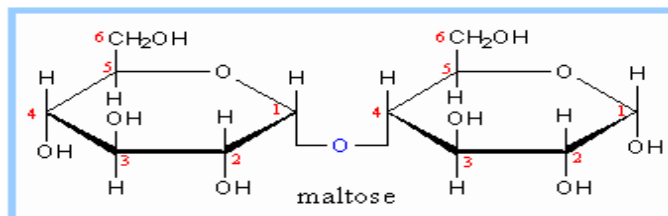


Figure 3: Chemical structure of disaccharides

(From Robert K.M et al: Harper's Biochemistry 1990.Courtesy of the Appleton and Lange Co.)

2.2.2.2 Complex Carbohydrates and Composition

Complex carbohydrates are composed of three or more units of sugar. Their complicated structure made them to be called “complex” carbohydrates. The chemical name for the largest type of complex carbohydrate is “polysaccharide”, meaning “many sugars.” Complex carbohydrates containing three to six units of simple sugar are called Oligosaccharides.

The term Oligosaccharides is derived from “oligo” – meaning, a few- and “saccharide” – meaning, sugar. Most of the few naturally occurring oligosaccharides are found in plants. Important oligosaccharide carbohydrates are raffinose and stachyose. These oligosaccharides are found in beans and legumes. They cannot be digested properly by small intestine but ended up being metabolized and expelled from the large intestine. Typically, they are low glycemic index carbohydrates, which help to maintain stable blood glucose levels when ingested as part of a meal (Jenkins, 2003).

Complex carbohydrates with more than six units of sugar are referred to as polysaccharides. They are the most naturally occurring complex carbohydrates. Starch is the main polysaccharide used by plants to store glucose and is the most common form of edible polysaccharide. Some polysaccharides even have more complex molecular structure and are known as dietary fiber, or “non-starch polysaccharides”. They include cellulose, hemicelluloses, pectin, gum and mucilage.

Cellulose cannot be digested by the human digestive system, so it passes through the digestive tract unabsorbed. Despite this fact, it remains essential for a healthy diet because it helps to “exercise” and maintain the integrity of the intestines. In lower animals like ruminants (e.g. cow), cellulose is digested and metabolized in their intestine by the intestinal flora (bacteria). Cellulose and most types of hemicelluloses are known as insoluble fibers, while

pectin, gum and mucilage are all soluble fibers that easily dissolve or swell when mixed with water.

2.2.3 Starch: Starch is one of the carbohydrates most widely produced by plants. A limited number of starchy plants are extensively grown and used for human consumption namely cereals, tubers and pulses. They also have the highest yield in starch (Wursch, 1990). Starch is deposited in the plants in the form of tiny granules, which are insoluble in water and ranging from 1-100 μ m in diameter depending on the plant species. These granules consist of amylose and amylopectin proportions which vary from one starch to another. Most starch consist of 15-25% amylose which is an essential linear polymer composed almost entirely of α -1,4-linked D-glucose and a limited number of long chain branching involving α -1,6-linkages (Bank et al, 1996). Its molecular weight varies depending on the plant source and maturity. It may contain 200-10000 anhydrous-glucose units.

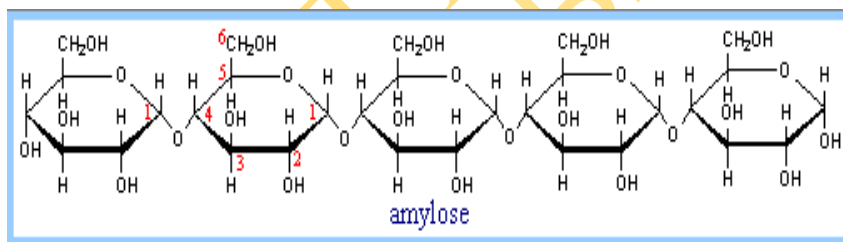


Figure 4a: Chemical Structure of amylose constituent of starch

(From Robert K.M et al: Harper's Biochemistry 1990.Courtesy of the Appleton and Lange Co.)

Amylopectin is one of the largest molecules in nature with a molecular weight between 10-500 millions unit (Greenwood et al, 1972). It is highly branched in α -1, 6-branch points. Debranching with enzymes reveals a bimodal distribution of linear chains. The longest chain has 50-60 glucose unit and short chains have 17-19 glucose units.

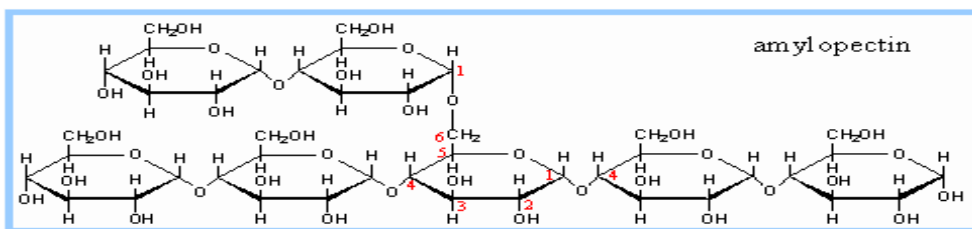


Figure 4b: Chemical Structure of amylopectin constituent of starch

(From Robert K.M et al: Harper's Biochemistry 1990. Courtesy of the Appleton and Lange Co.)

The morphology of starch is not unique but varies from each botanical type and even within species. Starch as a carbohydrate, represents the source of nutritive energy for humans (Bornet et al, 1990). With excessive treatment, the granules may even rupture and disintegrate with solubilization of the starch fraction. In many common plant foods the starch is only partly gelatinized because of the limited water content during processing. The starch granules are only slightly swollen while the internal structure is partly intact (Holm et al, 1988).

Physicochemically, starch is a white, odorless and tasteless carbohydrate, typically composed of long chains of glucose molecules (1000 or more). The granules are insoluble in water and heating them in excess water at a temperature above 50 degree centigrade results in alteration of the granules due to swelling, hydration and solubilization. Disruption of the starch molecules results in gelatinization, which involves melting of the crystallites present in the native granules. Retrogradation of the starch may occur upon cooling of the gelatinized starch, which is enhanced by low temperature and high starch concentration.

The state of starch may play an important role in determining susceptibility to the digestive enzymes (Evans et al, 1986). Processed food may therefore contain an appreciable quantity of starch, which is quite resistant to α -amylolysis and thus escape digestion in the intestinal tract (Englyst et al, 1985). This resistant starch is generally defined as the component of native or processed starch that is able to survive exhaustive digestion of amylolytic enzymes.

Nevertheless, it can be measured as α -glucan after solubilization with alkaline dimethyl sulfoxide (Berry, 1986). The formation of this resistant starch is influenced by amylose content, processing temperature and water content.

In plant cells, starch is presented as granules of starch polymers (amylose and amylopectin) tightly packed with a high degree of molecular order associated by hydrogen bonding. Raw granules contain highly crystalline regions, which are birefringent in polarized light. The granules are insoluble in cold water but when exposed to heat in the presence of water, they undergo irreversible swelling and destruction of the internal crystalline structure and the birefringence is lost. This process is referred to as gelatinization (Holmes et al, 1985).

2.3 DIGESTIONS AND ABSORPTION OF CARBOHYDRATES

2.3.1 Digestions

The utilization of starch from foods by human beings involves several well-recognized steps such as cooking, ingestion with chewing, digestion in the alimentary canal as well as absorption and metabolism of the monosaccharides unit consisting exclusively of glucose (Wursch, 1990).

In addition, there is increasing evidence that part of the ingested starch can escape digestion and absorption but then, utilized through colonic fermentation. The digestion rate of starch measured in-vitro correlated with glycemic response (Jenkins et al, 1982). The glycemic responses (in human and rats) of the processed cereals are generally in good agreement with the in-vivo rate of digestion, which is assumed to be the rate-limiting step in the in-vitro digestion process (Brand et al, 1985; Rose et al, 1987 and Krezowski et al, 1987). However, cereals fiber does not appear to affect the rate of hydrolysis except when it forms a physical barrier limiting access of the starch.

The principal dietary carbohydrates are polysaccharides, disaccharides and monosaccharides. Starches (glucose polymers) and their derivatives are the only polysaccharides that are digested to any degree in the human gastrointestinal tract. In glycogen, the glucose is mostly in long chains (glucose molecule with 1,4 α -linkage) while few branching produced by 1,6 α -linkages are also present. Amylopectin, which constitute 80-90% of dietary starch, is similar but less branched whereas amylose is a straight chain with only 1,4 α -linkages. Glycogen is found in animals whereas amylose and amylopectin are of plant origin. The disaccharide sucrose (table sugar) is only ingested along with the monosaccharides fructose and glucose. Starch is attacked by ptyalin, the α -amylase enzyme in the saliva. However, the optimal P^H for the enzyme is 6.7, and its action is inhibited by the acid gastric juice when food enters the stomach. The P^H sensitivity plays an important role in the functioning of salivary amylase.

In the small intestine, the potent pancreatic α -amylase also acts on the ingested polysaccharides. Both the salivary and the pancreatic α -amylases hydrolyze 1,4 α linkages but spare 1,6 α linkages, terminal 1,4 α linkages, and the 1,4 α linkages next to branching points. Consequently, the end products of α -amylase digestion are the Oligosaccharides-the disaccharide maltose, the trisaccharide maltotriose, some slightly larger polymers with glucose in 1,4 α linkage, and α -limit dextrin, branched polymers containing an average of about 8 glucose molecules. (Wheelan et al, 1960).

The Oligosaccharidases responsible for further digestion of the starch derivatives are located in the outer portion of the membrane of the microvilli principally in the ileum (Semenza, 1981). α -Limit dextrinase hydrolyzes the α -limit dextrans, and maltase split glucose from maltose, maltotriose, and other polymers of glucose in 1,4 α linkage (Semenza, 1986 and Marshal et al, 1977).

Most of the glucose molecules that are formed enter the mucosa cells although some reenter the intestinal lumen and are absorbed farther along. Ingested disaccharides are hydrolyzed by lactase or sucrase on the luminal surface of the mucosa cells (Gray et al, 1979). Deficiency of one or more of these disaccharidases leads to diarrhea, bloating and flatulence after ingestion of sugar. The diarrhea is due to the increased number of osmotically active oligosaccharide molecules that remain in the intestinal lumen, causing the volume of the intestinal contents to increase. The bloating and flatulence are due to the production of gas (carbondioxide and hydrogen) from disaccharide residues in the lower intestine and colon.

Lactase is of interest because in most mammals and in many races of humans, intestinal lactase activity is high at birth, declines to low level during childhood and remains low in adulthood. However, most western Europeans and their American descendants retain their intestinal lactase activity in adulthood. Lactose intolerance is found in a few African tribes but most blacks are intolerant. In the USA, 70% of the black populations are intolerant to lactose. The problem of milk intolerance can be relieved by administration of commercial lactase preparations but this is expensive. Yogurt is better tolerated in intolerant individuals because it contains its own bacterial lactase.

2.3.2 Absorption

Hexoses and pentoses are rapidly absorbed across the wall of the small intestine. Essentially, all of the hexoses are removed before the remains of a meal reach the terminal part of the ileum. The sugar molecules pass from the mucosa cells to the blood in the capillary draining into the portal vein. Glucose (not being a polymer) does not go through digestion process as do the starchy foods in focus and because of its high tonicity, may affect the rate of gastric emptying (Thompson et al, 1982).

The transport of some sugars is uniquely affected by the amount of Na⁺ in the intestinal lumen. A high concentration of Na⁺ on the mucosal surface of the cells facilitates while a low concentration inhibits sugar influx into the epithelial cells. This is because glucose and Na⁺ share the same symport. Intracellular Na⁺ is low and it moves into the cell along its concentration gradient. Glucose moves with it and is released in the cell. The Na⁺ is transported into the lateral intercellular spaces and the glucose diffuses into the interstitium and thence to the capillaries. Thus, glucose is an example of secondary active transport. The energy for the glucose transport is provided indirectly by the active transport of Na⁺ out of the cell. This maintains the concentration gradient across the luminal border of the cell so that more Na⁺ and consequently more glucose enter. The glucose mechanism also transports galactose.

Fructose apparently utilizes a different carrier and its absorption is independent of Na⁺ or the transport of glucose and galactose. Some fructose is converted to glucose in the mucosa cells. Pentoses are absorbed by simple diffusion. Insulin has little effect on intestinal transport of sugars. In this respect, intestinal absorption resembles glucose reabsorption in the proximal convoluted tubules of the kidneys; neither process requires phosphorylation and both are essentially normal in diabetes. The maximal rate of glucose absorption from the intestine is about 120g/h.

The amount of unabsorbed carbohydrate available for colonic fermentation can be estimated by following the increase of breath hydrogen after eating a test meal. This can be quantified by comparing the amount of hydrogen excreted after a standard dose of lactulose (Brand et al, 1972 and Wolever et al, 1986). The amount of starch reaching the colon was found to correlate well with the amount calculated from the production of hydrogen (Levitt et al, 1972). However, this would overestimate the level of malabsorbed starch presumably due to the dietary

fiber present in the foods tested and which by fermentation also produces hydrogen (Jenkins et al, 1978).

The tolerance test monitors the rise in plasma levels of glucose and insulin following oral ingestion of carbohydrate meals. The rapidity, duration and degree of rise in plasma glucose and insulin represent the end result of gastric emptying, digestion and absorption and also of glucose utilization. Recent investigations in man with the use of breath hydrogen test indicated the existence of malabsorption of most starch foods by the small bowel, which may only modify energy food value but induce abdominal discomfort as well (Anderson et al, 1981 and Levitt et al, 1987).

Digestibility of starch in foods by α -amylase can be modified in-vitro as well by numerous factors including starch origin (Goddard et al, 1984 and Bonnet et al, 1989), non-starch components such as fiber (Harber et al, 1977 and Potter et al, 1981) or glutinous proteins (Anderson et al, 1981 and Jenkins et al, 1987) and food processing (Bjorck et al, 1984 and Brand et al, 1985).

2.4 CARBOHYDRATES AND GLYCEMIC (BLOOD GLUCOSE) RESPONSES: EFFECTS OF PROCESSING ON GASTRIC EMPTYING AND RATE OF GLUCOSE ABSORPTION.

The glycemic response to carbohydrates varies from one form to another (Jenkins et al, 1983). The role of food on gastric emptying rate if starchy meals are consumed, and the correlation with blood glucose response has been studied by Torsdottir et al (1984) on healthy and diabetic subjects. A high correlation was found between the difference in gastric emptying and difference in blood glucose and insulin responses (Rice meal versus mashed Potato meal-the time was greater for rice). The meals were structurally different and apparently affect the gastric emptying correlation and hence the absorption of glucose. The high correlation observed may

imply that the digestion of starchy foods is not the sole limiting factor for glucose absorption (rice and potato).

However, no correlation was found between gastric emptying and blood glucose or plasma insulin after a whole white bean meal. As expected, blood glucose was blunted but the gastric emptying rate for the whole white bean meal fell in between the other two meals (rice and potato). Mashed potato and mashed white bean meal were observed to have the same gastric emptying rate although; they elicited different blood glucose and insulin responses (Wursch et al, 1986). Rate of digestion of starch was the limiting factor for glucose absorption in this case.

Effects of carbohydrate foods on blood glucose response depend on their glycemic index values (Buclossi et al, 1990). High-GI foods cause higher and rapid rise in blood glucose after a meal which triggers insulin secretory response from beta cells of the pancreas leading to an unfortunately sharp decrease in blood glucose with consequent lower insulin secretion from the pancreas (Schulze et al, 2004). Whereas, low-GI foods cause slower and more sustained rise in blood-glucose levels with moderate insulin secretory response from pancreas (Brand-Miller et al, 2003).

Legumes (a class of high fiber containing protein and anti-nutrients) with starch-digested in-vitro, produced slow glycemic responses and in long term results in improved diabetic control (Throne et al, 1983). Dry beans are a major part of the traditional foods in diets of many countries in Africa, India, Central and South America (Osilesi et al, 1993). Reddy et al, 1984 showed that the total starch in dry seeds range from 20% (winged beans) to 56.5% (pinto beans).

Processing of the leguminous seeds increases significantly the digestion rate and subsequently blood glucose and insulin responses up to nearly the level produced by bread (Jenkins et al, 1982). Cooking of leguminous seeds elicited less metabolic responses than those of cooked cereals and potato in both healthy and diabetic individuals (Bornet et al, 1987).

Simpsons et al (1981) suggested that dried leguminous seeds could play a valuable role in the carbohydrate tolerance and may be usefully incorporated into therapeutic diets of NIDDM. Substitution of white peas beans into a test meal in place of bread reduced the glycemic response of NIDDM to more than 50% (Jenkins et al, 1984). However, no significant reduction was seen with IDDM patients.

Fat slows down gastric empty thus, delays the glycemic response peak with overall reduction of the glycemic response and the effect is more pronounced for the rapidly absorbed carbohydrate from bread. Jenkins et al (1984) demonstrated that the addition of fat and protein to bread in the form of butter or skim milk cheese did not significantly reduce the overall glycemic response of the bread. Many comparative meal tolerance tests have been reported (Collier et al, 1986 and Bornet et al, 1987). The results were sometimes contradictory even when all the carbohydrates in the meals were starch.

Thus, the therapeutic application of methods of achieving a reduced rate of nutrient absorption with respect to carbohydrate includes high dietary fiber containing foods or use of viscous fiber supplements, enzyme inhibitors in foods/pharmacological agents e.g. acarbose and antinutrients, selection of low-GI starchy food, altered meal frequency e.g. nibbling and gorging, nature of starch e.g. amylose versus amylopectin and degree of gelatinization and retrogradation; and food form determined by the method of processing (cooking), degree of hydration or particle size (Jenkins et al, 1990).

In general, slowly digested foods produce flatter glycemic responses. Such foods are termed "lente" carbohydrate foods. Using isotopically labeled glucose, rate of carbohydrate absorption from the gut after a glucose load continue at a near maximum rate when the peripheral blood glucose levels had already returned towards baseline. It is the initial absorption

rate that appears related to the blood glucose rise. By inference, if absorption time were prolonged, the incremental blood glucose area would be smaller.

2.5 EFFECT OF PROCESSING ON DIETARY CARBOHYDRATES

Processing of foods affects carbohydrate micronutrient content and bioavailability in different ways with either desirable or adverse effects on the nutritional values (Foster- Powel et al, 2002). The bioavailability of starch is affected dramatically through processing regarding both rate and extent of small intestinal digestibility. This permits optimizing the digestion of starch by choice of raw materials and processing conditions (Foster- Powel et al, 2002). The role of food on gastric emptying rate if starchy meals are consumed, and the correlation with blood glucose response has been studied by Torsdottir et al (1984) on healthy and diabetic subjects.

The digestion rate of starch measured in-vitro correlated with glycaemic response (Jenkins et al, 1982). The glycaemic responses (in human and rats) of the processed cereals are generally in good agreement with the in-vivo rate of digestion, which is assumed to be the rate-limiting step in the in-vitro digestion process (Brand et al, 1985; Rose et al, 1987 and Krezowski et al, 1987). However, cereals fiber does not appear to affect the rate of hydrolysis except when it forms a physical barrier limiting access of the starch.

The rate and extent to which starch is digested and absorbed, and the resulting glucose and insulin responses vary considerably depending on the source and degree of food processing (Jenkins et al, 1980; Snow et al, 1981; Englyst et al, 1985-87 and Hearnton et al, 1988). One way of quantifying this variation in response to dietary carbohydrates is the glycaemic index (GI) pioneered by Jenkins et al (1981).

Differences in glycaemic response to carbohydrates are brought about by the method of cooking. A much greater blood glucose response occurs after the consumption of cooked starch

compared with raw starch and purred compared with whole foods (Collings et al, 1981 and O'Dea et al, 1980).

Many foods eaten today in Nigeria are prepared under factory processing conditions very different from the conventional cooking methods. The food industry has developed a wide range of convenient and novel snack products which are ready-to-eat or minimize preparation in the home and have increased storage life. New processes/methods such as extrusion cooking, explosion puffing and instantization which make use of extreme temperature and pressure or repeated wetting and drying may affect the digestibility of starch as they make starch readily available for digestion. Conditions which are known to increase the digestibility of starches are those which produce obvious hydration of the granules (gelatinization), distinct changes in chemical nature or disruption in the organized structures (Boohher et. al, 1951)

Such conditions increase the availability of starch to amylase and are more likely to occur during factory processing because of the higher temperature and pressure involved. In extrusion puffing or cooking, enough energy and heat must be applied to thoroughly gelatinize or cook the ingredients (Kent et. al, 1987)

In contrast, conventional cooking methods such as boiling involves less physical disruption in moderate heat, and are therefore less likely to cause starch damage or complete gelatinization. Difference in fiber content, trace minerals concentrations and other nutrient could contribute to difference in the glycemic response to ingested food.

The metabolic response to carbohydrate ingestion plays an important role in healthy and in disease states such as diabetes mellitus and hyperlipidaemia (Jenkins et al, 1986). Many metabolic studies now have shown that food sources of carbohydrate vary greatly in their rate of absorption and effect on blood glucose and insulin concentrations. Present knowledge of the

variation in glycemic response to carbohydrate-containing foods comes largely from measurement of glycemic index.

In principle, the glycemic index is calculated as the measured glycemic response to a portion of a test food containing 50g “available” carbohydrate expressed as a percentage to the same amount of “available” carbohydrate from a standard food eaten by the same subjects (Jenkins et al, 1981 and Wolever et al, 1991; 1996). Glycemic index values have been published for a wide range of foods (Foster-Powel et al, 2002) and have been used in several studies to design low-glycemic load diets for diabetic subjects.

Studies have shown that processing of foods with different methods results in significant differences in the rate and extent of carbohydrate digestion and hence the glycemic index (GI) value. Two different brands of the same type of food such as a plain cookie may look and taste almost the same, but difference in the type of flour used, moisture content and the cooking time can result in differences in the degree of gelatinization and consequently the GI values (Wolever et al, 2002).

The human diet contains many types of carbohydrates, each of which contributes to different physiologic responses. Diets rich in rapidly digested carbohydrates (High-glycemic index diets) have been suggested to have detrimental effect on health due to high level of postprandial blood glucose and insulin responses associated with them (Salmeron et al, 1997) while slowly digested carbohydrates (Low-glycemic index diets) may protect against chronic diseases (FAO/WHO, 1997).

The nutritional quality of the carbohydrates and the effects of processing on that quality is a major concern when considering foods to be incorporated into diabetic diets because processing in a number of ways can alter both the content and the nutritional quality of food carbohydrates.

Clinical studies of persons with diabetes have shown a good positive correlation between improved glycemic control and diets with low glycemic index (Brand et al, 1991; Wolever et al, 1992; Fontvielle et al, 1992 and Frost et al, 1994). Wolever et al (1992) studied the beneficial effects of a low-glycemic index diets in type 2 diabetes mellitus.

Studies on the control of diabetic complications have shown a decrease in progression of diabetic complications as a result of euglycemic control from dietary therapy (Hodges et al, 2004). Dietary advice based on the glycemic index improves dietary profile and metabolic control in type 2 diabetes mellitus (Frost et al, 1994). Thus, food processing favouring low glycemic index should be advocated.

Processing effects on dietary fibre include solubilization and depolymerization that can influence the physiological response both in the upper and lower gastrointestinal tract. Formation of resistant starch and use of resistant oligosaccharides as food ingredients provide new opportunities to increase the amount of carbohydrate available for colonic fermentation. Pasta has a medium-GI value of 40-50, which can be further reduced by cooking it less (al dente). This is because al dente pasta resists the effect of digestive enzymes more than regular cooked pasta and so has a lower GI (foster-Powell et al, 2002).

Low molecular weight carbohydrates are lost during food processing. The degree of loss varies from one form to another (Nyman et al, 1987). Losses of water-soluble nutrients at blanching and boiling can be minimized by use of small amounts of water and by adding back the processing water. During wet heat treatment, as in blanching, boiling and canning of vegetables and fruits, there is a considerable loss of low molecular weight carbohydrates (i.e. mono- and disaccharides) as well as micronutrients, into the processing water. Nyman et al (1987) in his study revealed that during the blanching of carrots and swedes (rutabagas) there

was a loss of 25% and 30%, respectively of these carbohydrates. With subsequent boiling, another 20% was lost. In peas, green beans and Brussels sprouts the loss was less pronounced – about 12% following blanching and another 7-13% at boiling (Nyman et al, 1987).

Loss of low molecular weight carbohydrates during food processing varies with carbohydrates type, species, storage duration and time of harvest. The loss of glucose and fructose at boiling was higher than that of sucrose (Svanberg et al, 1997). The losses of low molecular weight carbohydrates in carrots which seems to be relatively easy to predict by knowing initial concentrations and process conditions of the raw material have also been shown to differ between various cultivars (species), time of harvest and storage duration. After storage, the loss of low molecular weight carbohydrates increases following boiling most probably due to the higher water content and possibly to a higher diffusivity (Oliviera et al, 1989).

Loss of dietary fiber during food processing was studied by Nyman et al (1987). In his study, no leaching of dietary fiber into the processing water was reported for carrots, green peas, green beans and Brussels sprouts following blanching, boiling and canning. However with Swedes, there was a 40% loss of dietary fiber (mainly insoluble) with boiling. Also with canning there was a leakage of insoluble fiber into the processing water.

The production of resistant oligosaccharides by enzyme technology is an expanding area. More than half of the “functional foods” on the Japanese market contain prebiotic oligosaccharides as active component, with the aim of promoting favourable gut microflora. Fructo-oligosaccharides synthesized from sucrose (Bornet et al, 1994) and galacto-oligosaccharides synthesized from lactose are the most extensively used types of resistant oligosaccharides. Alternatively, fructo-oligosaccharides can be produced by hydrolysis of inulin.

Non-enzymatic browning reactions (Maillard reactions) have been observed to occur between reducing sugars and amino groups in foods at processing and in storage. These reactions are temperature dependent and most extensive at intermediate water activities. They are important nutritionally as they may diminish the bioavailability of amino acids, especially lysine, thus diminishing the protein nutritional value. The carbohydrate content and availability is influenced only marginally. When a non-reducing disaccharide such as sucrose is replaced by, for example, high fructose corn syrup containing glucose and fructose, Maillard reactions occur much more rapidly and extensively. This has to be kept in mind in selecting processing procedures and storage conditions.

Heat-induced effects on starch include: Gelatinization, Retrogradation, and Starch-texturization. Upon heating in the presence of water, a process called gelatinization (irreversible loss of the crystalline regions in starch granules) occurs. The temperature range during which the crystalline structure of the starch granule is lost is dependent on the water content, and on the type of starch. The gelatinization dramatically increases the availability of starch for digestion by amylolytic enzymes (Granfeldt et al, 1995).

Usually, the starch granules are not completely dissolved during food processing, and a food can be regarded as dispersion in which starch granules and/or granular remnants constitute the dispersed phase. The degree of gelatinization achieved by most commonly used food processes, however, is sufficient to permit the starch to be rapidly digested. Consequently, food processes that even result in a low degree of gelatinization (e.g. steaming and flaking of cereals) produces a postprandial blood glucose and insulin increment similar to that with completely gelatinized foods (Granfeldt et al, 1995 and Hagander et al, 1987).

A progressive re-association of the starch molecules as a result of thermodynamic inequilibrium of gelatinized starch occurs upon ageing (Eliasson et al, 1996). This recrystallization referred to as retrogradation, may reduce the digestibility of the starch. The retrogradation of the amylopectin component is a long-term phenomena occurring gradually upon storage of starchy foods. Amylose, however, re-associates more quickly. The crystallinity of retrograded amylopectin is lost following re-heating to approximately 70°C, whereas temperatures above 145°C are required to remove crystallinity of retrograded amylose. This is a temperature well above the range used for processing of starchy foods. This implies that retrograded amylose, once formed, will retain its crystallinity following re-heating of the food (Eliasson et al, 1996).

Par boiling, another method of food processing, affects the content and the quality of carbohydrate foods. During par boiling of rice, the kernels are subjected to a pre-treatment involving heating and drying. This process reduces the stickiness of the rice, possibly by allowing leached amylose to retrograde and/or form inclusion complexes with polar lipids on the kernel surface. Parboiling also affects the final cooking properties of the rice.

In pasta products, gluten forms a viscoelastic network (texturization) that surrounds the starch granules, which restricts swelling and leaching during boiling. Pasta extrusion is known to result in products where the starch is slowly digested and absorbed (Granfeldt et al, 1991 and Wolever et al, 1986). Available data on spaghetti also suggest that this product group is a comparatively rich source of resistant starch (Englyst et al, 1992).

The slow-release features of starch in pasta probably relates to the continuous glutenous phase. This not only restricts swelling, but possibly also results in a more gradual release of the starch substrate for enzymatic digestion. Pasta is now generally acknowledged as a low glycemic index food suitable in the diabetic diet. However, it should be noted that canning of pasta

importantly increases the enzymic availability of starch, and hence the glyceemic response (Holm et al, 1992).

During milling of cereal grains to refined flours the outer fibre-rich layers are removed, resulting in a lower content of total dietary fibre. This reduction is due mainly to a decrease of insoluble fibre. The dietary fibre composition in both whole-grain and refined flours is different. Refined flours of oats, barley, rice and sorghum contain mainly glucans, while arabinoxylans dominate in refined flours of wheat, rye and maize. Whole-grain flours all contain considerable amounts of cellulose. The husk, which surrounds barley, rice and oats, also contains considerable amounts of xylans. This fraction is generally removed before consumption, but oat and rice husks are used for fibre preparation to enrich foods.

Processes involving heat-treatment may affect the dietary fibre in different ways. An increased temperature leads to a breakage of weak bonds between polysaccharide chains. Also glycosidic linkages in the dietary fibre polysaccharides may be broken. These changes are important from analytical, functional and nutritional points of view. A decreased association between fibre molecules, and/or a depolymerization of the fibre, results in a solubilization. If the depolymerization is extensive, alcohol soluble fragments can be formed, resulting in a decreased content of dietary fibre with many of the currently used fibre methods. Moderate depolymerization and/or decreased association between fibre molecules, may have only minor influence on the dietary fibre content, but functional (e.g. viscosity and hydration) and physiological properties of the fibre will be changed.

Other reactions during processing that may affect the dietary fibre content and its properties are leakage into the processing water, formation of Maillard reaction products thus adding to the lignin content, and formation of resistant starch fractions. Also structural alterations in the cell

wall architecture are important to follow during processing as these are highly correlated to sensory and nutritional characteristics. The architecture of the fibre matrix in the cell wall differs between various types of plant material. The cross-linking of constituent polysaccharides and phenolics within the cell wall is important in determining the properties of the fibre matrix, as the solubility of the fibre is highly dependent on the type and amount of cross-links present.

During heat-treatment the cell wall matrix is modified and the structural alterations that occur may be important not only for the nutritional properties of the product but also for its palatability.

With extrusion-cooking of wheat-flour, even at mild conditions, the solubility of the dietary fibre increases (Björck et al, 1984). The solubilization (at least for whole-grain wheat flour and wheat bran) seems to be dependent on the water content used in the process, and the lower the content of water, the higher the solubilization of the fibre (Ralet et al, 1990). The screw speed and the temperature had minor effects in those experimental studies. An increased solubility of the fibre has also been obtained with 'severe' popping of wheat (Nyman et al, 1987), whereas baking (conventional and sour-dough baking), steam-flaking and drum-drying had only minor effects on dietary fibre components (Siljeström et al, 1986).

One reason why popping caused an increased solubility of the fibre was that the outer fibrous layers were removed and the content of insoluble fibre decreased. Considerable amounts of Maillard reaction products were also formed during this process. A loss of insoluble dietary fibre has also been reported with autoclaving of wheat flour, which was attributed to degradation of the arabinoxylans (Siljeström et al, 1986).

Most raw materials containing cereal fibres are ground for better acceptance of the final product and this process can affect hydration properties. Swelling and water-binding capacity of

pea hull fibres are decreased by grinding, whereas the water-holding capacity was slightly increased (Ralet et al, 1993). The kinetics of water-uptake was also different, and the ground product hydrated instantaneously in contrast to the unground product, which reached equilibrium only after 30 minutes. This was related to the differences in surface area.

Caprez et al (1986) showed that heat-treatment could also change hydration properties. For example, boiling increased the water-binding capacity slightly in wheat bran and apple fibre products, whereas autoclaving, steam-cooking and roasting had no significant effects. The kinetics of water uptake, however, was different for steam-cooking and roasting. Thus, both products exposed to steam-cooking had a very rapid water-uptake, whereas the roasted sample had a slow uptake. Extrusion-cooking of pea-hulls, sugar-beet fibres, wheat bran and lemon fibres had only slight effects on the water-binding capacity.

Cooking of leguminous seeds elicited less metabolic responses than those of cooked cereals and potato in both healthy and diabetic individuals (Bornet et al, 1987). Simpsons et al (1981) suggested that dried leguminous seeds could play a valuable role in the carbohydrate tolerance and may be usefully incorporated into therapeutic diets of NIDDM. Substitution of white pea's beans into a test meal in place of bread reduced the glycemic response of NIDDM to more than 50% (Jenkins et al, 1984). However, no significant reduction was seen with IDDM patients.

2.6 CARBOHYDRATES AND GLYCEMIC INDEX

Over the years, it has been demonstrated that the ingestion of various types of starchy foods has various effect on postprandial glucose and insulin responses in healthy and diabetic subjects. Recently, the Glycemic Index (GI) has become the benchmark for classifying carbohydrates. It replaces the older method of classification based on their structure. The glycemic index is a measure of how rapidly a particular food causes blood sugar to rise

compared with glucose (Jenkins et al, 2002). By this index, according to Chlup et al (2004), foods are classified into high-GI foods (foods with a GI rating of 70 and above, causing a strong and rapid rise in blood glucose levels), intermediate (medium)-GI foods (foods with a GI rating between 55 and 69 causing a medium rise in blood glucose levels) and low-GI foods (foods with GI rating of 54 or less, causing a slower rise in blood glucose levels).

In the determination of glycemic index of food, different foods have been used as the reference or standard meal. Wolever (1990) in his study used white bread or 75g of glucose while Chlup et al (2004) used 50g of pure glucose as the reference food. The choice of bread as a reference or standard food is not unanimously accepted because the glycemic response to bread differs consistently from one report to another when compared to glucose (Jenkins et al, 1986 and Crapo et al, 1981).

Insulin secretion and therefore insulinaemic index was found to vary from one food to another in the same direction as the glycemic response in normal and type 2 diabetes (Crapo et al, 1997; 1981). The glycemic index, invented in 1981 by David Jenkins and Thomas Wolever of the University of Toronto, is a new system for classifying carbohydrate-containing foods, according to how fast they raise the blood glucose levels after a meal. This new system for classifying carbohydrates calls into question many of the old assumptions about how carbohydrates affect health. This approach of the glycemic index in the classification of food is a useful one provided the possible effects of other specific meal constituents are recognized (Jenkins et al, 1984). The approach is likely to apply well with respect to planning dietary pattern for 80-90% of the diabetic population (NIDDM).

The glycemic index (GI), measures how fast and how far blood sugar rises after carbohydrate meal consumption (Jenkins et al, 2002). White bread, for example, is converted

almost immediately to blood sugar, causing it to spike rapidly. It is classified as having a high glycemic index. Brown rice, in contrast, is digested more slowly, causing a lower and gentler change in blood sugar. It has a low glycemic index (Jenkins et al, 2003). Foster-Powell et al, (2002) published the most comprehensive list of the glycemic index of foods.

Diets filled with high-GI foods have been linked to an increased risk for both diabetes (Schulze et al, 2004; Willet et al, 2002) and heart disease (Liu et al, 2002; Pereira et al, 2003). On the other hand, low-GI foods have been shown to help control type 2 diabetes mellitus (Brand-Miller et al, 2003).

According to Coulston et al (1984), the establishment of glycemic index ignores the insulin response entirely despite the fact that insulin response might be of more clinical importance in the non-diabetic population. Results of recent studies have documented that the insulin response to specific carbohydrate can be quite divergent in the face of similar glycemic responses (Jenkins et al, 1986). This is true for subjects with normal glycemic response, impaired glucose tolerance and non-insulin dependent diabetes mellitus (NIDDM).

2.6.1 Factors Influencing the Glycemic Index Value of Carbohydrates

It has been observed by various researchers (Thorburn et al, 1986; Laine et al, 1987; Marion, 2000; Gannon et al, 2001; Foster-Powell et al, 2002; Jenkins et al, 2003; Chlup et al, 2004) that the GI values of carbohydrates are influenced by several factors namely:

- (1) **Chemical and physical structure of the carbohydrate:** -Carbohydrate with molecular structure facilitating quick metabolism or digestion is associated with high GI value. The GI of glucose is 100 due to that the body processed it efficiently. Fructose has a low GI of 23 because the body cannot easily

metabolize it. Ordinary table sugar (sucrose) has a glycemic index of 65, midway between 23 and 100 in the medium-glycemic-index range. (Foster-Powell et al, 2002). Finely ground grain is more rapidly digested due to great surface area provided for metabolism by the digestive enzymes than coarsely ground grain. As a result, the former has a higher GI than the latter. (Foster-Powell et al, 2002).

(2) Degree of refinement of the carbohydrate:

Highly refined or processed carbohydrates have higher GI values than less refined carbohydrates (Brand miller et al, 2003). In general, refined or processed carbohydrates have had most of their 'natural' fiber and other 'inconvenient' constituents (which for example may affect the food's shelf-life) removed. The carbohydrate is incapable of resisting the digestive enzymes and is rapidly metabolized into glucose. Processed foods have higher GI value compared with conventionally cooked food of the same type but not always e.g. potatoes (Brand Miller et al, 2003).

(3) Methods of preparation or cooking: -

Pasta has a medium-GI value of 40-50, which can be further reduced by cooking it less (*al dente*). This is because *al dente* pasta resists the effect of digestive enzymes more than regular cooked pasta and so has a lower GI (foster-Powell et al, 2002). This observation was also made in this study using white yam as a test diets. According to Fasanmade and Anyakudo (2007), the difference in the processing method of wheat as white bread and semolina showed significant difference in their observed glycemic index values and responses.

(4) Fiber content: -

Fiber (either in the carbohydrate itself or in the stomach) protects the starchy carbohydrate from rapid attack by digestive enzymes, or slows digestion in the digestive tract. Either of these consequences will slow down the conversion of the carbohydrate to glucose (Jenkins et al, 2003 and Hodges et al, 2004). Thus, fiber reduces the glycemic index of carbohydrates

(5) Fat and protein content: -

Protein has minimal effect on the absorption of carbohydrates or the glucose response peak whereas fat has little effect on blood glucose levels. Fat delays the peak but not the total glucose response (Marion, 2000 and Gannon et al, 2001). The more fat or acid a carbohydrate food contains, (or, the more fat or acid in the stomach, during digestion) the slower the carbohydrate food is converted to glucose and absorbed into the bloodstream. The presence of fat and/or acid retards the emptying of the stomach.

(6) State of ripeness of the fruits: -

Ripe fruits and vegetables tend to have more sugar than unripe ones, and so tend to have a higher glycemic index. Process of ripening of fruits converts complex carbohydrates to simple carbohydrates. This mechanism tends to reduce the polysaccharides and disaccharides often present in the unripe fruits to monosaccharides which are present in abundant in ripe fruits (Brand miller et al, 2003).

(7) Presence of metabolic disorder: -

In certain disease conditions such as diabetes mellitus, glycemic responses to carbohydrates have been found to be affected. People with diabetes mellitus have higher glycemic responses and mean incremental areas under the curve for blood

glucose response curve when compared with the non-diabetic healthy people. Therefore, the glycemic index-classification of foods has provided means of selecting carbohydrates that may be useful in the management and control of such metabolic disorder (Donduran et al, 1999 and Thorburn, 1986).

All these factors above influencing the glycemic index of carbohydrates, lead to Counterintuitive results sometimes. Some foods that contain complex carbohydrates such as potatoes, quickly raise blood sugar levels, while some foods that contain simple carbohydrates, such as whole fruit, raise blood sugar levels more slowly.

Sex, age and time of the day do not alter the GI values of carbohydrates (Chlup et al, (2004). Thus, selection of male participants only in this study was not biased. Chlup et al, (2004) in their study, determined the GI of foods in males and females at different time of the day in different age groups and observed that there was no statistically significant difference in the GI of the tested foods in females and males of different age groups at different time of the day.

2.6.2 Glycemic Values of Meals

Generally, human beings do not typically consume measured portions of single-carbohydrate foods. Rather, they ingest meals containing a combination of foods that contain varying amounts of carbohydrates, plus protein, fat and fiber. So, it is often more important to know the glycemic value of a meal, rather than simply the GI of individual foods.

Wolever et al (1984) found good correlation between the calculated glycemic index of the meals and the respective glycemic responses of the component food. They also demonstrated that the mean glycemic responses of different mixed meals in diabetic subjects are proportional to the GI for individual carbohydrate food present in the meal. The situation is more confusing when the mixed meal contains starch and sugar from various sources (Coulston et al, 1981).

Workers who tested mixed meals with comparable distribution of carbohydrate, fat and protein on NIDDM patients found a good correlation between incremental blood glucose area and the calculated glycemic index (Wolever et al, 1986).

Effect of various carbohydrate rich foods on glycemic and insulin responses when fed as part of a standard meal with representative portions of fat, carbohydrate, protein and calories is essential in order to judge the potential therapeutic utility of prescribing diabetic diets based on the glycemic index of the individual carbohydrate-rich food (Coulston et al, 1984).

2.6.3 Calculating the GI value of a meal

The GI value of a meal according to Jenkins et al, 1981 and Wolever et al, 1991 is calculated by:

1. Adding up the total grams of carbohydrates in the meal.
2. Calculating the percentage of the carbohydrate-total contributed by each food and finally,
3. Multiplying the percentage contributed by each food by the food's GI

Example If a meal contains 60g carbohydrate and includes two slices of bread (26 grams of carbohydrates), the bread accounts for 43 percent of the total. Since the GI for bread is about 70, therefore 43 percent of 70, which is about 30, are the GI value of the whole meal.

2.6.4 Reducing the GI values of a meal

Since glycemic index only concerns carbohydrates, if the only carbohydrate in a meal has a high GI, then the GI of the whole meal is high. For example, if the only carbohydrate in a meal is bread, the GI of that meal is 70. However, the glycemic effect of the meal can be reduced in several ways by:

- Including a low GI food like beans or berries.

- Adding vinaigrette or any acidic extra, like lemon juice so as to retard the gastric emptying thus, reducing the glycemic index.
- Adding a little olive oil which reduces GI by the effect of its fat content that also delays gastric emptying.

2.6.5 Measurement of GI of particular foods

The glycemic index of a food is measured under strict conditions. Portions of a carbohydrate food are fed to a group of volunteers whose blood samples are tested at regular intervals, over 2 hours, in order to check blood-glucose levels (Jenkins et al, 2003). In measuring the glycemic index of a particular food, Wolever (1991) and Jenkins (1981) described the procedures as follows in this order:

5-10 or more volunteers are given a serving of the test-food containing 50 grams of digestible (available) carbohydrate. The actual portion size of each tested food will vary according to how much carbohydrate it contains. The smaller the percentage of carbohydrates in the food, the larger the portion needs to be given to provide the standard 50-gram amount of digestible carbohydrates. To test how the food raises blood-glucose levels, a sample of blood is taken from each subject at interval of 60 minutes for two-hour duration. The glucose level of these blood samples is then measured and recorded using a glucometer.

The glucose level (mg/dl) against time (minutes) is plotted on a graph to obtain the blood glucose response curve. From the graph the incremental area under the curve is calculated and the results are interpreted using a computer program.

The volunteer's response to the food being tested is compared with his/her blood-glucose response to 50g of pure glucose dissolved in a given amount of potable water. (Glucose is the reference food and the testing of glucose on the subject's blood sugar levels is done on a separate

occasion).

The average blood sugar response from 5-10 people will determine the glycemic index (GI) value of that food (Chlup et al, 2004).

2.6.6 Glycemic Index Variability

The glycemic index of foods is relatively constant between people and even, mixed meals have a fairly predictable effect according to most (but not all) studies (Wolever et al, 1990). However, there are some instances of wide and large inter- and intra-individual variability for the same food due to inherent botanical differences from country to country rather than to methodological differences. A good example of this is rice. The difference in the amylose content, which is digested more slowly than amylopectin starch, could explain much of the variation of the glycemic index values of rice and other similar foods (Donduran et al, 1999 and Rasmussen et al, 1993).

Variations amongst investigators may be as a result of different protocol. Crapo et al (1983) have consistently used protocol of individual carbohydrate containing foods equivalent to 50g glucose along with 500ml of water. Jenkins et al (1981) controlled the total amount of carbohydrate to equal 50g of glucose but have added variable amount of other foods such as milk. Chlup et al (2004) used protocol of individual carbohydrate containing foods equivalent to 50g glucose along with 300ml of water while Coulston et al (1987), provided the test meals in the context of a standard meal in which the carbohydrate represent 45% of the caloric content of the meal.

The methodology used in determining the carbohydrate content of the test foods might also explain the variation of the GI of similar food. An essential preliminary step in GI testing is to measure or obtain accurate laboratory measurement of the available carbohydrate content of

foods. GI testing requires that both portion of the standard and the test food, contain the same amount of carbohydrate typically 50g (Chlup et al, 2004 and Wolever et al, 1991).

Foods with low-GI may be used to achieve the reduction of fat intake without increasing postprandial glycaemia. Despite the addition of fat, lentils (a low glycemic food) still produced an appreciably lower glycemic response than potatoes, a food with higher glycemic index.

2.6.7 Drawback of glycemic index

By using the Glycemic Index alone, the glycemic effects of foods containing a small percentage of carbohydrates are likely to be *overstated*, while the glycemic effects of foods containing a high percentage of carbohydrates are likely to be *understated*. For example, foods that are mostly water or air rich will not cause a surge in blood sugar levels even if their glycemic index is high (Liu et al, 2000). As a result of this drawback, scientists have developed the idea of **Glycemic Load**. It ranks foods according to actual carbohydrate content (e.g. in a typical portion-size), not how fast a 50g amount of carbohydrates raises blood sugar levels.

2.6.8 Glycemic Load

Glycemic Load is the application of the glycemic index to a standard serving of food. The glycemic index (GI) of a food is not based on commonly consumed portion-sizes of foods. Instead, GI is measured by giving volunteers a portion size sufficient to contain 50g of useable carbohydrates. Therefore, the portion size of each GI-tested food will vary according to how much carbohydrate it contains. For example, carrots contain only about 7 percent carbohydrates, so the test-portion of carrots eaten by the test-volunteer will be huge. Serving sizes of foods (like bread), which contain a higher percentage of carbohydrates, will be smaller (Liu et al, 2000). The glycemic load (GL) is a new way of classifying foods that takes into account both the amount of carbohydrate in the food and the impact of that carbohydrate on blood sugar levels.

2.6.8.1 Measurement of Glycemic Load

Food's glycemic load is determined by multiplying its glycemic index by the amount of carbohydrate it contains (Liu et al, 2000). To calculate glycemic load in a typical serving of food, the GI of that food is divided by 100 and multiplied by the useable carbohydrate content (in grams) in the serving size. For example, the glycemic index of carrots is about 47. Carrots contain about 7 grams of carbohydrate per 100g of carrots. So, to calculate the glycemic load for a standard 50g serving of carrots, the GI (47) is divided by 100 (0.47) and multiplied by 3.5. The glycemic load (GL) of carrots is therefore 1.6 (Liu et al, 2000).

2.6.9 Significance of glycemic Index

Although the glycemic index was invented originally to help diabetic patients manage their blood-sugar levels, dietitians and weight experts now use it as a tool to treat obesity, reduce cravings and appetite swings, and improve eating habits (Brand Miller et al, 2003). As originally proposed, the GI was intended to provide the physiological data on the blood glucose response in man to a range of foods which can be used as a supplement to tables based solely on chemical analysis (Jenkins et al, 1981). According to O'Dea (1983), it is an over simplification to imply that all foods with low-GI are appropriate to inclusion in the diet of people with diabetes. This does not take into account the longer-term effect of other nutrients (Wolever et al, 1983).

An important function of glycemic index is that it allows for the identification of starchy carbohydrate foods that may be incorporated into higher carbohydrate diets now being recommended for the management of diabetes (Wolever et al, 1983).

2.6.9.1 Glycemic Index Food Chart

table 1 shows the list of glycemic index values of foods. The reference value of the glycemic-index chart is Glucose (GI = 100) (Foster-Powell et al, 2002).

Table 1

Glycemic Index of Common Foods (Temperate region) Foster-Powell et al (2002).

Glycemic Index of Cereals	Glycemic Index of Grains	Glycemic Index of Pasta
Kellogg's All Bran 51 Kellogg's Bran Buds 45 Kellogg's Cornflakes 84 Kellogg's Rice Krispies 82 Kellogg's Special K 54 Oatmeal 49 Shredded Wheat 67 Quaker Puffed Wheat 67	Buckwheat 54 Bulgur 48 Basmati Rice 58 Brown Rice 55 Long grain White Rice 56 Short grain White Rice 72 Uncle Ben's Converted 44 Noodles (instant) 46 Taco Shells 68	Spaghetti 43 Ravioli (meat) 39 Fettuccini (egg) 32 Spiral Pasta 43 Capellini 45 Linguine 46 Macaroni 47 Rice vermicelli 58
Glycemic Index of Fruits	Glycemic Index of Vegetables	Glycemic Index of Breads inc. Muffins & Cakes
Apple 38 Banana 55 Cantaloupe 65 Cherries 22 Grapefruit 25 Grapes 46 Kiwi 52 Mango 55 Orange 44 Papaya 58 Pear 38 Pineapple 66 Plum 39 Watermelon 103	Beets 69 Broccoli 10 Cabbage 10 Carrots 49 Corn 55 Green Peas 48 Lettuce 10 Mushrooms 10 Onions 10 Parsnips 97 Potato (baked) 93 Potato (mashed, instant) 86 Potato (new) 62 Potato (48rench fries) 75 Red Peppers 10 Pumpkin 75 Sweet Potato 54	Bagel 72 Blueberry Muffin 59 Croissant 67 Donut 76 Pita Bread 57 Pumpernickel Bread 51 Rye Bread 76 Sour Dough Bread 52 Sponge Cake 46 Stone Ground Whole wheat bread 53 Waffles 76 White Bread 70 Whole Wheat Bread 69
Glycemic Index of Beans	Glycemic Index of Snacks	Glycemic Index of Dairy
Baked Beans 48 Broad Beans 79 Cannellini Beans 31 Garbanzo Beans (Chickpeas) 33 Lentils 30 Lima Beans 32 Navy Beans 38 Pinto Beans 39 Red Kidney Beans 27 Soy Beans 18 White Beans 31	Cashews 22 Chocolate Bar 49 Corn Chips 72 Jelly Beans 80 Peanuts 14 Popcorn 55 Potato Chips 55 Pretzels 83 Snickers Bar 41 Walnuts 15 Glycemic Index of Sugars Fructose 23 Glucose 100 Honey 58 Lactose 46 Maltose 105 Sucrose 65	Milk (whole) 22 Milk (skimmed) 32 Milk (chocolate flavored) 34 Ice Cream (whole) 61 Ice cream (low-fat) 50 Yogurt (low-fat) 33 Glycemic Index of Cookies Graham Crackers 74 Kavli Crispbread 71 Melba Toast 70 Oatmeal Cookies 55 Rice Cakes 82 Rice Crackers 91 Ryvita Crispbread 69 Soda Crackers 74 Shortbread Cookies 64 Stoned Wheat Thins 67 Vanilla Wafers Water crackers 78

2.7 Dietary Carbohydrates and Health

The relationship between diet and disease according to Mera (1994) has long been established. This relationship is increasingly assuming public health importance due to increase in the prevalence of diet-related non-communicable diseases in developing countries and the functional financial burden of these emerging diet related diseases on individual and health systems.

Non-communicable chronic diseases (NCCD) include such degenerative diseases as coronary heart diseases (CHD), cerebrovascular diseases, various cancers, diabetes mellitus, dental caries and osteoporosis (FAO/WHO, 1992). While excessive and unbalanced intake of food or dietary components, weaved with certain lifestyle changes have been implicated in the etiology of these diseases (Wielgoz, 1995), some specific dietary components and foods have been found to play important roles in reducing the risk and preventing the incidence of these diseases (Block et al, 1994).

The marked dietary changes related to these diseases include less consumption of complex carbohydrates and fiber, increased fat and saturated fat consumption and increased consumption of animal protein (Byer et al, 1995). A high intake of carbohydrates especially those with high glycemic indices are associated with increased risks for diabetes and coronary heart disease (Willet et al, 2002 and Schulze et al, 2004).

One of the mechanisms of the effect of these dietary habits and component in the etiology of these non-communicable chronic diseases is the increase elevation of the blood glucose concentration which if unregulated by normal body mechanism is associated with increased risk for diabetes (Schulze et al, 2004) and coronary heart disease (Liu et al, 2002 and Pereira et al, 2003). Jarret (1996) supported this view while implicating impaired glucose tolerance and

hyperglycemia as a risk factor in the development of cardiovascular disorder. Donahue and Orchard (1992) in their study revealed hyperglycemia as an indicator of cardiovascular risk.

Carbohydrates, as a major source of energy in the food, are macronutrients required in reasonably large quantities. When ingested in unreasonably large or excessive amount, it results in increased risk for metabolic disorder such as diabetes mellitus (Willet et al, 2002). In the past, studies on the relationship between carbohydrates and health were focused on simple carbohydrates like glucose and fructose (Jukes, 1986). However in recent years, much knowledge has been gained on the metabolic effect of complex carbohydrates in food partly from the study of nutritional and metabolic properties of dietary fibers, which are found to be edible plants in association with starch.

Brand et al (1990) in their study supported the hypothesis that slowly digested and absorbed carbohydrates in traditional food are a factor that helped to protect susceptible populations from developing diabetes mellitus. Thorburn et al (1987) showed that the traditional Australian Aboriginal and Pacific island foods produced significantly lower glycemic responses than the western starchy foods. According to their work, it was hypothesized that slow digestibility of native foods protected the genetically susceptible populations such as Australian Aborigines from developing diabetes. The studies of Jenkins et al (1983) and Brand et al (1990) have supported the hypothesis that the nature of carbohydrates in traditional foods is an important factor in protecting indigenous population from developing diabetes.

Nutritional factors affect our predisposition to wide varieties of diseases both acute and chronic. Nutritional problems broadly fall into two categories: Those due to insufficient intake relative to needs and infections and those due to an excessive or imbalanced intake of food or particular dietary component (FAO/WHO, 1992). The latter in association with changes in lifestyle bears strong relationship with chronic non-communicable diseases such as coronary

heart diseases, hypertension, diabetes mellitus, osteoporosis, different kinds of cancers and dental caries.

In 1982, Zimmet revealed in his study that during the century, the abandonment of subsistence activities (gathering, hunting and farming) among several indigenous populations has been associated with dramatic increase in prevalence of non-insulin dependent diabetes mellitus (NIDDM).

Many epidemiological studies have shown that diet related non-communicable disease that were formerly associated with developed prosperous countries are at present making themselves felt in developing countries (even the poorest) and they are at present the leading cause of death globally. Many factors have been identified as risks for developing these diseases. They include age, sex, genetics, dietary pattern, urbanization, alcohol consumption, tobacco smoking, obesity, high blood cholesterol and imbalance in antioxidant protective mechanisms (Jones et al, 1985).

2.8 Health Effect of Dietary Carbohydrate: High-GI Values versus Low-GI Value Carbohydrates

High-glycemic-index foods trigger strong insulin responses, thereby exposing the body to all the negative effects of insulin. By comparison, low-glycemic value foods do not provoke this insulin response. (Buclossi et al, 1990). Diets containing high-glycemic-index meals, which cause rapid and strong increases in blood sugar levels, have been linked to an increased risk for diabetes mellitus (Willet et al, 2002) and an increased risk for heart disease (Ludwig, 2002 and Pereira et al, 2003) while lower glycemic index diets have been shown to help control type 2 diabetes and reduce symptoms of insulin resistance (Hodges et al, 2004).

Over-consumption of high-glycemic-index foods has been linked to food cravings and

disordered eating patterns, as a result of repeated surges and falls in blood-glucose called “sugar spikes” (Donduran et al, 1999). Also it has been shown that over-consumption of high-GI carbohydrates may aggravate insulin resistance in patients predisposed to the condition. Insulin resistance (called Metabolic Syndrome X, or more properly, Insulin Resistance Syndrome) is believed to be a precursor of type 2 diabetes (Brand Miller et al, 2002).

Insulin resistance is believed to be a genetic condition, aggravated by obesity. However, some experts consider that it may be the result of a separate inherited sensitivity to high-glycemic-index carbohydrates (Liese et al, 2003 and Reaven, 2003). Despite the apparent associations between high-glycemic-index carbohydrates and insulin resistance, and the fact that the latter can be reduced by a lower glycemic index diet plan, there is no scientific evidence that insulin resistance is caused by eating carbohydrates with a high-glycemic-index rating. Insulin resistance is largely the result of genetics, strongly influenced by body fitness and body weight. (Brand miller et al, 2003

2.8.1 Diabetes and Diet: Role of Dietary Carbohydrates in Type 2 Diabetes Mellitus (The Glycemic Index Concept)

Diabetes mellitus, a major complex metabolic disorder, presents major challenges to public health. Most striking understanding of this disorder was acquired after the discovery of insulin in 1921. It is characterized by polyuria, polydypsia, polyphagia, weight loss in spite of polyphagia, hyperglycemia, glycosuria, ketosis and acidosis and if poorly managed, could result in coma (Passmore and Eastwood 1986).

Hyperglycemia, which is one of the chief signs of diabetes, is believed to be as a result of deficient production of insulin or could be due to the decrease sensitivity of the target organs to insulin or decreased action of insulin. Higher postprandial blood glucose levels have been shown

to be associated with all cause mortality even in healthy people. Therefore, lowering blood glucose levels is a major means of reducing the risk and minimizing the complications associated with type 2 diabetes mellitus (Hodges et al, 2004).

Classically, diabetes mellitus is classified into two main types: Insulin dependent diabetes mellitus (IDDM) also referred to as type 1- an auto immune disease which destroys the beta cells in the pancreas. Without insulin the patient dies. The second main type is referred to as Non-insulin dependent diabetes (NIDDM) also known as type 2 diabetes mellitus is due to an insulin resistance and reduced capacity to produce extra insulin to overcome this resistance (Petrie et al, 1997). In addition to these two main types, other classes of diabetes mellitus have been identified such as malnutrition related diabetes mellitus (MRDM), gestational diabetes mellitus (GDM) and impaired glucose tolerance (IGT).

Prevalence varies greatly between different communities but has been noted to increase as life expectancy increases. Over 100 million people of all walks of life throughout the world may be affected. (Guest and O'Dea, 1992). Diabetes has been found to be extremely high in people who go directly from hunter-gatherer existence and skip the agricultural part of development into an urbanized life (Jervell, 1995).

According to World Health Organization (WHO, 1991), diabetes is diagnosed if the 2 hours postprandial blood glucose is greater than 200mg/dl. Values below 140mg/dl are said to be normal while those between 140 and 200mg/dl are said to have impaired glucose tolerance. Many studies have shown that in all population studied, NIDDM is more common consisting of 90-95% (Petrie et al, 1997).

Aetiologically, several contributory/causative factors have been identified such as hereditary, race, lifestyle, nutritional status, stress, infection, altered immune function, altered metabolic and or physiological status, drugs and hormones (Osuntokun et al, 1971, Adetuyibi,

1976; nutritional news, 1992). It is widely presumed that genetic factors are probably more important in those who develop diabetes before the age of 40.

Dietary pattern are important factors contributing to health. Diets that are inadequate in energy and certain nutrients can lead to serious deficiency diseases and even death while dietary patterns reflecting excessive or imbalance intakes are linked with diet-related non-communicable diseases both of which are of growing global public health importance (FAO/WHO/ICN, 1992).

The fundamental principle underlying the dietary management of diabetes is the balancing of energy expenditure with nutritional sources of energy under the influence of insulin and other hormonal factors.

The aim of diet therapy is to achieve normoglycemia and maintain ideal body weight. Dietary advice is primarily given in diabetes mellitus to avert symptoms of hyper- and hypoglycemia and to eliminate or postpone secondary complications, which may arise e.g. atherosclerotic cardiovascular disease due to micro-, and macroangiopathy complication (Vessby, 1994). Thus, dietary recommendations in diabetes aim at normalizing blood glucose concentrations, serum insulin concentrations, blood lipid abnormalities and blood pressure elevation.

In type 1 diabetes, with the administration of insulin, the underlying treatment is the establishment of regular meals consistent with day-to-day caloric and carbohydrate intake. However in type 2 diabetes, the management principle is to decrease caloric intake. Thus, the cornerstone for its treatment is diet and exercise with oral hypoglycemic agents. In earlier management of diabetes, restriction of carbohydrate intake was recommended. Carbohydrate restriction to about 40% of total calories intake proved successful, particularly in Type 1 diabetes, but caused an elevation of dietary fat intake. In Type 2 diabetes, carbohydrate

restriction was shown to improve glucose tolerance, particularly when there is accompanying weight loss.

Many investigators noted that a high carbohydrate calorie restricted regimen resulted in improved carbohydrate tolerance (Brunzell et al, 1974 and Weinsier et al, 1974). Isocaloric regimen of low and high carbohydrate content was compared to eliminate the confusing influence of calorie restriction. The mechanism for improved glucose tolerance with high carbohydrate regimen appears to be related to increased peripheral sensitivity to insulin action. Simpson et al (1979) confirmed that high carbohydrate diet does not cause diabetic control to deteriorate over weeks.

Studies in man suggest that successful dietary treatments are associated with a flattening of the postprandial glucose curve with resultant reduced demand for insulin. Indeed, the substantial reduction in basal glucose concentration in the presence of a slight reduction in insulin requirement suggests that high carbohydrate diet will cause an improvement in diabetic control (Wolever, 1990).

The main rationale for providing a high carbohydrate intake has been the possibility of decreasing dietary fat and cholesterol intake, since diabetics who have their carbohydrate intake restricted consume greater proportion of fat. Such high fat intake has been associated with raised blood lipids and an increased risk of cardiovascular diseases (Jenkins et al, 1980).

However, very high carbohydrate diet has been observed to result in a rise in fasting triglycerides in hyperlipidemic patients, in diabetics and in normal subjects (MacDonald, 1978). Such carbohydrate-induced lipidemia has been linked to the high insulin levels stimulated by the high carbohydrate diet (Olefsky et al, 1974). Thus, the factors leading to the genesis of triglyceride rise may be relevant to the pathogenesis of endogenous hypotriglyceridaemia (Albrink et al, 1979). Since this might be an undesirable side effect of low fat, high carbohydrate

diets aimed at reducing cholesterol, it is important to know whether a rise in plasma triglycerides and insulin is an inevitable effect of very- high-carbohydrate diets or is modified by the type of high carbohydrate foods that comprise the diet.

Most of the studies demonstrating carbohydrate-induced lipidaemia have used diets in which over 70% of the calories are derived from carbohydrate but these carbohydrates has been supplied as sugars, dextrin or a mixture of these often as a liquid formula though sometimes as mixed natural foods. If dietary energy derived from fat is diminished, and the protein content not increased because of possible negative effects on renal function, there has to be a secondary increase of carbohydrate content of the diet to meet the need for dietary energy. There are also suggestions that carbohydrate rich foodstuffs may have beneficial effects of their own with regard to glucose homeostasis and serum lipid concentrations.

An increase content of fiber-rich, carbohydrate containing foods may also promote satiety and increase volume of foods, thereby reducing energy density and assisting in body weight reduction in overweight subjects (Vessby, 1994). Reduced rates of carbohydrate absorption resulting in less potential demand for insulin may be associated with improved diabetic control (Anderson et al, 1979). Recent recommendations for dietary management of diabetes mellitus state that diets need to be individualized so that there is improved glucose and lipid control in the patient (Wursch et al, 1997).

2.9 FIBER IN DIETARY MANAGEMENT OF DIABETES MELLITUS

The gastric effect of vegetal fiber has been noted as far back as 1883.it was suggested that dietary fiber might provide health benefits. The dietary fiber hypothesis of Burkitt and trowel (1975) suggested that the high incidence of colon cancer, coronary heart diseases, diabetes, obesity, hypertension and certain other related diseases among Caucasians might be

related to their low intake of dietary fiber.

Epidemiological data were provided in support of the hypothesis. Clinical and physiological studies, however confirmed that fiber does affect gastrointestinal function, glucose homeostasis as well as modulate serum lipid levels in man (Anderson et al, 1984). Dietary fiber has been observed to have important therapeutic Implications for certain conditions such as diabetes mellitus (Anderson et al, 1979 and Jenkins et al, 1980), hypertension, coronary heart disease and intestinal disorders (Jenkins et al, 1983).

Evidence is however emerging that plant fibers have important influences on human nutrition and gastrointestinal physiology because they alter the absorption and metabolism of many nutrients. The finding of reported rarity of diabetes in the rural Africans and the increased incidence in urban African suggest that high-fiber, high-carbohydrate diet might be protective against diabetes (Trowell, 1978). Conversely, it was suggested that low-fiber starchy food is diabetogenic factor in the susceptible human phenotype. This is called the dietary fiber hypothesis of aetiology of diabetes mellitus (Trowell, 1975).

Several studies have shown that diets high in fiber improve the glycometabolic balance of diabetic patients (Wolever et al, 1976 and Kiehm et al, 1976). The effect of fiber-supplemented diets on the glycaemic control of diabetic patients has been documented by various investigators such as Jenkins et al, 1980; Cohen et al, 1980; Ray et al. 1980 and Tagliaferro et al, 1985. Some of these studies used guar supplements and reported a change in fasting serum glucose, with values ranging from +4 to -13%. Significantly reduced glucosuria was also observed (Jenkins et al, 1980).

A statistically significant reduction of 39% in glucose values was reported when a combination of wheat fiber and guar was used (Ray, 1983). Serum cholesterol level was observed to decrease by all types of fiber supplement (Jenkins et al, 1980; Usitupa et al, 1981;

and Tagliaferro, 1985). All average reduction of 40% in insulin doses across studies has been observed when high carbohydrate, high fiber diets were consumed by diabetic patients (Kiehm, 1976; Anderson et al, 1979; and Simpson, 1979). All the studies reported a drop in average fasting serum glucose values, with reductions ranging from 6% -27% and averaging 17% across studies (Kiehm et al, 1976, Simpson, 1979, Anderson et al, 1980, Simpson et al, 1981 and Ney et al, 1982).

A significant reduction in glucosuria was observed in 2 of these studies (Simpson et al, 1981, Ney, 1982). Serum cholesterol values were significantly reduced in all the studies cited above with an average reduction of 20% across studies and a range of 10 to 32%. When standard carbohydrate and fiber were used, a significant reduction in fasting serum glucose of 14% and 35% respectively was observed (Kimoth et al, 1982 and Karlstrom et al, 1984). Thus diets high in carbohydrate and fiber improve glucose metabolism without increasing insulin secretion. High carbohydrate, high fiber diets lower fasting serum and peripheral insulin concentrations in response to oral glucose administration in both diabetic and non- diabetic individuals (Anderson, 1982) and insulin receptor binding to circulating monocytes increased with these diets (Aro, 1981). The effect of carbohydrate-rich diets in diabetes mellitus seems to be due to the amount of carbohydrate, the concomitant increase and type of dietary fiber and the metabolic status of the patient. Riccardi et al (1984) showed that an increase of carbohydrates from 42% -53% of energy without concomitant increase of dietary fiber did not confer any benefit compared with diet with a higher fat content and a low carbohydrate content.

However, when dietary fiber content also was increased, mainly from vegetable fiber, both the blood glucose profile and the serum lipoprotein composition were improved compared with the high-fat low-Carbohydrate diet. This indicated that there are beneficial effects of an increased content of dietary fiber when added to a diet with a moderately increased content of

carbohydrate.

It is generally accepted that diabetic patients should avoid foods containing “simple” carbohydrates so as to minimize blood glucose excursion after meal. However, test meals studies have indicated an overlap in the ranges of blood glucose to simple and complex carbohydrates (Jenkins et al, 1984). Thus, the distinction, which for many years, has been made between sugars and starch may not be relevant to the blood glucose control achieved by diabetic patients (Wolever et al, 1985).

2.9.1 Lipid Profile and Glycemic Index

The main rationale for providing a high carbohydrate intake has been the possibility of decreasing dietary fat and cholesterol intake, since diabetics who have their carbohydrate intake restricted consume greater proportion of fat. Such high fat intake has been associated with raised blood lipids and an increased risk of cardiovascular diseases (Jenkins et al, 1980).

However, very high carbohydrate diet has been observed to result in a rise in fasting triglycerides in hyperlipidemic patients, in diabetics and in normal subjects (MacDonald, 1978). Such carbohydrate-induced lipidemia has been linked to the high insulin levels stimulated by the high carbohydrate diet (Olefsky et al, 1974). Thus, the factors leading to the genesis of triglyceride rise may be relevant to the pathogenesis of endogenous hypotriglyceridaemia (Albrink et al, 1979). Since this might be an undesirable side effect of low fat, high carbohydrate diets aimed at reducing cholesterol, it is important to know whether a rise in plasma triglycerides and insulin is an inevitable effect of very- high-carbohydrate diets or is modified by the type of high carbohydrate foods that comprise the diet.

Most of the studies demonstrating carbohydrate-induced lipidaemia have used diets in which over 70% of the calories are derived from carbohydrate but these carbohydrates has been

supplied as sugars, dextrin or a mixture of these often as a liquid formula though sometimes as mixed natural foods. If dietary energy derived from fat is diminished, and the protein content not increased because of possible negative effects on renal function, there has to be a secondary increase of carbohydrate content of the diet to meet the need for dietary energy. There are also suggestions that carbohydrate rich foodstuffs may have beneficial effects of their own with regard to glucose homeostasis and serum lipid concentrations.

The lipid profile is a group of tests often ordered to determine the risk of coronary heart disease and other dietary lipid related disorder. They have proved to be good indicators to predict whether an individual is likely to have a heart attack or stroke caused by occlusion of blood vessels or hardening of the arteries (atherosclerosis). The lipid profile typically includes:

- Total cholesterol
- High density lipoprotein cholesterol (HDL-C) — often called good cholesterol
- Low density lipoprotein cholesterol (LDL-C) — often called bad cholesterol
- Triglycerides (TG)

An extended profile may also include:

- Very low density lipoprotein cholesterol (VLDL-C)
- Non-HDL-C.

It was recommended by American Society of Cardiologists that healthy adults with no other risk factors for heart disease be tested with a fasting lipid profile once every five years (Gannon et al, 2005). Individuals may also be screened using only a cholesterol test and not a full lipid profile. However, in the face of high cholesterol value, there may be the need to have follow-up testing with a lipid profile (Gannon et al 2005).

In the presence of other risk factors or past history of high cholesterol level in the individual, regular lipid profile test is necessarily advocated. For children and adolescents at low risk, lipid profile test is usually not routinely ordered. However, screening with a lipid profile is recommended for children and youths who are at an increased risk of developing heart disease as adults. Some of the risk factors are similar to those in adults and include a family history of heart disease or health problems such as diabetes, high blood pressure (hypertension), or being overweight. High-risk children should have their first lipid profile between 2 and 10 years old, according to the American Academy of Pediatrics. Children younger than 2 years old are too young to be tested (Gannon et al, 2003).

A total cholesterol reading can be used to assess an individual's risk for heart disease; however, it should not be relied upon as the only indicator. The individual components that make up total cholesterol reading – LDL, HDL, and VLDL – are also important in measuring risk (Gannon et al, 2003). For instance, one's total cholesterol may be high, but this may be due to very high good (HDL) cholesterol levels – which can actually help prevent heart disease. Therefore, while a high total cholesterol level may help give an indication that there is a problem with cholesterol levels, the components that make up total cholesterol should also be measured (Gannon et al, 2003).

Regular interval of lipid profile ordering may be necessary in evaluating success of lipid-lowering lifestyle changes such as diet and exercise or determining the effectiveness of drug therapy such as statins.

CHAPTER THREE
MATERIALS AND METHODS

3.1 LOCATIONS OF STUDY

The first phase of these experimentally controlled designed research studies was carried out at the department of Human Nutrition of the University of Ibadan and the Dietetic department of the University College Hospital (UCH) Ibadan, Nigeria while the second phase was carried out in the departments of Chemical Pathology (UCH), Physiology and Veterinary Anatomy/Pathology of the University of Ibadan, Ibadan Nigeria.

STUDY PHASE I: Effects of Food Processing Using Human Subjects

3.2 SUBJECT SELECTION

A non-probability sampling technique was used to select subjects who volunteered to participate in the study. The selection was made after the explanation of the protocol involved and the rationale behind the study.

3.3 SAMPLE SIZE

3.3.1 Diabetic volunteers

Ten type 2 diagnosed diabetic male patients managed and well-controlled only on oral hypoglycemic agents were recruited from the diabetic patients regularly attending the medical out-patient (MOP) diabetic clinic in the University College Hospital, Ibadan, Oyo state, Nigeria. The diabetic status and management profile of the selected volunteers were ascertained by checking their hospital case files. Good clinical history was taken to rule out exclusion criteria for diabetic subjects. Their fasting blood sugar (FBS) levels were determined using a portable glucometer (On Call Plus Blood Glucose Monitoring System, ACON Laboratories, Inc. San Diego, USA.). The study protocol did not prevent their daily routine prescribed medications.

They were allowed to take their drugs after the collection of the last two-hour postprandial blood samples for the assessment of the blood glucose concentrations.

3.3.2 Exclusion Criteria for Diabetic subjects

- (i) Female gender
- (ii) Use of insulin in the management and control of the diabetes
- (iii) Patients with type 1 diabetes
- (iv) Irregular attendance in the diabetic clinic
- (v) Poorly controlled patients with type 2 diabetes
- (vi) Diagnosis of diabetes mellitus less than one year

3.3.3 Healthy volunteers

Ten healthy male volunteers, age-matched hospital workers were recruited as control from the University college Hospital, Ibadan, Oyo State, Nigeria. Thorough clinical history and common baseline investigations including fasting blood sugar test, sphygmomanometer test and weight/height measurements were performed to establish that the volunteers met the inclusion criteria for healthy subjects.

3.3.4 Inclusion Criteria for Healthy Subjects

- (i) No history of hypertension in the subjects.
- (ii) No history of diabetes mellitus in the subjects.
- (iii) No family history of hypertension and diabetes in the subjects.
- (iv) No history of alcohol drinking on regular basis.
- (v) No history of smoking on regular basis.
- (vi) Values of common baseline investigations within normal range.

3.4 AGE

The age range of the volunteers was between 30 and 70 years for both study groups. The healthy volunteers were matched for age and sex with the diabetic volunteers

3.5 ETHICAL APPROVAL

The U.I/U.C.H Institutional Review Committee of the Institute for Advanced Medical Research and Training (IMRAT) approved the experimental protocol with the assigned number UI/EC/07/0092.

3.6 INFORMED CONSENT

The volunteers gave their written consent after the explanation of the procedure involved and the rationale behind the study with the evidence of signed informed consent-form.

3.7 TEST FOODS.

Five different processed yam meals with a known content of nutrients (determined in the laboratory) were tested while the reference (standard) food used in this study was 50g pure glucose as recommended by the World Health Organization/Food and Agriculture Organization (WHO/FAO) Expert Consultation Panel (1998). Each food was tested twice at six weeks interval to obtain two glycemic index values for a particular food being tested. The standard meal (pure glucose) dissolved in 300mls of water was served twice at six weeks interval like the tested foods to obtain two values for incremental area under the curve for standard meal (i.e. IAUCS₁ and IAUCS₂). Therefore, the study covered a period of seventy two weeks for the two sessions.

The composition (proximate), the calories and the amount of the tested food (expressed in grams) containing 50g of digestible carbohydrates were determined and calculated in the laboratory (Human Nutrition Analytical Laboratory, University of Ibadan, Nigeria.) by employing standard methods of food analysis and also by using the table of food composition for

use in Africa (1968) by Woot-Tsuen wu Leung of United State department of health, education and welfare, Public Health Service, Bethesda, Maryland, USA. Other sources of information used in the determination of the composition, calories and the amount of the tested food (expressed in grams) containing 50g of digestible carbohydrates include:

(i) Tables of representative values of foods commonly used in tropical countries by B.S Platt (1962)

(ii) The composition of foods by Paul and Southgate (1967); 4th Revised Edition. Amsterdam. NY Oxford, Elsevier. North Holland. Biomedical Press. NY 10017 USA.)

Dioscorea rotundata by dry weight of 100g is composed of 67% moisture, 80% starch, 7% protein, 7% mineral, 3% fiber and 1.7% lipids. 100g of the edible portion of the yam give 385kcal energy.

Raw weight of Dioscorea rotundata is composed of 80.8% moisture, 16.4% starch, 1.5% protein, 1.2% mineral, 0.6% fiber and 0.1% lipids. 100g of the edible portion of the yam give 71kcal food energy.

3.7.1 COMPOSITION (PROXIMATE) OF THE PROCESSED AND PREPARED YAM MEALS

A. Yam flour Paste meal [Amala, Yoruba];

Composition: Carbohydrates 84.12g, Protein 5.47g, Fat 0.00g; fiber 3.38g; Ash 0.74g; Moisture 77.08%. One serving of 280g of cooked Amala equal 50g of carbohydrate; Energy 375.71kcal.

B. Pounded Yam [Iyan, Yoruba];

Composition: Carbohydrates 84.26g, Protein 7.23g, Fat 0.36g; fiber 2.11; Ash 1.14g; Moisture 65.52%. One serving of 225g of pounded yam equal 50g of carbohydrate; Energy 381.19kcal.

C. Boiled Yam [Isu sise, Yoruba];

Composition: Carbohydrates 83.06g, Protein 7.66g, Fat 0.56g; fiber 2.41; Ash 0.82g; Moisture 61.01%. One serving of 180g of boiled yam [equal 50g of carbohydrate; Energy 380.71kcal.

D. Roasted Yam [Isu sisun, Yoruba];

Composition: Carbohydrates 80.40g, Protein 9.41g, Fat 0.59g; fiber 3.27g; Ash 0.51g; Moisture 59.39%. One serving of 135g of roasted yam equal 50g of carbohydrate; Energy 379.52kcal.

E. Fried Yam [Isu dindin, Yoruba];

Composition: Carbohydrates 79.07g, Protein 8.10g, Fat 2.20g; fiber 2.67g; Ash 1.57g; Moisture 54.33%. One serving of 60g of fried yam equal 50g of carbohydrate; Energy 383.81kcal.

3.7.2 STANDARD (REFERENCE) FOOD

The standard food was 50g Pure Glucose which was dissolved in 300mls of water before drinking. One serving equals 50g of digestible carbohydrates.

3.7.3 FOOD PREPARATION/PROCESSING

The yam species *Dioscorea rotundata* (White Yam) was purchased at the local Bodija market of the Ibadan North Local Govt. Oyo State, Nigeria with the help of an Agriculturist (IITA) and the Dietician (UCH). The five yam meals were processed and prepared professionally in expected quality and quantity by the Dietetic and Catering Department of the University College Hospital, Ibadan, Nigeria as follows:

3.7.3.1 BOILED YAM

The raw yam tuber was peeled to expose the fleshy part of the yam. This was later sliced, soaked and rinsed in clean potable water` prior to cooking on fire. The cooking lasted for 35minutes with 2000mls of water added until the yam softened. Salt was added to taste.

3.7.3.2 POUNDED YAM

The raw yam tuber was peeled to expose the fleshy part of the yam. This was later sliced, soaked and rinsed in clean potable water` prior to cooking on fire. The cooking lasted for 35minutes with 2500mls of water added until the yam softened. No salt was added to taste. The cooked yam was then pounded for 30minutes in a mortar with pestle to consistent, smooth and paste-like dough. 60mls of the water used for boiling the yam was added during the pounding.

3.7.3.3 FRIED YAM

The raw yam tuber was peeled to expose the fleshy part of the yam. This was later sliced, soaked and rinsed in clean potable water prior to frying in vegetable oil. Sliced yam was allowed to air-dry for about 5minutes and then shallow-fried for about 30minutes in a frying pan containing 300mls of unsaturated cholesterol free vegetable oil purchased at a local market. The frying was allowed to continue until a softened slightly brown fried yam was obtained.

3.7.3.4 AMALA (YAM FLOUR PASTE)

This was prepared from browned yam flour called 'Elubo Isu' in the western part of Nigeria. The yam flour is conventionally made by par-boiling yam chips at about 70-75°C till the chips are pliable and thereafter sun-dried for about 2-3 days depending on the atmospheric humidity. The dried chips are then ground into brown coloured yam flour ready for reconstitution in boiled water. Amala was prepared by adding the yam flour into boiling water and stirred continuously for about 15-25minutes until a thick brown or grey coloured smooth paste (Amala) was formed. Some quantity of water (about 125mls) was added to influence the desired constituency of the Amala.

3.7.3.5 ROASTED YAM

The raw yam tuber was peeled to expose the fleshy part of the yam. This was later soaked and rinsed in clean potable water` prior to roasting in an oven at about 100°C. The roasting lasted for

about 35-45 minutes until softened slightly brown roasted yam was formed ready for consumption.

Throughout the study, same type of soup (Ewedu with stew) was served which was prepared from local 'Ewedu' vegetable (*Corchorus Olithorus*) and tomato sauce. The food types (test meals) are eaten in Western Nigeria culture with a bowl of soup and so it was necessary to serve the meals with soup prepared in a standard and uniform way to avoid the introduction of possible variables that may affect the results. The boiled beef meat per serving (25g) was equal in weight throughout the study.

The portions of the food served were packed and wrapped properly. Each serving contained 50g of digestible carbohydrates. For the standard meal, the 50g pure glucose was dissolved in 300ml of water before drinking.

Table 2 shows the serving size (weights) of various processed/prepared yam meals containing 50g digestible carbohydrates.

Table 2
Serving Size (Weights) of processed/prepared yam meals containing 50g digestible carbohydrates

Yam meal Types	Weight (g)
Amala	280
Boiled	180
Pounded	225
Fried	60
Roasted	135

3.8 DETERMINATION OF FOOD COMPOSITION (PROXIMATES) AND ENERGY

Following the processing and preparation of the various yam (test) meals, portions of each (Table 2 above) containing 50g digestible carbohydrates as previously determined and calculated were taken to the laboratory for further analysis of their chemical (proximate) composition i.e. the carbohydrates, protein, fat, fiber, ash and moisture content prior to determination of their GI and the GR to each meal in the study subjects. Their calorific contents were also determined using Bomb Calorimeter (Cal 2k Eco Bomb Calorimeter, Digital Data Systems Ltd. Randburg, South Africa). The methodology of laboratory determination of the proximate and the energy according to the standard official methods of food analysis of the association of official analytical chemist (1990) and principles and techniques in food analysis by Dieter et al, (1980) and Joslyn (1970) was used in this study. The values of the proximate were determined after three (3) determinations.

3.8.1 Determination of Moisture Content Using Moisture Extraction Oven Method

PRINCIPLE

The lost in weight following drying of each sample at 100-135°C (usually 105°C) for 4 hours (or till weight was constant) in a hot (draft) air oven (Gallenkamp Hot Air Oven, Griffen and George Ltd, England) was reported as moisture content of the food sample.

APPARATUS

1. Well-ventilated moisture extraction oven
2. Desiccator with silica gel
3. Top loading and Analytical balances accurate to 0.1mg (Mettler Analytical and Top loading balances, H78AR and P121O respectively, Gallenkamp, UK.)
4. Aluminum or porcelain silica dishes
5. Spatulas

6. Labeling papers and magic markers

REAGENTS

No reagent required.

PROCEDURE

- Five porcelain dishes were washed and dried in the moisture oven and then put inside the desiccator to cool. Thereafter, each dish was weighed and recorded.
- 5g of macerated sample of each was put into the weighed dish and then weighed to take the weight of the dish and wet sample.
- Both sample and dish were dried in the moisture oven at 105°C for 4hrs until weight is constant.
- Sample was cooled in the desiccators and the dry weight of the sample and the dish was measured

CALCULATION

The percentage moisture content was calculated using the formula:

$$\% \text{ moisture content} = \frac{\text{weight of moisture}}{\text{Weight of sample}} \times 100$$

Or
$$\frac{\text{wet weight} - \text{dry weight}}{\text{Wet weight}} \times 100$$

$$\% \text{ Dry matter} = 100 - \% \text{ moisture content}$$

3.8.2 Determination of the Ash Component

PRINCIPLE

The inorganic minerals remaining in the residues after ignition of the food samples in a muffle furnace (Muffle furnace model SM9080, Surgifriend Medicals, Shanghai China) at temperature of 500-600 °C constitutes the ash in the food products.

APPARATUS

1. Muffle Furnace
2. Porcelain or silica crucibles
3. Desiccator
4. Analytic Balance
5. Hot Plate
6. Fume Cupboard

REAGENT

No reagent is required.

PROCEDURE

- Porcelain Crucible was washed and dried in the oven which was then
- Allowed to cool in a desiccator and weighed
- 5g of finely ground dried sample was put into an empty porcelain crucible which was previously ignited and weighed.
- The sample was then charred on a hot plate in the fume cupboard to drive most of the smoke.
- The sample was thereafter transferred into a preheated muffle furnace maintained at a temperature of 600°C for 2-8 hours.
- The crucible after 4hrs in the furnace was transferred directly to a desiccator, cooled and weighed to obtain the white ash.

CALCULATION

$$\% \text{ Ash} = \frac{(\text{weight of crucible + ash}) - (\text{weight of empty crucible})}{\text{Sample weight}} \times 100$$

Sample weight

3.8.3 Determination of Crude Protein Using Kjeldahl Nitrogen Method

PRINCIPLE

This method measured the crude protein content in the food samples by giving the amount of all the reduced nitrogen in the form of amines, ammonium compounds, urea, amino acids etc. The procedure involved digesting the sample with conc. H_2SO_4 and converting the nitrogen to ammonium nitrogen sulphate. The digestion was accelerated by adding a catalyst (selenium anhydrous copper sulphate salt) to increase the boiling point. The mixture was made alkaline by adding NaOH solution. The NH_3 produced was distilled into boric acid. The exact amount of NH_3 was determined by titrating with HCl. Protein values were obtained by multiplying the total nitrogen with conversion factor depending on the food source.

APPARATUS

1. Digester with in-built temperature controller (Electrothermal Heating Mantle, thermo Fisher Scientific. ESSEX, SS2 5PH, 419 Sulton Road, UK).
2. Digestion tubes
3. Heat resistant gloves
4. Automatic pipettes
5. Measuring cylinder
6. Complete titration units
7. Weighing papers
8. Kjeldahl flasks
9. Conical flasks
10. Tube mixer
11. Markham distillation apparatus
12. Glass beads

REAGENTS

1. conc. H₂SO₄
2. Kjeldahl catalyst (selenium anhydrous copper sulphate salt)
3. NaOH ((40-60%)
4. Boric acid 4% with bromocresol green/methyl red indicator solutions
5. 0.01HCl

PREPARATION

Boric acid indicator

This was prepared by mixing 24cc of 0.1% methylene blue in water and 16cc of 0.1% of methyl red in ethanol in 1000cc vol. flask. To the mixture, 0.5% of boric acid in water was added to make up to the mark. This was left for three days before use.

40-60% NaOH

40-60g NaOH pellets were dissolved in the 1000cc volumetric flask containing some quantity of water. Water further added to make up the mark.

0.01HCl

To 1000cc standard flask containing 600cc of water, 8.9cc of 35% of HCl was added and made up to the mark. This was later standardize by titration and diluted 10 times.

PROCEDURE

- 2g of sample was weighed and transferred into kjeldahl flask. 5g of anhydrous Na₂SO₄ was added followed by 1g of CuSO₄ and 1 tablet of kjeldahl catalyst.
- Into the mixture, 25c of conc. H₂SO₄ and 5 glass beads which prevent bumping during heating were introduced.

- In the fume cupboard, the mixture was heated initially gently then later strongly with occasional shaking until the mixture assume green colour.
- Thereafter the mixture was allowed to cool and distilled water used to wash dark particles at the neck of the flask down and then reheated as above until the green colour disappeared. This was then allowed to cool.
- After the cooling, the digest was washed severally into the 250cc volumetric flask. Distilled water added to make up the mark.
- Through Markham Distillation Apparatus (Griffen and George Ltd, UK), the digest was steamed for 15minutes.
- A 100cc of conical flask containing 5cc of boric acid indicator was placed under a condenser such that the tip of the condenser was under the liquid.
- With calibrated pipette, 5cc of the digest was introduced via a small aperture into the body of the Kjeldahl distillation flask. This was followed with distilled water and 5cc of 60% NaOH
- The solution was then steamed for 5-7 minutes to collect enough NH_4SO_4 as indicated by the green coloration of the boric acid indicator.
- Distillation continues for the next 15minutes and the solution in the receiving flask (Distillate) taken for titration with 0.01N of HCl acid

CALCULATION

$$\% \text{ total Nitrogen} = \frac{14.01 \times (\text{Sample titre} - \text{blank titre}) \times N}{10 \times \text{sample weight}}$$

N = Normality of the titrating acid

$$\% \text{ Protein (Crude)} = \text{Total Nitrogen} \times \text{Conversion Factor (6.25)}$$

3.8.4 Determination of crude fat using Soxhlet method

PRINCIPLE

This method measured crude fat which was extracted by petroleum ether based on the sparingly solubility of lipids in water and their considerable solubility in non-polar organic solvents which was evaporated subsequently to get the fat. The measured fat consists of all the soluble materials present in the food sample (test meals).

APPARATUS

1. Thimbles
2. Cotton wool
3. Soxhlet apparatus
4. Extraction flask (250mls)
5. Metallic cups
6. Filter paper

REAGENTS

- Petroleum ether

PROCEDURES

- 250mls of extraction flask was washed and dried in the oven at temperature of 105-110 °C for about 30 minutes. This was allowed to cool in the desiccator and then weighed.
- Soxhlet extractor with reflux condenser was fit up and water flow through the condenser started.
- 2g of dried sample of each test meal was placed on a filter paper which was folded and transferred into a fat-free extraction thimble plugged tightly with cotton wool. The thimble was placed in the extraction barrel.

- 300mls of petroleum ether was added into the extraction flask which was heated. The sample was allowed to reflux for about 6hrs.
- After extraction, the thimble was removed with care and petroleum ether collected in the top container of the set-up and drain into a container for re-use.
- The extraction flask was removed when almost free of petroleum ether and dried at temperature of 105-110 °C for 1 hour. Thereafter it was transferred into a desiccator, cooled and weighed to get the weight of flask and fat content.

CALCULATION

$$\% \text{ Fat} = \frac{(\text{weight of flask + fat}) - (\text{weight of empty flask})}{\text{Sample weight}} \times 100$$

3.8.5 Determination of the Crude Fiber Using Trichloroacetic Acid Method

PRINCIPLE

The indigestible matter or roughage in the food constitutes the crude fiber of any food. This method involved defatting the sample of each test meal followed by hydrolysis of protein and carbohydrate content of the sample using a mixture of acids.

APPARATUS

1. 600mls long beaker
2. Porcelain crucible/silica dish
3. Muffle furnace
4. Crude fiber refluxing apparatus
5. Filter paper (Whatman No 4; 15cm)

REAGENTS

- Distilled Water

- Methylated Spirit
- Petroleum Ether
- Trichloroacetic acid (TCA)

PREPARATION

TCA Digestion Reagent: This was prepared by mixing 500mls of glacial acetic acid with 450mls of water and 50mls conc. HNO_3 (nitric acid). Then, 20g of TCA dissolved in the mixture.

PROCEDURE

- Each sample of test meal was defatted with petroleum ether from which 1g of defatted sample was taken and introduced into 600mls beaker. 100mls of TCA reagent then added.
- The mixture was boiled and refluxed for 40 minutes beginning from the time of commencement of boiling.
- The flask was removed and cooled slightly under cold tap.
- The content was filtered through 15cm No 4 Whatman filter paper and the residue washed 6 times with hot distilled water and once with methylated spirit.
- The filter paper was opened in order to remove the residue with spatula and then transferred the fiber to a silica dish for drying overnight in an oven at 105 °C followed by cooling in the desiccator and weighing.
- Thereafter, ashing in a muffle furnace at 600 °C for 6hrs was carried out. Then cooled in a desiccator and weighed. Loss in weight during the incineration was equivalent to the amount of crude fiber.

CALCULATION

$$\% \text{ Crude Fiber} = \frac{\text{Difference in weighing}}{\text{Sample weight}} \times 100$$

3.8.6 Determination of Carbohydrate Content by Method of Difference

The carbohydrate content of the samples of each test meal was determined by difference i.e. by subtracting the % of ash, moisture, protein and fat from 100.

The calorific values were estimated by multiplying the % of carbohydrate and crude protein by 4 and that of fat by 9kCal/g. However, the gross energy was determined by the use of the Ballistic Bomb Calorimeter.

3.8.7 Energy Determination of Various Processed/Prepared Test (Yam) Meals Using Ballistic Bomb Calorimeter

PRINCIPLE

A known weight of a sample of each test meal was ignited electrically and burned in an excess of oxygen in the bomb. Maximum temperature rise of the bomb was measured using the thermocouple and galvanometer system. The calorific value of each sample was determined by comparing the temperature rise with another sample of known calorific value such as benzoic acid.

APPARATUS

1. Ballistic Bomb Calorimeter (Gallenkamp, UK)
2. Full Oxygen Cylinder
3. Mettler Analytical Balance
4. Benzoic Acid (Standard)
5. Clean spatulas
6. Cotton Thread

7. Iced or Tap water

8. Cloth

PREPARATION OF SAMPLE MATERIALS

- Each test meal was ground and mixed properly such that a sample taken represented the bulk of the meal.
- Prior to combustion, each sample was dried without volatilizing or destroying any of the combustible materials.
- The size of the sample taken was determined by the total heat released.

PROCEDURE

- Each food sample was dried and ground to a fine powder followed by thorough mixing of the powder.
- 0.5g of finely mixed sample above was measured for each test meal and placed in a clean crucible while 0.5g of benzoic acid (standard material) with a known calorific value was also taken.
- Each crucible was placed on the support pillar at the base of the bomb while a 5cm length of sewing cotton was placed between the coils of the firing wire (platinum) and the center of the crucible containing the sample. Same was also done for the standard material.
- The bomb body was then lowered and locked for the thread to engage firmly. Thereafter, a thermocouple was plugged into the hole at the top of the bomb body
- Oxygen cylinder valve was opened while the pressure was closed. Bomb valve was released while the front panel valve of the control box was opened (about ¼ turn)
- Cylinder Pressure was allowed to rise to 25 atm. in a time of about 20 sec. adequate for complete combustion of the sample.

- The temperature of combustion was made stable by means of the Galvanometer zero knob.
- The firing button was pressed and released and the maximum deflection of the galvanometer noted.
- Thereafter, the gas in the bomb was released by releasing the pressure valve. The body of the bomb then removed and rinsed two to three times and the dried.

CALCULATION

$$\text{Calorific value of sample} = \frac{\text{Heat released from sample (Kcal)}}{\text{Weight of sample}}$$

3.8.8 DETERMINATION OF THE GLYCEMIC INDEX AND GLYCEMIC RESPONSE

[1] Getting the basic data.

To determine the Glycemic Index (GI), measured portions (Table 2 above) of tested foods containing 50g of carbohydrates were eaten by each of the 20 volunteers after an overnight fast (12 hrs). Prior to ingestion of the test meal, blood samples were taken to determine their fasting blood sugar concentrations. Capillary blood samples used for blood glucose concentrations assessment were obtained through finger pricking using fine disposable lancets (to minimize pain) at 30 minutes interval over the next two hours after the meal (i.e. at times 0, 30, 60, 90 and 120 minutes; time 0 was just before food intake). The blood-glucose concentration of each volunteer was measured by means of a portable glucometer (On Call Plus Blood Glucose Monitoring System, ACON Laboratories, Inc. San Diego, USA.) employing the electronic clinometric method of blood-glucose analysis. At the end of each week test period, the blood-glucose values were recorded from the memory of the glucometer into a record paper for further

data analysis. The duration of the test period lasted for 14 weeks to obtain the glucose concentrations for all the tested foods and the reference food (pure glucose).

[2] Construction of Mean incremental Blood-glucose (Glycemic) response curves.

The averages of the respective blood-glucose concentrations after the meal were used to construct a mean incremental blood-glucose response curve for the two-hour period. The blood glucose concentration (mg/dl) was plotted on the ordinate (Y-) axis while the time duration (minutes) was plotted on the abscissa (X-) axis. For the purpose of statistical evaluation, all tests that were not complete were excluded.

[3] Calculations of the Individual GI values in every volunteer.

The incremental area under the curve (IAUC) was calculated for each meal in every volunteer separately (as the sum of the surface triangles and trapezoids between the blood-glucose curve and the horizontal baseline going parallel to X-axis from the beginning of the blood-glucose curve at time 0 to the point at time 120 minutes) to reflect the total rise in the blood-glucose concentration after eating the tested foods. The IAUCS for the reference (standard) food (i.e. 50g of pure glucose) was obtained similarly to the mean from the first independent IAUCS₁ and IAUCS₂ in the same volunteer.

In the IAUC/IAUCS calculations, all the blood-glucose values in the course of the test lower than the first value (at time 0) were equalized to the respective first value. In each volunteer, the GI (%) was calculated by dividing the IAUC for the tested food by the IAUCS for the standard food multiplied by 100. The following formula which is a modified version of Wolever et al was used to calculate the GI:

$$GI = \frac{IAUC}{1/2(IAUCS_1 + IAUCS_2)} \times 100 \text{ [%]}$$

IAUC = Incremental Area Under the blood glucose response Curve for the tested meal.

IAUCS = Incremental Area Under the blood glucose response Curve for the Standard meal.

The **glycemic response** (GR) was calculated as the incremental area under the curve above the fasting glucose concentration, (as the sum of the surface triangles and trapezoids between the blood-glucose curve and the horizontal baseline going parallel to X-axis from the beginning of the blood-glucose curve at time 0 to the point at time 120 minutes) ignoring any area below the fasting value, according to the equation given by Wolever et al (1991).

[4] Working out the average of GI's for tested food in each volunteer.

In each volunteer, each food item was tested twice so that two GI's were obtained and the average was calculated.

[5] Final calculation of the GI for each tested food.

The GI for each tested food was calculated as the mean from the respective average GI's of the ten volunteers (n = 10) in each group (i.e. the final GI for each tested food in diabetic and healthy subjects separately).

[6] The variability of GI for each tested food.

This was assessed as the standard deviation of the mean.

3.9.1 STUDY DESIGN

1. Every participating volunteer was instructed on how to keep to the principles of the study protocol without any prejudice to their social status:
 - To consume the tested food six weekly (on every Thursday) for breakfast at 8.00am according to the given schedule. No other food was allowed for breakfast.
 - To keep to the same extent of physical exercise during the whole 14 week test period.

- To consume no alcohol and not to smoke cigarette.
- 2. Each volunteer received 10 servings of tested foods and 2 servings of standard food.
- 3. Each volunteer kept a diary on food intake, exercise and results of blood-glucose values.

Both MS Excel and Statistical program SPSS v 17 were used to analyze the data.

3.9.2 STATISTICAL ANALYSIS

The Glycemic Index (GI) was expressed in percentage (%) as Mean \pm SEM (Standard Error of Mean) while other results were expressed as Mean \pm Standard Deviation [S.D]. Comparisons of the mean values between the two groups were performed by student t test using appropriate statistical methods and program of Microsoft Excel and SPSS v. 17. Linear Correlations of the GIs with the glycemic responses to various meals prepared by different processing methods between both groups were calculated with Pearson correlation test. P values of < 0.05 were considered statistically significant.

STUDY PHASE II:

Comparison of Low- and High-GI diets Using Animal Model (Rats)

3.1 LOCATION

This phase two study of the research work using experimental animals (rat) was carried out in the departments of Chemical Pathology (UCH), Physiology and Veterinary Anatomy/Pathology of the University of Ibadan, Ibadan Nigeria.

3.2 MATERIALS

The following materials/apparatus used in the Phase II of the research study (apart from those readily available in the laboratory of study) were purchased from reputable market and laboratory in Ibadan, Oyo State, Nigeria:

Modified Plastic Cages for keeping rats; Flat clay Tray for feeding rats; 1ml insulin syringes for injection and withdrawal of specimen; Metal Cannula for oral intubation of glucose into rats stomach, ink markers for tagging the rats for identification; On Call-Plus Glucometer with glucometer strips for determining the timed blood glucose concentrations and also for recording values obtained; Disposable Gloves for hygienic handling of rats and specimens; Cotton wool for mopping and sterilization with Methylated spirit; Dissecting set for rats dissection; Standard/Volumetric Flasks for constituting glucose and alloxan; Glucose D for oral glucose challenge; k₃ EDTA Specimen bottle for blood sample collection for lipid profile assay; Normal Saline infusion fluid for constitution of reagents; Alloxan reagent for induction of diabetes in rats; Digital weighing scales for weight measurement; Petri Dish as containers.

3.3 DIETS

Two of the five processed and prepared meals obtained from the first phase of this study research with known determined composition and glycemic index values were used for this Phase II study to compare their effects on the body and organ weights, glycaemic tolerance, pancreas histoarchitecture and lipid profile in alloxan-induced adult rats.

A. FRIED YAM (Low-GI Diet)

Average of the two values was taken to determine the classification of food. Since the average (30%) is less than 55% on GI Reference Range (WHO), fried yam is classified Low-GI food. Reason for choice among others with Low-GI was because of its preferred palatability to the rat as compared to others which was observed during preliminary study.

B. ROASTED YAM (High-GI Diet)

Average of the two values was taken to determine the classification of food. Since the average (79%) is greater than 70% on GI Reference Range, fried yam is classified Low-GI food.

Reason for choice among others with High-GI was because of its preferred palatability to the rat as compared to others which was observed during preliminary study.

C. STANDARD FEED (Oladokun Rat Feed)

The standard feed used in this study was purchased from one of the market dealers of Oladokun feeds Nigeria Limited at Mokola market, Ibadan, Oyo State.

Feed Ingredients:

- Maize
- Wheat middling
- Fish meal
- Groundnut cake
- Dried grains
- Brewer's yeast
- Bone meals
- Oyster shell
- Salt
- Antioxidant
- Antibiotics

Feed composition (% per 25kg feed):

- Protein 21% min.
- Fat 3.5% min.
- Fibre 6.0% max.
- Calcium 0.8%
- Phosphorus 0.8% (Total)

Table 3a: Proximate Composition of fried yam:

Test meals	Food energy (kcal)	Moisture (%)	Protein (g)	Fat (g)	Carbohydrate (g)	Fiber (g)	Ash (g)
Fried Yam	383.81	54.33	8.10	2.20	79.07	2.67	1.57

GI (Glycemic Index): 25% (healthy) or 36% (diabetics).

Table 3b: Proximate Composition of roasted yam:

Test meals	Food energy (kcal)	Moisture (%)	Protein (g)	Fat (g)	Carbohydrate (g)	Fiber (g)	Ash (g)
Roasted Yam	379.52	51.39	9.41	0.59	80.40	3.27	0.51

GI (Glycemic Index): 64% (healthy) or 93% (diabetes).

3.4 EXPERIMENTAL ANIMALS AND DIETS

Forty (48) male Albino Wistar rats weighing 130-150g (aged 2 months) were purchased from the disease-free stock of the animal house of the department of Veterinary Physiology, University of Ibadan, Ibadan, Oyo state, Nigeria. The rats were kept in polypropylene plastic cages and maintained at normal and standard laboratory conditions of temperature (28 ± 2 °C) and relative humidity ($46 \pm 6\%$) with 12-hour light-dark cycle and adequate ventilation. The rats were fed with standard rat feed continuously for four weeks until they weighed 250g and above which was suitable for the experimental study. While being fed, the rats were allowed to acclimatize to their environment.

The rats were weighed twice weekly to ensure that no rat outside the initial weight range of 250g-300g was used. This entry point weight range was chosen to ensure that the rats used were mature enough for the laboratory experiment at the end of 6 weeks period of study. The rats were initially fed with commercially available standard rat feed (Ladokun feeds Nig. Ltd.) purchased from a commercial branch depot in Ibadan, Oyo State with water ad libitum during the period of acclimatization. Thereafter, they were grouped and fed with the test feed (as mentioned above) and water ad libitum according to the experimental design for the next six (6) weeks prior to the laboratory investigations. The weights of the rats were recorded for the six weeks period of the study prior to laboratory investigations.

The Investigations using experimental animals use and care as found in the United States Guidelines (United States National Institutes for Health Publication No. 85-23, revised in 1985) were conducted in accordance with the internationally accepted principles for laboratory animals.

3.5 INDUCTION OF DIABETES

After 15 hour overnight fast, rats in groups A, B and C were injected intraperitoneally with freshly prepared alloxan monohydrate (Sigma chemicals, USA) dissolved in sterile normal

saline at a dose of 150 mg/kg body weight. Diabetes was confirmed in alloxan- induced animals seven days later by using glucometer to monitor the glucose level in the blood sample from the tail vein (Tail Snipping). Rats with Fasting Blood Glucose (FBG) level > 150mg/dl (WHO, 1985) were considered diabetic (hyperglycemic) and used for this study.

3.6 ANIMAL CATEGORIZATION AND EXPERIMENTAL DESIGN

The animals were allowed 28-day acclimatization period, after which they were randomly divided into six broad categories of eight (8) rats each:

GROUP A: Diabetic (hyperglycemic) rats fed on high-GI (Roasted Yam) diet – DRG group.

GROUP B: Diabetic (hyperglycemic) rats fed on Low-GI (Fried Yam) diet – DFG group.

GROUP C: Diabetic rats fed on normal standard rat diet (Control Diabetic) – DCG group.

GROUP D: Non-diabetic rats fed on high-GI (Roasted Yam) diet – NRG group.

GROUP E: Non diabetic rats fed on Low-GI (Fried Yam) diet – NFG group.

GROUP F: Non-diabetic rats fed on normal standard rat diet (Normal Control) – NCG group.

For the groups D, E and F category, the fasting blood glucose level was determined and confirmed by glucose oxidase method of Trinder (1969), using glucometer (On Call Plus Blood Glucose Monitoring System, ACON Laboratories, Inc. San Diego, USA.) by withdrawing blood from the tail end and testing. Those with blood glucose level of < 150 mg/dl were confirmed to be in the normoglycemic category and were used for the study.

3.7 BLOOD COLLECTION

The blood samples were collected from both the tail veins (OGTT) and posterior vena cava veins (Lipid Profile) of the rats. The blood samples from the tail veins were applied directly to the glucometer strips to determine the blood glucose concentrations while the blood samples from the posterior vena cava veins were collected in the k₃ EDTA (Ethylene Diamine Tetraacetic Acid) sample bottles for the analysis of the lipid profile.

3.8 BIOLOGICAL ASSAYS

3.8.1 Oral Glucose Tolerance Test (OGTT)

Animals in all groups were fasted 15 hours before the day of experiment with free access to water and were treated with an oral D-glucose load of 2 gm kg⁻¹ (dissolved in distilled water) administered by means of cannula, Blood samples were withdrawn from the cordal (tail) vein of each animal (tail snipping) to determine the fasting blood sugar concentration at time 0 minute and subsequently at intervals of 30mins, 60mins, 90 mins, 120 and 150 mins just after oral glucose administration of glucose challenge.

3.8.2 Lipid profile test (LPT)

Blood samples from the Posterior Vena Cava vein were collected and transferred into the k₃ EDTA (Ethylene Diamine Tetraacetic Acid) sample bottles. Samples were centrifuged at 3000 revolutions to obtain the plasma fractions which was kept in a refrigerator (at -70⁰C) until used and the sera obtained were used for the biochemical assay of the lipid profile using a dry-chemical auto analyzer (AU-5200, OLYMPUS). Total Cholesterol (TC), HDL-cholesterol and Triacylglycerol (TAG) were quantified using enzymatic kits (Randox Laboratories, San Francisco, USA) methods according to the manufacturer's instructions. LDL-cholesterol was calculated by Friedwald formula (Friedwald et al, 1972). The lipid profile was conducted at the beginning and at the end of the study for comparison.

3.9.1 EXTRACTION OF ORGANS AND ORGAN WEIGHTS

After 6 weeks of test feed consumption, animals in all groups were given light anaesthesia using Ethyl Ether in a glass dome and then dissected to extract some organs. Organ weights were measured and the pancreas of normal control, diabetic control and diabetic rats were histologically examined. Organ weights (liver, heart, kidney, lungs, spleen and testes) were measured and recorded as a percentage of final body weight together with the absolute values.

3.9.1.1 Histology of Pancreas: Histological examination was based on an earlier protocol (Humason, 1979). Slices of the pancreatic tissue were fixed in 10% formalin solution for 24 h. All samples were then dehydrated in graded ethanol series, cleared in toluene and embedded in paraffin wax; 5-6 μm sections were routinely stained with Harris hematoxylin and eosins stains (Sigma-Aldrich) and were assessed under light microscope (Nikon Eclipse E400).

3.9.2 STATISTICAL ANALYSIS

Data was analyzed using appropriate statistical methods and program of Microsoft Excel and SPSS v. 17. Results (all mean values) are expressed as group Mean \pm SEM (Standard Error of Mean). Comparisons between groups and the significant difference between the control and the treated groups were analyzed using one way analysis of variance (ANOVA) followed by Duncan's multiple range tests. P values of < 0.05 were considered statistically significant.

CHAPTER FOUR

RESULTS

STUDY PHASE I: EFFECT OF PROCESSING

DIABETIC GROUP

4.1.1 ANTHROPOMETRIC CHARACTERISTICS OF THE DIABETIC SUBJECTS

Ten type 2 (NIDDM) diabetic male patients volunteered to participate fully in both sessions of this study. Table 4 shows the anthropometric characteristics of the diabetic subjects. The average age of the subjects at the time of the study was 63.4 ± 9.2 years (range 43-70 years). Generally, the diabetic were older than the control healthy subjects even though they were age-matched. The average duration of diabetes was 8.4 ± 8.2 years (range 1-23 years). The mean body weight was 73.1 ± 14.6 kg (range 47-103kg) while the mean body mass index (BMI) was 25.98 ± 4.04 kg/m² (range 18.83-33.25kg/m²). This was comparable with the control group with a significant statistical difference. All the patients had good glycemic control at entry point as evidenced by their fasting and 2 hours postprandial blood sugar levels at the onset of the study. Five (50%) of the diabetics were Pre-Obese ($25-29.9$ kg/m²) while 1(10%) was obese. The remaining 40% fell within normal range of BMI ($18.5-24.9$ kg/m²). Current therapeutic regimen of the subjects included modified diet and oral hypoglycemic agents except one (10%) subject who was managed only on diet alone. 80% of the diabetic were also hypertensive and were managed on oral hypotensive combination chemotherapy with moderate partial exercise. The occupations of the subjects varied widely as shown by the table of clinical characteristics below - (Table 4).

4.1.2 GLYCEMIC RESPONSE CURVES TO TEST MEALS AND THEIR CORRESPONDING INCREMENTAL AREAS IN DIABETIC SUBJECTS

Tables 5 and 6 respectively show the incremental areas and the peak blood glucose concentrations of the glycemic response curves to various processed yam meals in diabetic subjects after consumption of 50g carbohydrates while figures 5 - 9 show the individual meal glycemic response curve pattern. A collective representative picture of the glycemic response curves pattern to all meal tested is shown by figure 10. Figure 11 shows the diagrammatic representation of the incremental areas of the standard and test meals.

The mean incremental area under the glycemic response curve following 50g pure glucose (Standard or reference meal) consumption occupied the largest area (8691 mg/dl.min) when compared with the five other test meals. The mean incremental area for the pure glucose was 4-65% higher than the rest. These differences were statistically significant ($p < 0.05$). Next to pure glucose incremental area was the Roasted yam (8332mg/dl.min) while Fried yam has the least incremental area (3082.5 mg/dl.min) - figure 11. The differences in the incremental areas of the test meals were also statistically significant ($p < 0.05$).

Fried yam has the flattest glycemic response curve pattern while the Roasted yam displayed a robust glycemic response pattern. (Figure 10)

At 60 minutes postprandial, Roasted yam glycemic response curve peaked with the highest mean blood glucose concentration value (211.95mg/dl) while Fried yam has the least mean peak blood glucose concentration (154.45mg/dl). Table 6

4.1.3 GLYCEMIC INDEX (GI) AND THE EFFECT OF METHOD OF PROCESSING /PREPARATION ON TEST DIETS (DIABETIC SUBJECTS)

Table 7 and figure 12 respectively show the tabular and diagrammatical representations of the glycemic indices (GIs) of the Test and the Standard meals expressed as Mean \pm SEM. The differences in their GIs which were statistically significant ($P < 0.05$) when compared between them revealed the impact of various processing methods on the GIs as quantitatively indicated by

their various different incremental areas under the glycemic response curves relative to that of the standard meal.

Roasted yam has the highest glycemic index of $93.34 \pm 4.04\%$ followed (in descending order) by boiled yam, pounded yam, amala and fried yam ($36.16 \pm 2.71\%$) which has the lowest GI.

4.1.4 CORRELATION OF GI IN DIABETIC AND NON-DIABETIC SUBJECTS

Table 8 shows very strong positive correlation between the GI and the glycemic responses to different method of processing and preparation in all foods tested while 60% of which are statistically significant (Boiled Yam ($p=0.000$); Fried Yam ($p=0.009$) and Roasted Yam $p=0.015$).

4.1.5 VARIABILITY OF THE GLYCEMIC INDEX (GI) IN DIABETIC SUBJECTS

There were substantial variations of the GIs of the test meals as shown by figure 13. Table 8 shows that wide variations in the GI of each meal were manifest in the correlation between the GI of the food types in patients with diabetes and healthy subjects.

TABLE 4**ANTHROPOMETRIC CHARACTERISTICS OF DIABETIC SUBJECTS**

Subject Numbers	Ages (Yrs)	Weights (Kg)	Heights (M)	BMI (kg/m²)	FBS (Mg/dl)	SBP (MmHg)	DBP (MmHg)	DOI (Yrs)	OCCUPATIONS
D1	70	75	1.68	26.57	75	110	70	10	Retired Police Officer
D2	70	61	1.6	28.53	108	140	80	15	Muslim Priest
D3	53	76	1.74	25.1	96	140	70	2	Travel Agency
D4	43	72	1.78	22.72	101	120	80	1	Engineering
D5	58	47	1.58	18.83	122	130	90	2	Teaching
D6	70	72	1.61	27.78	117	140	80	5	Banking (retired)
D7	70	85	1.7	29.41	120	140	70	23	Banking (retired)
D8	66	68	1.69	23.81	113	150	80	20	Retired Police Officer
D9	69	103	1.76	33.25	112	150	80	2	Retired Prison Officer
D10	65	72	1.74	23.78	105	140	70	4	Building Contractor
MEAN	63.4	73.10	1.69	25.98	106.90	136.00	77.00	8.40	
SD	9.24	14.55	0.07	4.04	13.91	12.65	6.75	8.18	

BMI-Body Mass Index, SBP- Systolic Blood Pressure, DBP- Diastolic Blood Pressure
DOI- Duration of Illness; FBS – Fasting Blood Sugar

TABLE 5

Incremental and Mean Incremental Areas under the Glycemic Response Curves to Test and Reference Diets in Diabetic Subjects

Subject numbers	Incremental Areas under the Glycemic Response Curves (IAUC) Mg/dl.min					
	Boiled Yam	Pounded Yam	Amala	Fried Yam	Roasted Yam	Glucose
D1	6855	1650	3315	825	8490	4350
D2	8200	5475	3990	3435	8475	8790
D3	6758.5	6405	4655	3945	4785	7800
D4	8655	5550	6045	2820	8265	8790
D5	7560	9615	5700	4470	9645	12150
D6	8782.5	8220	3450	5385	11190	8130
D7	7965	8910	5325	3360	9120	10740
D8	6030	5640	5160	2115	7675	8940
D9	8835	2655	4215	1410	5925	8430
D10	7257.5	4267.5	4350	3060	9750	8790
Mean	7689.85	5838.75	4620.5	3082.5	8332	8691
S.D	960.09	2583.92	924.52	1377.23	1867.27	2007.64

S.D – Standard Deviation

TABLE 6

Peak and Mean Peak Blood Glucose Concentrations of Glycemic Response Curves to Test and Reference Diets at 60 minutes Postprandial in Diabetic Subjects

Subject Numbers	Peak Blood Glucose values (Mg/dl)					
	Boiled Yam	Pounded Yam	Amala (yam flour Paste)	Fried yam	Roasted Yam	Glucose
D1	180.5	117	136	105.5	208.5	167.5
D2	213.5	150.5	149	158.5	228	207
D3	145	156	158.5	161.5	204	192
D4	213.5	154.5	177.5	135.5	214	240
D5	188.5	198	198.5	167.5	224.5	231
D6	211	181	190.5	194	183.5	249
D7	227	235	219	161	165	240
D8	202.5	181	194	158.5	266	229
D9	183.5	160	188	137.5	221.5	173
D10	191.5	150.5	181	165	204.5	209
Mean	195.65	168.35	179.2	154.45	211.95	213.75
S.D	23.37	32.20	24.93012	23.60	27.03	28.85

S.D – Standard Deviation

TABLE 7

Comparison of Glycemic Indices (GIs) of Various Processed Yam Meals between the Diabetic and Healthy Study Subjects

Food Items	Glycemic Indices (%) (Mean \pm SEM)		Significance Levels	
	Diabetic N=10	Healthy N=10	Equal Variance Assumed	Equal Variance not Assumed
Boiled Yam	88.65 \pm 3.11	52.32 \pm 6.46	➤ 0.0001	➤ 0.00022
Pounded Yam	70.75 \pm 4.93	53.74 \pm 8.82	0.11	0.108
Amala	50.09 \pm 4.66	36.12 \pm 7.05	0.12	0.165
Fried Yam	36.16 \pm 2.71	24.50 \pm 2.88	➤ 0.009	➤ 0.01
Roasted yam	93.34 \pm 4.04	64.00 \pm 10.17	➤ 0.015	➤ 0.012

➤ Significant P < 0.05

TABLE 8

Correlations of Glycemic Indices with Glycemic Responses to various yam meals prepared with different Processing Methods between Diabetic and Healthy Study Groups
(n = 20)

Food/Meal Types	Pearson Correlations (r)	Significance levels (p)
Boiled Yam	0.767	0.000
Pounded Yam	0.369	0.110
Amala (Yam Flour Paste)	0.363	0.116
Fried Yam	0.571	0.009
Roasted Yam	0.534	0.015

P < 0.05 Significant

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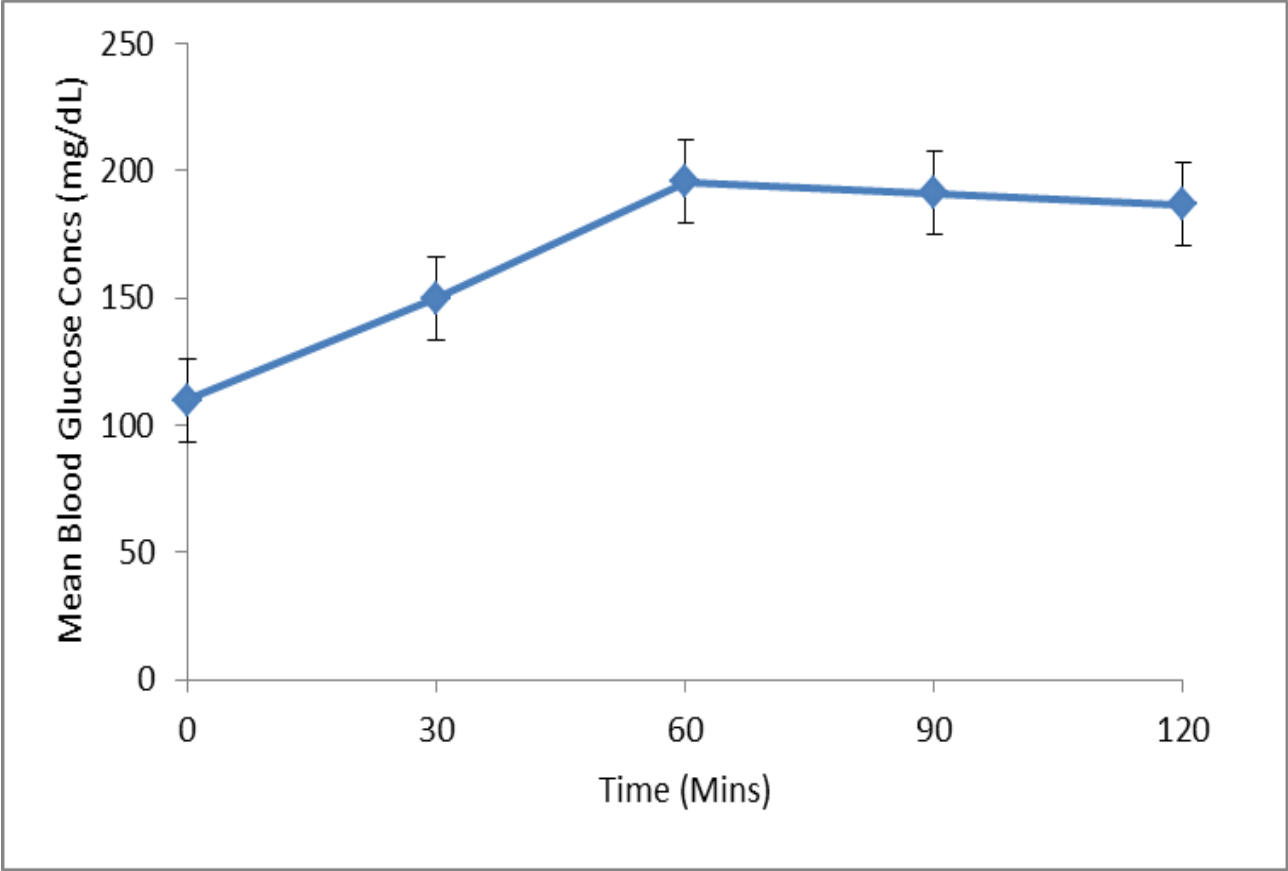


Figure 5: Glycemic response curve to boiled yam (Diabetic Subjects)

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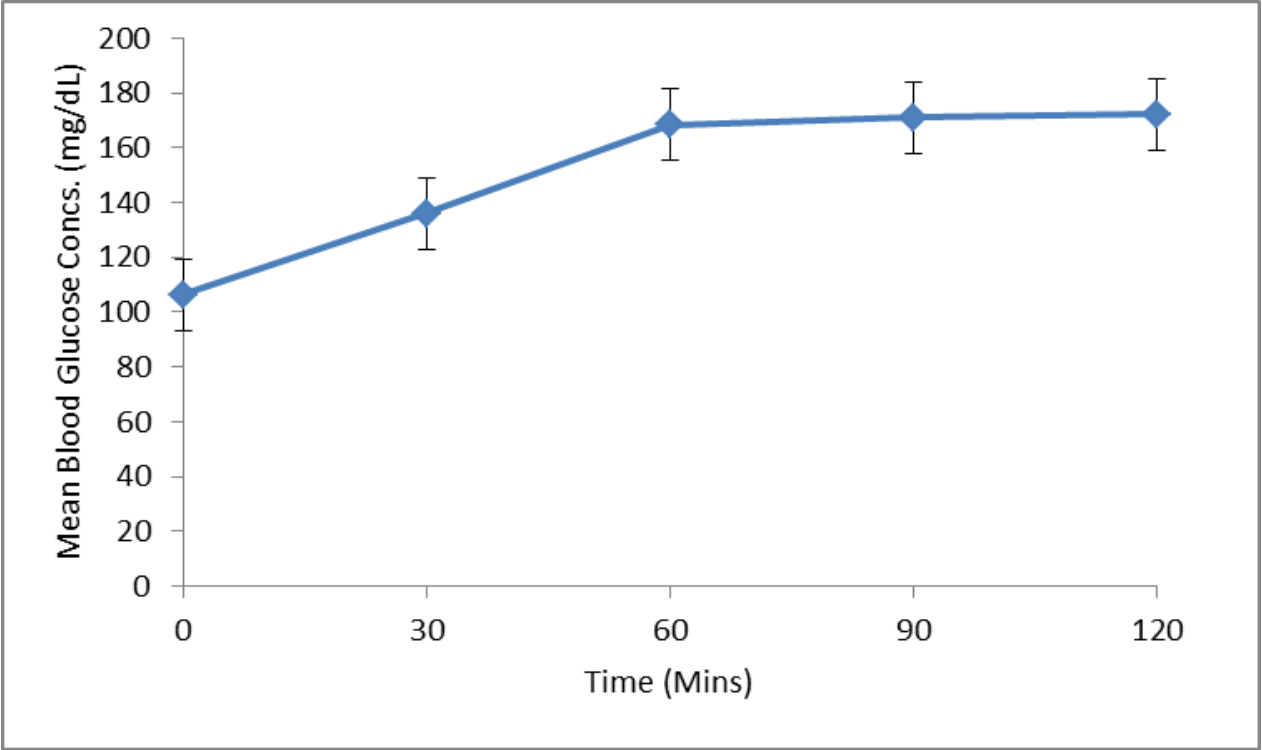


Figure 6: Glycemic response curve to pounded yam (Diabetic Subjects)

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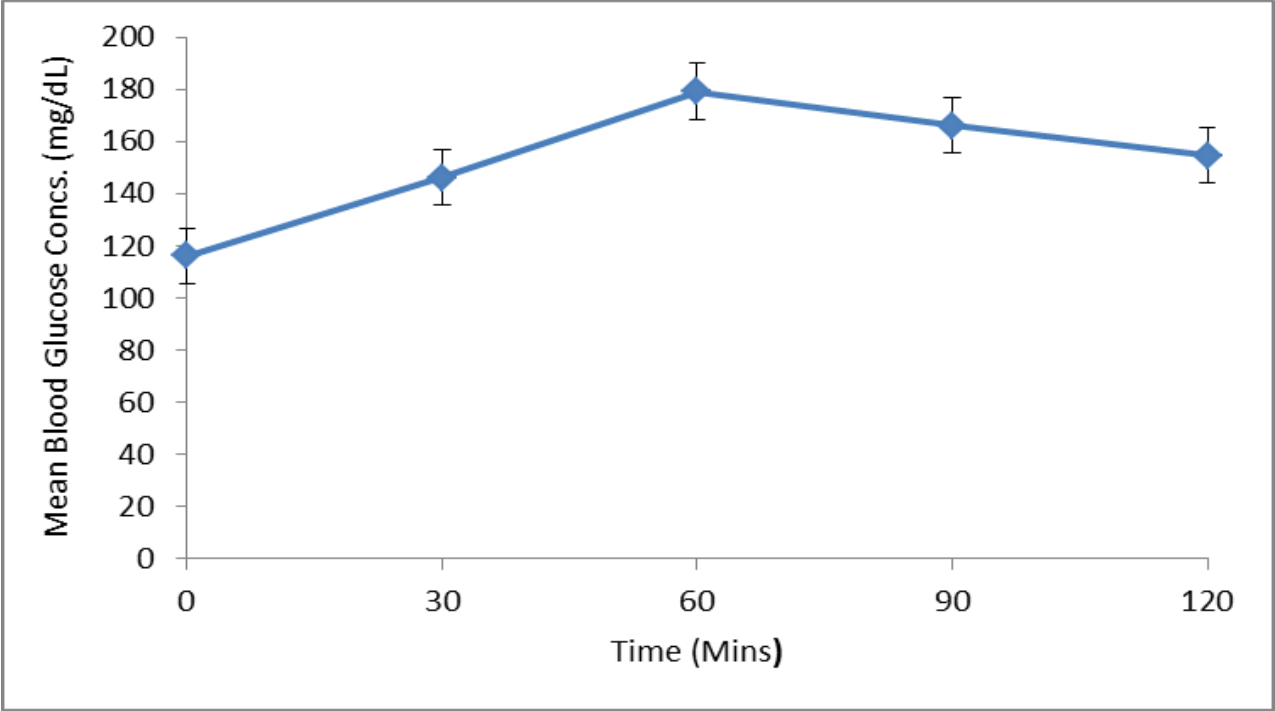


Figure 7: Glycemic response curve to Amala (Diabetic Subjects)

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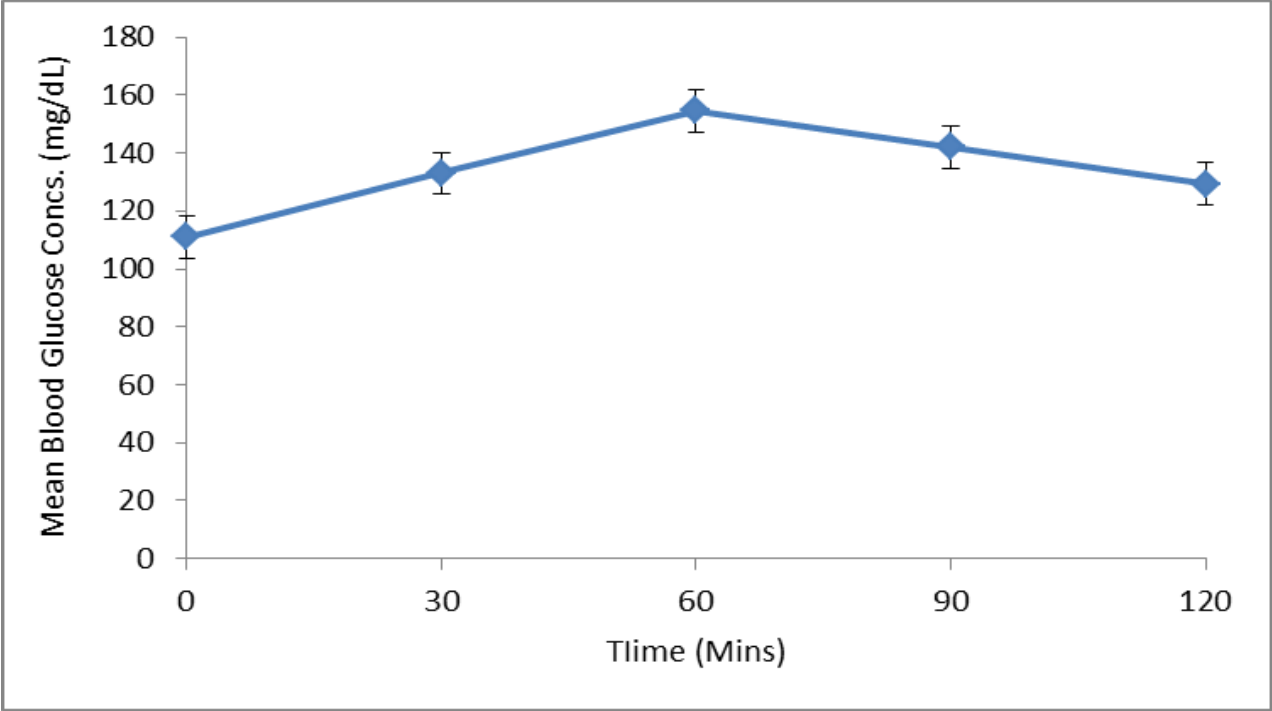


Figure 8: Glycemic response curve to fried yam (Diabetic Subjects)

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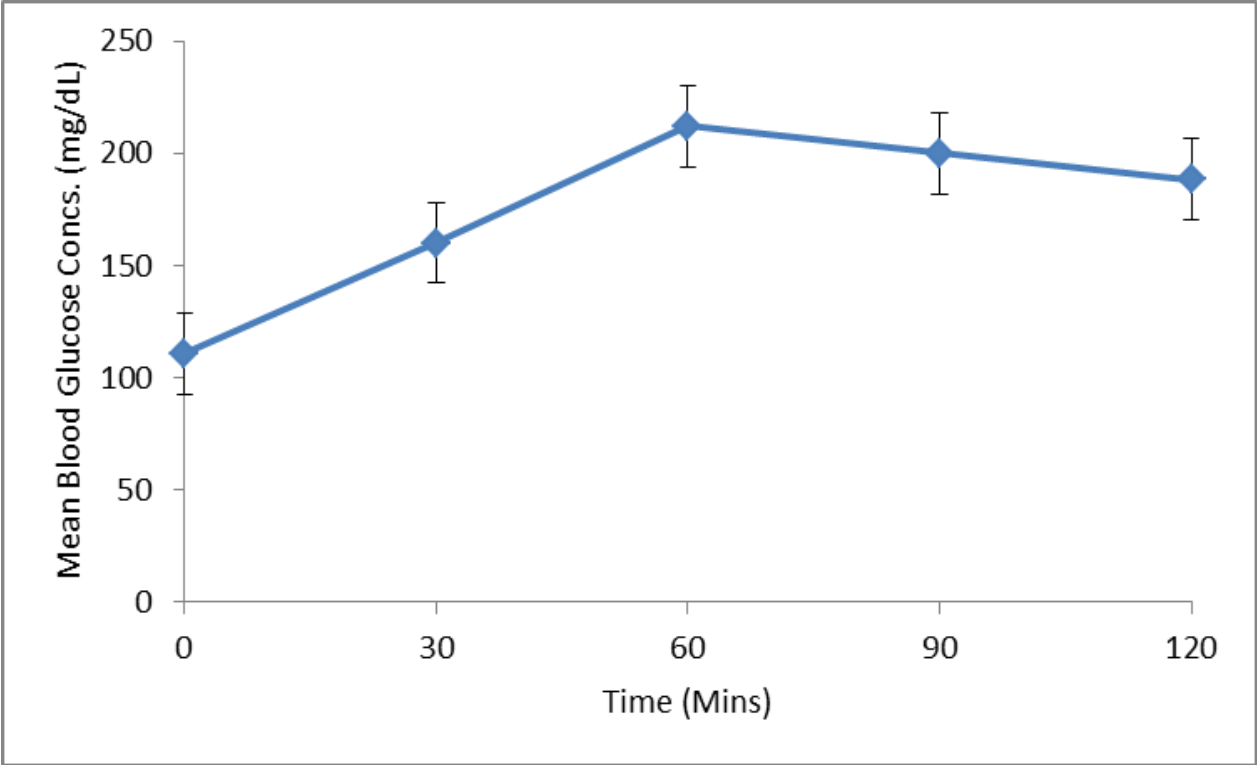


Figure 9: Glycemic response curve to roasted yam (Diabetic Subjects)

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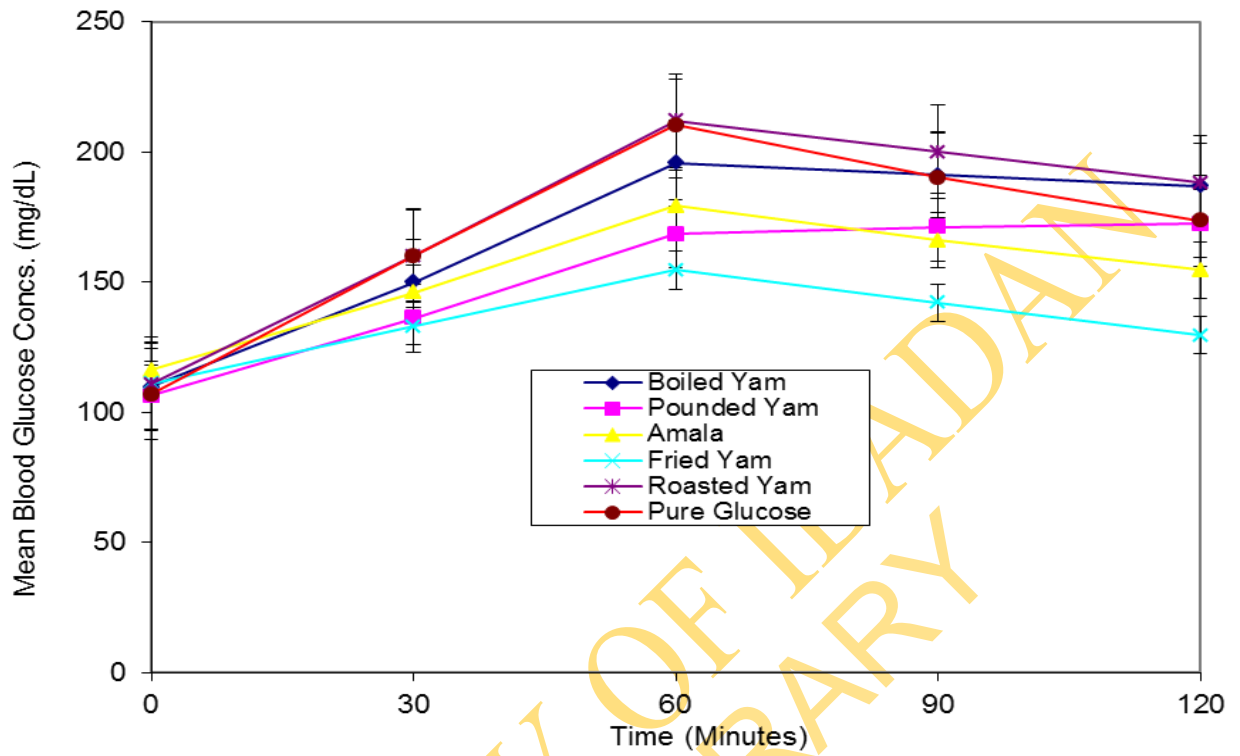
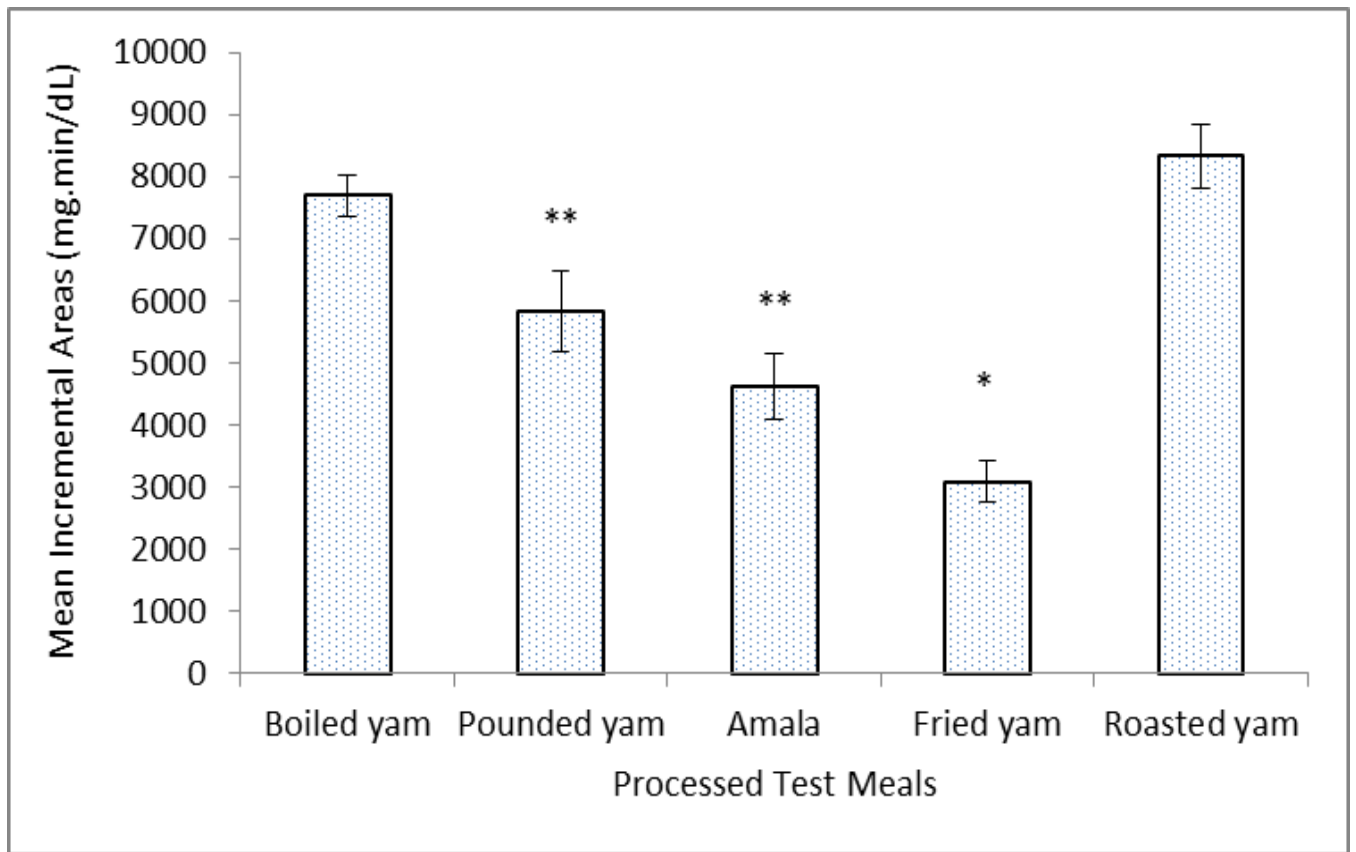


Figure 10: Glycemic response curves to reference (glucose) and various processed yam meals in diabetic subjects (after consumption of 50g of digestible carbohydrate)

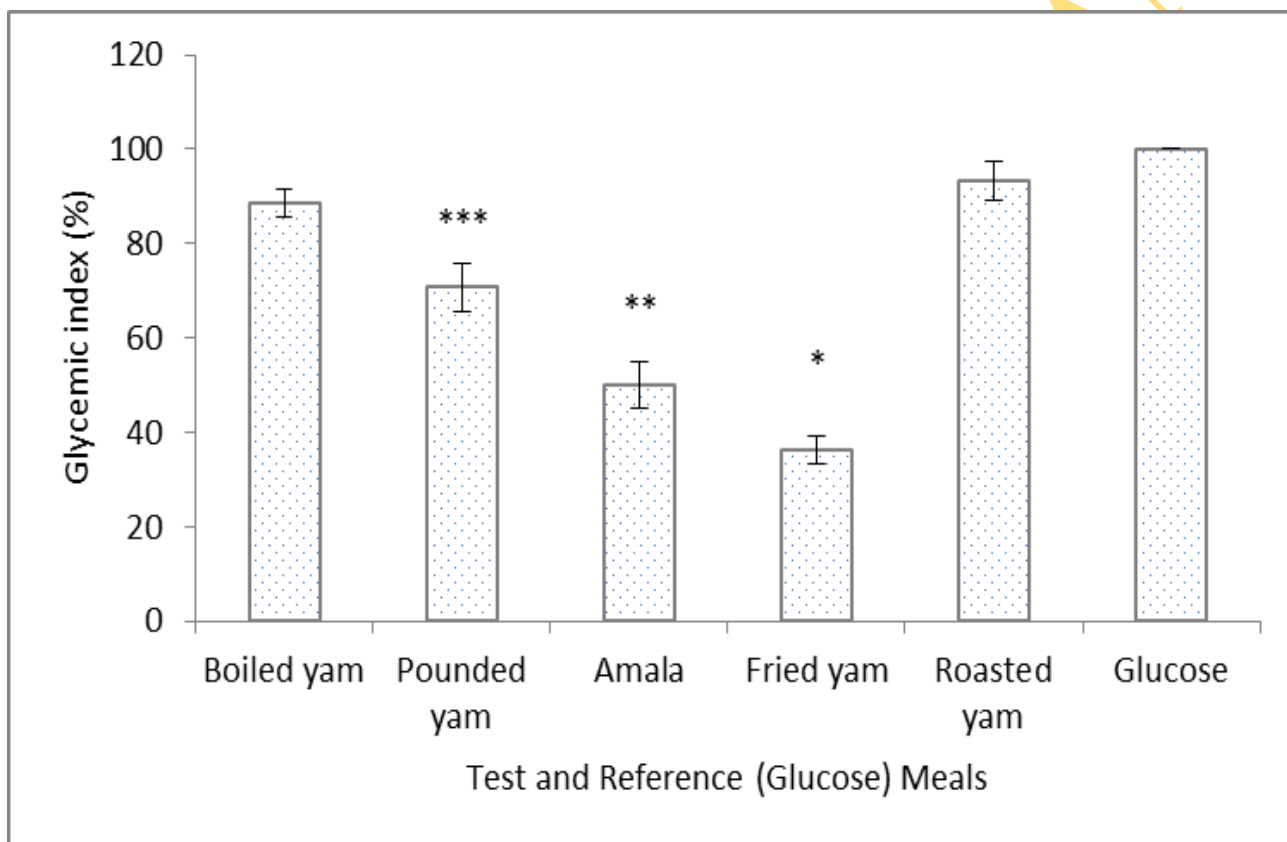


Values are in mean±SEM

*Significantly different from amala;pounded yam;fried yam;roasted yam

**Significantly different from boiled yam;roasted yam

Figure 11: Mean incremental areas under glycemic response curves to various processed test meals (diabetic subjects)



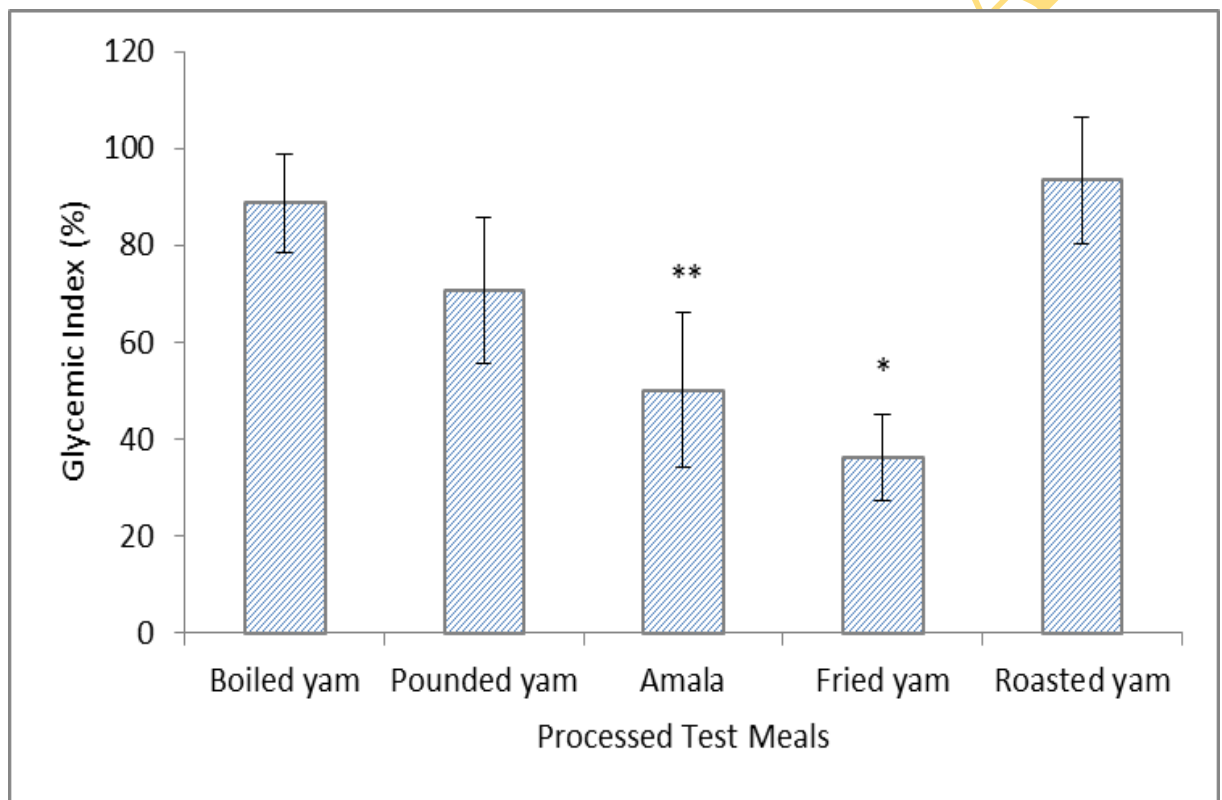
Values are expressed in mean±SEM.

*Significantly different from Amala;Pounded yam;Boiled yam;Roasted yam

**Significantly different from Pounded yam;Boiled yam;Roasted yam

*** Significantly different from Boiled yam;Roasted yam

Figure 12: Glycemic index (GI) of test and reference (glucose) meals in diabetic subjects.



Values are expressed in mean±SD

*significantly different from boiled yam;Pounded yam;Roasted yam

**significantly different from boiled yam; roasted yam

Figure 13: Variability of Glycemic index of processed test meals (Diabetic Subjects)

HEALTHY (NON-DIABETIC) CONTROL GROUP

4.2.1 ANTHROPOMETRIC CHARACTERISTICS OF THE CONTROL SUBJECTS

Ten healthy non-diabetic volunteers participated fully in both sessions of this study. Table 9 shows the anthropometric characteristics of the healthy subjects. The average age of the subjects at the time of the study was 46.5 ± 5.4 years (range 38-55 years). All satisfied the inclusion criteria for the study and were age-matched with the diabetic subjects. The mean body weight was 66.2 ± 8.4 kg (range 52-80 kg) while the mean body mass index (BMI) was 20.67 ± 2.2 kg/m² (range 18.72-26.12 kg/m²). This was comparable with the diabetic group with a significant statistical difference. All the patients had good glycemic control at entry point as evidenced by their fasting and 2 hours postprandial blood sugar levels at the onset of the study. 1(10%) of the healthy subjects was Pre-Obese ($25-29.9$ kg/m²) while others were within normal range of BMI ($18.5-24.9$ kg/m²). The occupations of the subjects varied widely as shown by the table of clinical characteristics below (Table 9).

4.2.2 GLYCEMIC RESPONSE CURVES TO TEST MEALS AND THEIR CORRESPONDING INCREMENTAL AREAS IN HEALTHY SUBJECTS

Tables 10 and 11 respectively show the incremental areas and the peak blood glucose concentrations of the glycemic response curves to various processed yam meals in healthy subjects after consumption of 50g carbohydrates while figures 14 - 18 show the individual meal glycemic response curve pattern. A collective representative picture of the glycemic response curves pattern to all meal tested is shown by figure 19. Figure 20 shows the diagrammatic representation of the incremental areas of the standard and test meals.

The mean incremental area under the glycemic response curve following 50g pure glucose (Standard or reference meal) consumption occupied the largest area (2434 mg/dl.min) when compared with the five other test meals. The mean incremental area for the pure glucose

was 30-77% higher than the rest. These differences were statistically significant. Next to the pure glucose incremental area was the Roasted yam (1863mg/dl.min) while the Fried yam has the least incremental area (3082.5 mg/dl.min) (Table 10). The differences between the incremental areas of the test meals were also statistically significant ($p < 0.05$).

Fried yam has the flattest glycemic response curve pattern while the Roasted yam displayed a robust glycemic response pattern (Figure 19).

At 60 minutes postprandial, Roasted yam glycemic response curve peaked with the highest mean blood glucose concentration value (115.95mg/dl) while Fried yam has the least mean peak blood glucose concentration (96.35mg/dl). Table 11.

4.2.3 GLYCEMIC INDEX (GI) AND THE EFFECT OF METHOD OF PROCESSING AND PREPARATION ON TEST DIETS (HEALTHY SUBJECTS)

Table 7 and figure 21 respectively show the tabular and diagrammatical representations of the glycemic indices (GIs) of the Test and the Standard meals expressed as mean \pm SEM. The differences in their GIs which were statistically significant ($P < 0.05$) when compared between them revealed the impact of various processing methods on the GIs as quantitatively indicated by their various different incremental areas under the glycemic response curves relative to that of the standard meal.

Roasted yam has the highest glycemic index of $64\pm 10.17\%$ followed (in descending order) by Pounded yam, Boiled yam, Amala and Fried yam ($24.5\pm 2.88\%$) which has the lowest GI.

4.2.4 CORRELATION OF GI IN NON-DIABETIC (HEALTHY) AND DIABETIC SUBJECTS

Table 8 above shows very strong positive correlation between the GI and the glycemic responses (GR) to the method of processing and preparation in all foods tested while 60% of

which were statistically significant (Boiled Yam ($p=0.0002$); Fried Yam ($p=0.009$) and Roasted Yam $p=0.015$).

4.2.5 VARIABILITY OF THE GLYCEMIC INDEX (GI) IN HEALTHY SUBJECTS

There were substantial variations of the GIs of the test meals as shown by figure 22. Table 8 shows that wide variations in the GI of each meal were manifest in the correlation between the GI of the food types in healthy subjects and diabetic patients

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TABLE 9**ANTHROPOMETRIC CHARACTERISTICS OF HEALTHY STUDY SUBJECTS**

Subjects Numbers	Ages (Yrs)	Weights (Kg)	Heights (M)	BMI (kg/m²)	FBS (Mg/dl)	SBP (MmHg)	DBP (MmHg)	OCCUPATIONS
C1	50	80	1.75	26.12	86	110	70	Engineering
C2	40	65	1.83	19.41	81	110	80	Social Worker
C3	53	52	1.65	19.1	93	120	70	Lab. Technology
C4	43	72	1.89	20.16	96	100	65	Dietetics
C5	48	59	1.77	18.83	94	120	70	Secretary
C6	45	58	1.76	18.72	97	110	80	Driving
C7	38	64	1.73	21.38	95	110	70	Dietetics
C8	45	67	1.74	22.13	90	120	80	Nursing
C9	55	71	1.89	19.88	91	120	80	Administration
C10	48	74	1.88	20.94	88	110	70	Record keeping
MEAN	46.50	66.20	1.79	20.67	91.10	113.00	73.50	
SD	5.40	8.40	0.08	2.23	5.00	6.75	5.80	

BMI-Body Mass Index SBP- Systolic Blood Pressure
 FBS – Fasting Blood Sugar

DBP- Diastolic Blood Pressure

TABLE 10

Incremental and Mean Incremental Areas under the Glycemic Response Curves to Test and Reference Diets in Healthy Study Subjects

Subject numbers	Incremental Areas under the Glycemic Response Curves (IAUC) Mg/dl.min					
	Boiled Yam	Pounded Yam	Amala	Fried Yam	Roasted Yam	Glucose
C1	1150	1200	1020	630	1200	3690
C2	1020	870	1530	750	1560	3060
C3	1230	480	1020	300	1200	1320
C4	840	1290	1530	1050	3990	3690
C5	1440	720	240	690	1260	2040
C6	2820	2010	600	630	1050	1200
C7	1440	540	480	180	2430	3065
C8	840	2370	750	1050	2760	3060
C9	1440	840	1170	750	1980	2045
C10	1950	1350	600	1290	1200	1170
Mean	1417	1167	894	732	1863	2434
S.D	594.78	620.72	435.84	337.99	948.63	1000.28

S.D – Standard Deviation

TABLE 11

Peak and Mean Peak Blood Glucose Concentrations of Glycemic Response Curves to Test and Reference Diets at 60 minutes Postprandial in Healthy Subjects

Subject Numbers	Peak Blood Glucose values (Mg/dl)					
	Boiled Yam	Pounded Yam	Amala (yam flour Paste)	Fried yam	Roasted Yam	Glucose
C1	121	121	109.5	102.5	119.5	156
C2	108	95	106	89.5	102	102
C3	118	116.5	110	90.5	121.5	152
C4	112	115.5	101	89	121.5	123
C5	126	116.5	102.5	95	106.5	93
C6	112.5	111	98	93	102	104
C7	115.5	114	113.5	107.5	121.5	122
C8	113.5	113.5	106	97.5	122.5	108
C9	112.5	116	97	109.5	115.5	154
C10	109.5	103	98	89.5	127	121
Mean	114.85	112.20	104.15	96.35	115.95	123.50
S.D	5.47	7.65	5.74	7.69	9.12	23.14

S.D – Standard Deviation

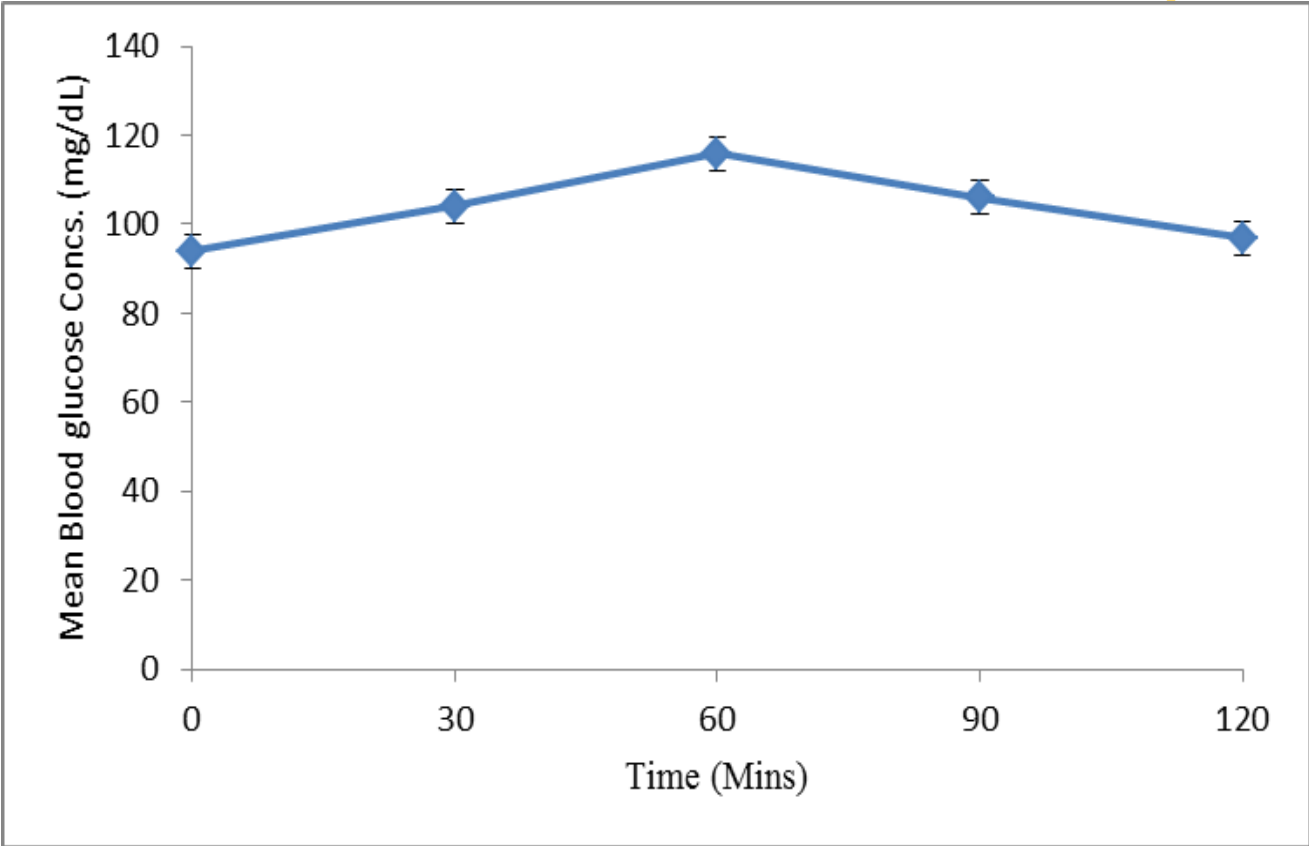


Figure 14: Glycemic response curve to boiled yam (Healthy Subjects)

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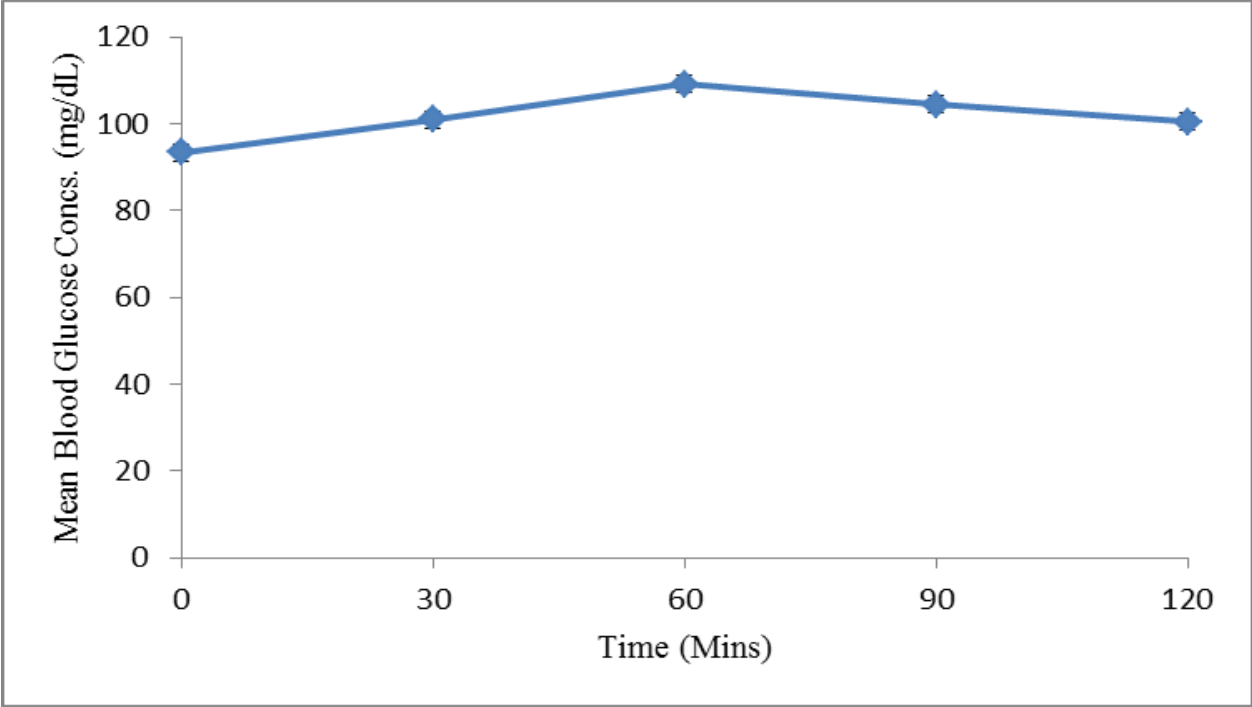


Figure 15: Glycemic response curve to pounded yam (Healthy Subjects)

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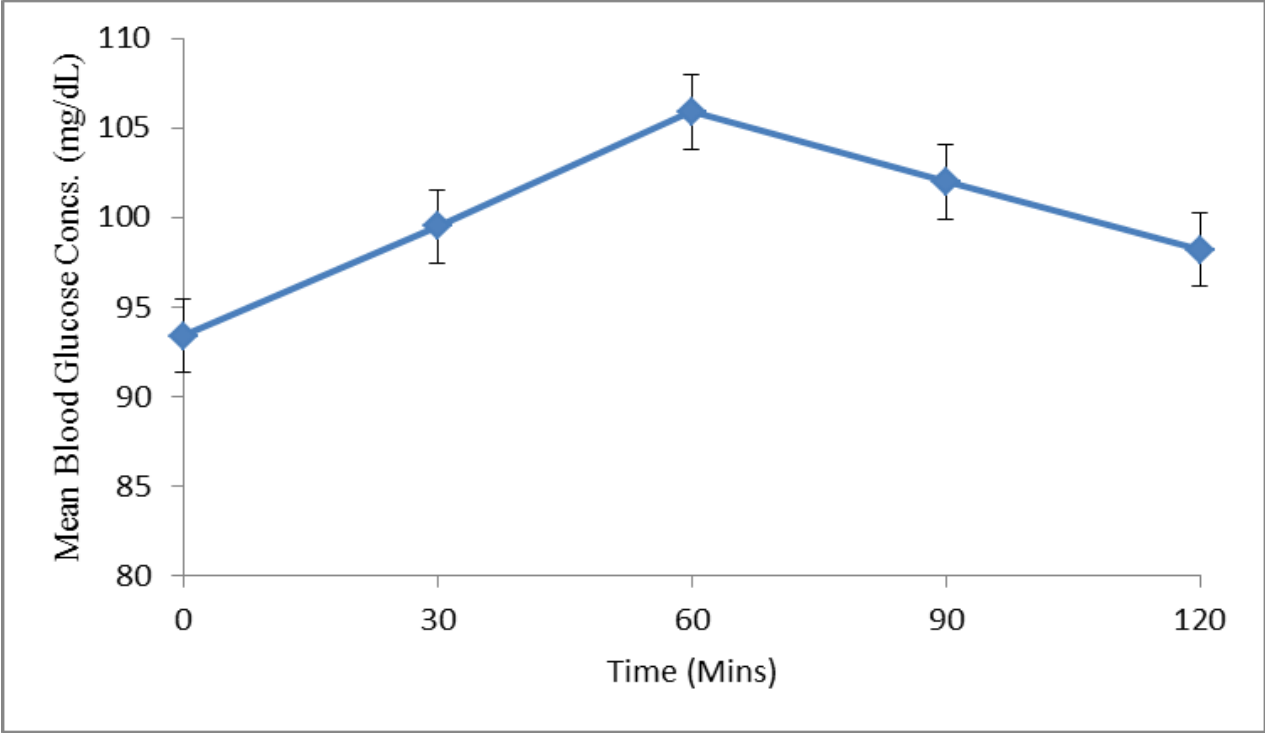


Figure 16: Glycemic response curve to Amala (Healthy Subjects)

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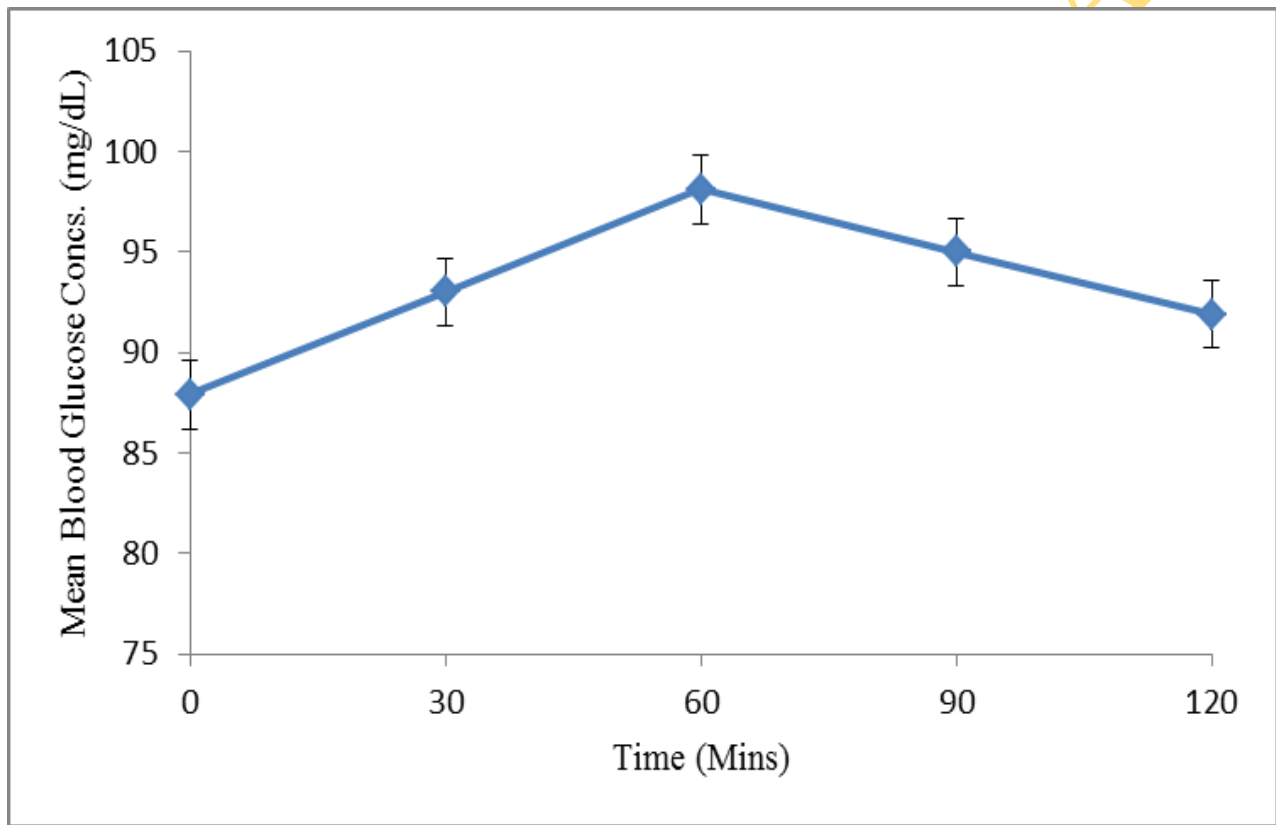


Figure 17: Glycemic response curve to fried yam (Healthy Subjects)

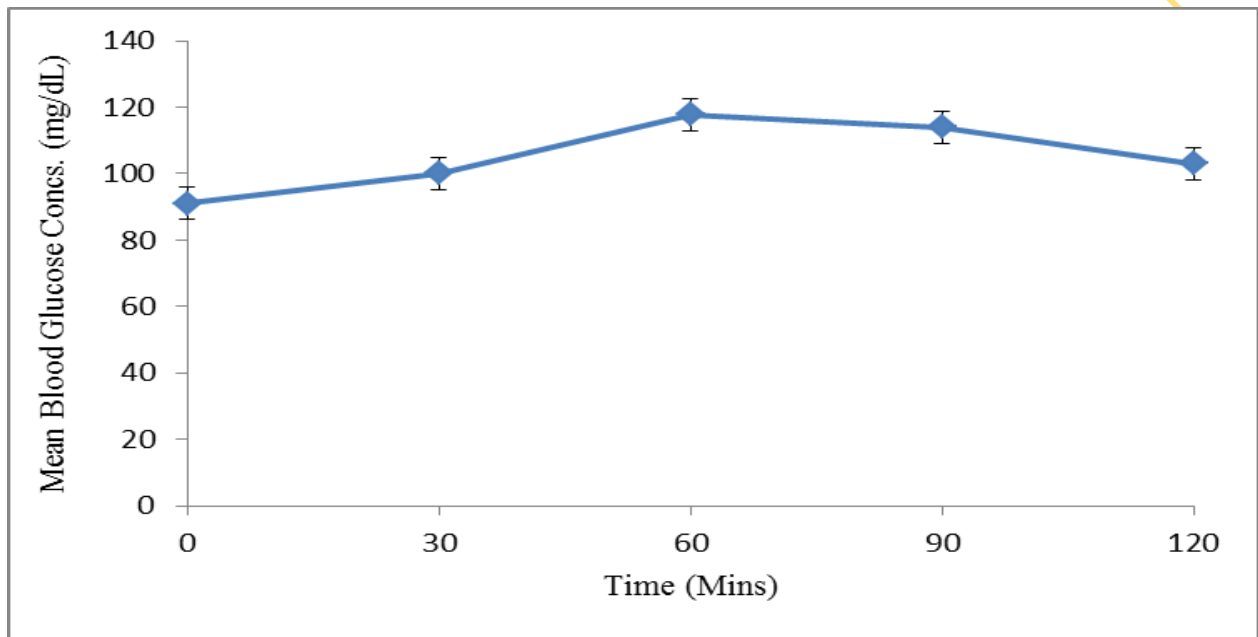


Figure 18: Glycemic response curve to roasted yam (Healthy Subjects)

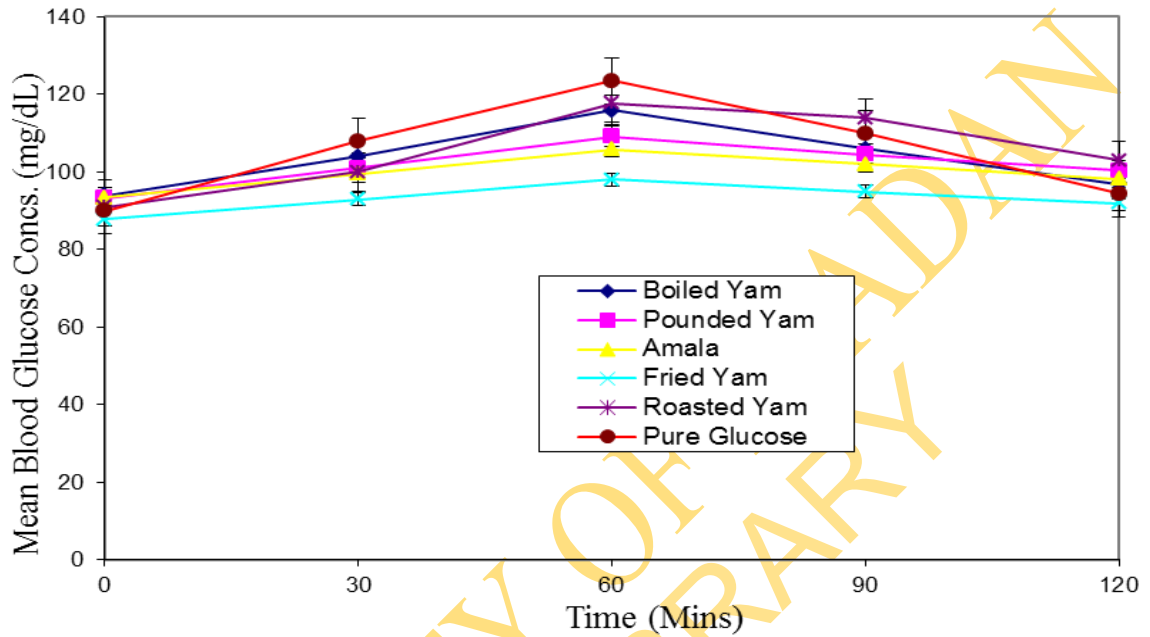
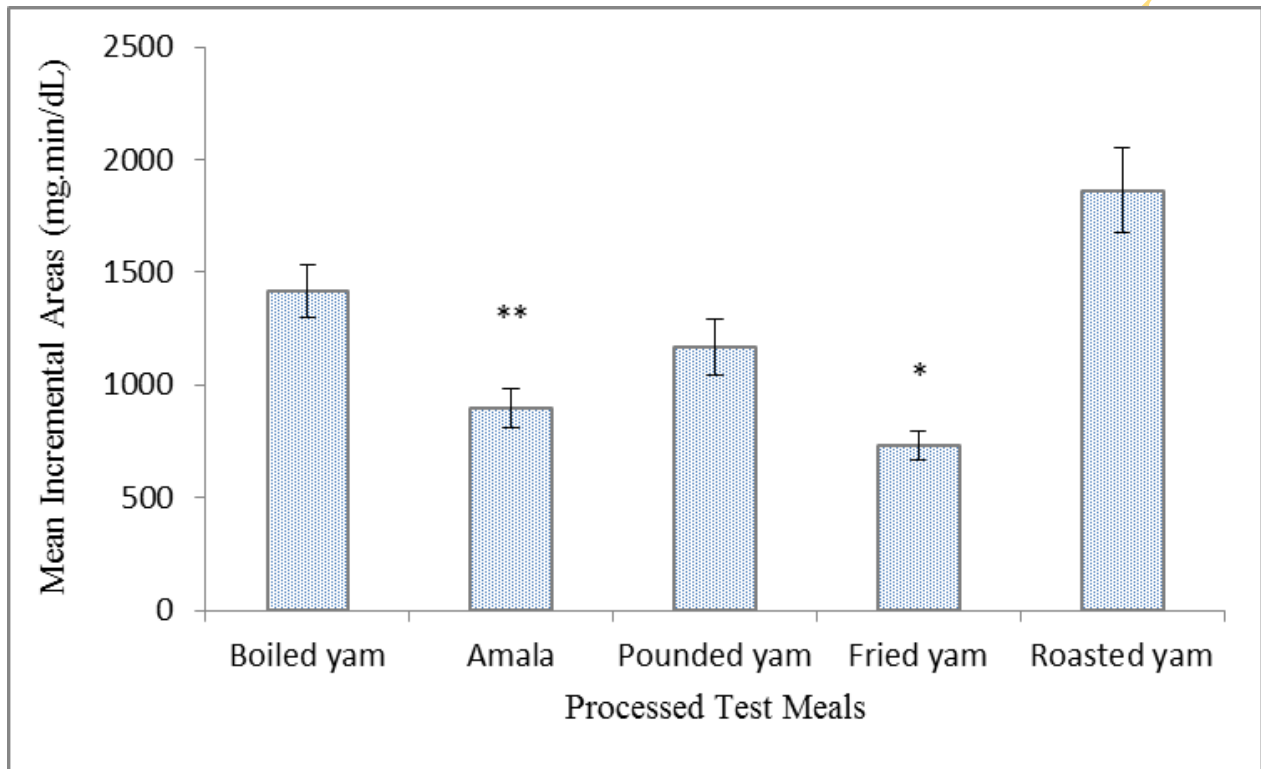


Figure 19: Glycemic response curves to reference (glucose) and various processed yam meals in healthy subjects (after consumption of 50g of digestible carbohydrate)

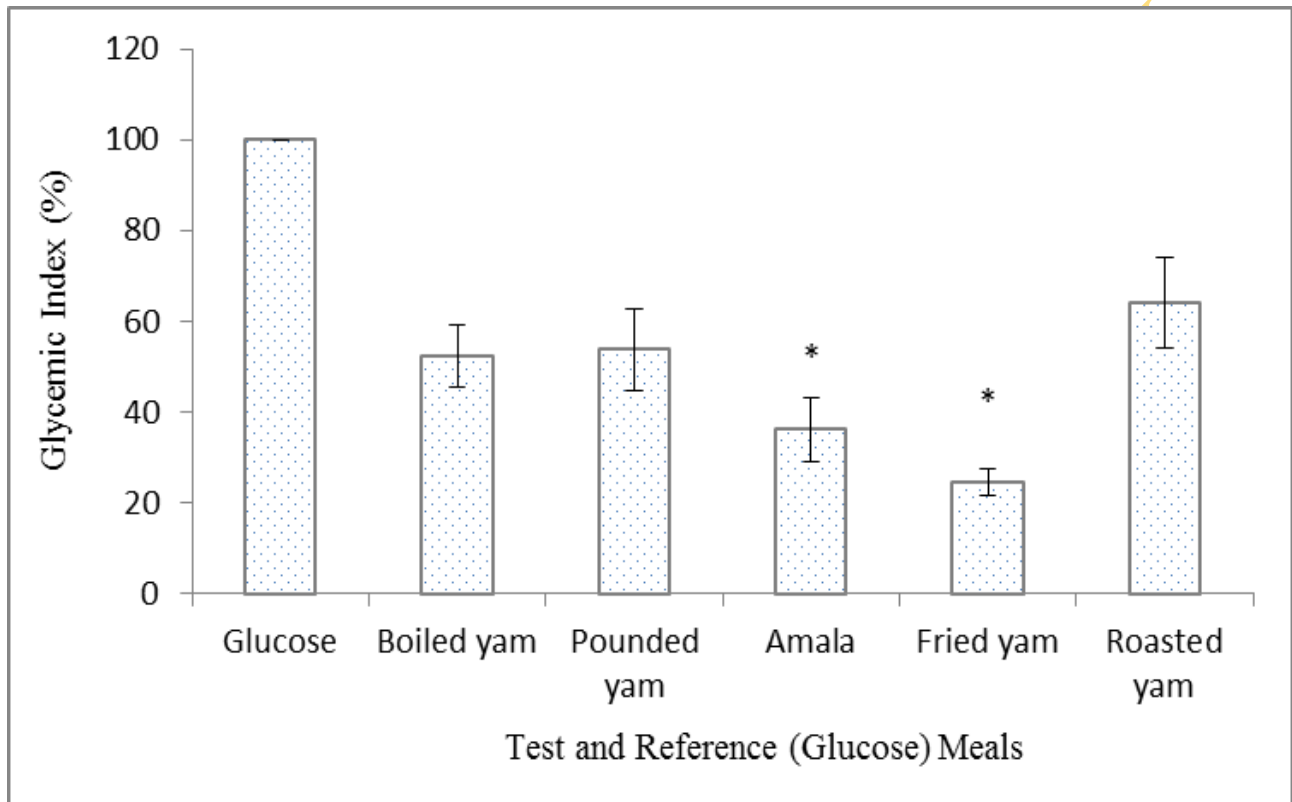


Values are in mean±SEM

*Significantly different from amala;pounded yam;fried yam; roasted yam

**Significantly different from boiled yam; pounded yam; roasted yam

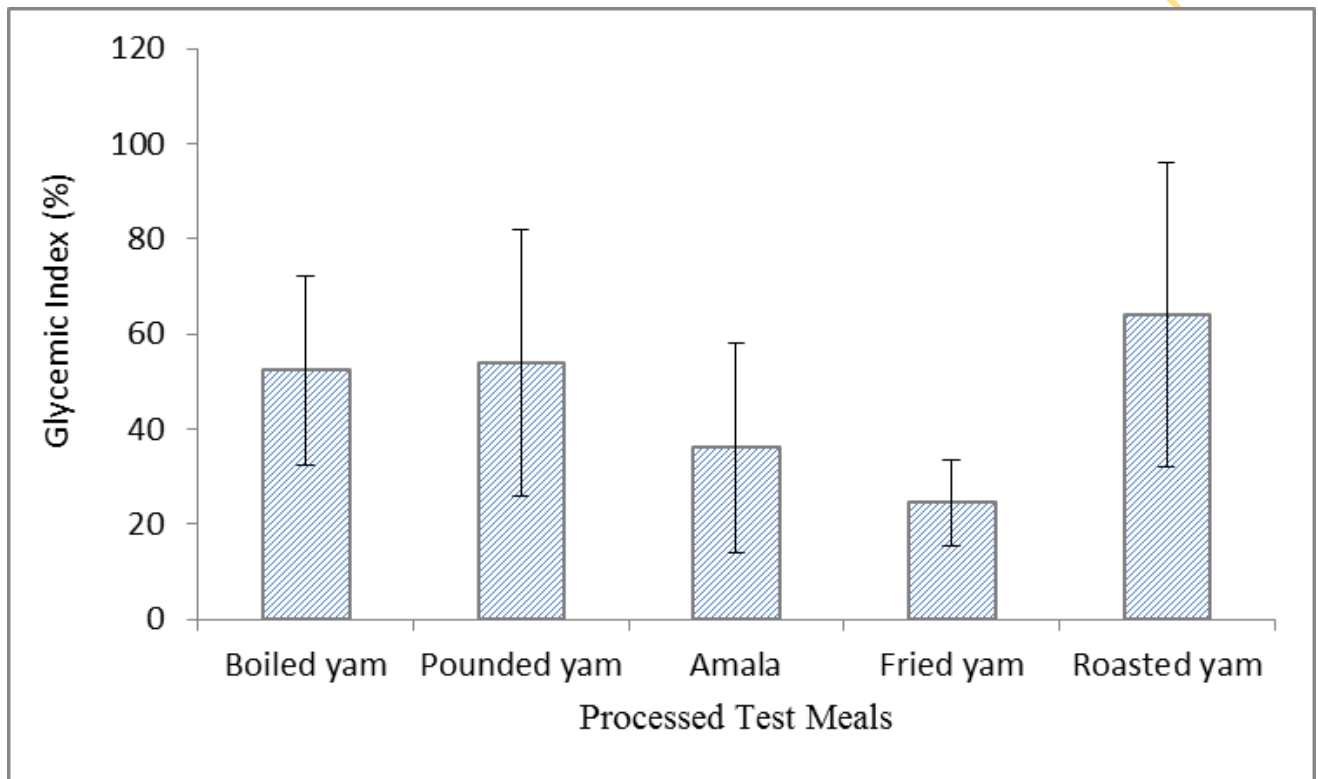
Figure 20: Mean incremental areas under glycemc response curves to various processed test meals (diabetic subjects)



Values are expressed in mean \pm SEM.

*Significantly different from Pounded yam;Boiled yam;Roasted yam

Figure 21: Glycemic index (GI) of test and reference (glucose) meals in healthy subjects.



Values are expressed in mean±SD

Figure 22: Variability of Glycemic index of processed test meals (Healthy Subjects)

4.3 FOOD NUTRIENTS COMPOSITION (PROXIMATES) AND ENERGY (Kcal)

4.3.1 FOOD PROXIMATES AND CALORIFIC VALUE BEFORE PROCESSING

Table 12 below shows the proximate analysis and energy values of the raw white yam before processing and preparation into various test meals used for the study. The protein, fat, carbohydrate, fiber and ash were expressed in grammes (g) while the moisture and the energy contents were expressed in percentage (%) and kilocalories (Kcal) respectively. Values are expressed as means \pm standard errors of means (SEM) of three (3) determinations.

4.3.2 FOOD PROXIMATES AND CALORIFIC VALUES AFTER PROCESSING AND PREPARATION

The proximate analysis and the energy values of the test meals after processing and preparation are shown in Table 13 below. The protein, fat, carbohydrate, fiber and ash were expressed in grammes (g) while the moisture and the energy contents were expressed in percentage (%) and kilocalories (Kcal) respectively. Values are expressed as means \pm standard errors of means (SEM) of three (3) determinations.

The proximate analysis of the test meals after processing and preparation showed that Amala has the highest percentage of moisture content (77.08%) while the roasted yam has the least value (51.39%). These values for Amala and Roasted yams were significantly lower statistically ($p < 0.05$) than the moisture content of the raw yam tuber (80.8%) prior to processing. The change in moisture contents of various test meals reflects the effect of various processing and preparatory methods on the moisture content of food which has implication on the overall effect on the glycemic response and glycemic index of the foods.

The protein content of the test meals was higher in all the processed foods than the raw food. Roasted yam has the highest protein value (9.41g) while Amala has the least value (5.47g).

The difference in the protein values of the test meals when compared to the raw food was statistically different ($p < 0.05$).

Amala displayed very low lipid content (0.0001g) while other test meals showed higher lipid contents compared to the unprocessed raw yam tuber. Fried yam has the highest fat content (2.20g). The difference in their values was statistically significant compared to the lipid content of the raw yam tuber.

The carbohydrate content of the entire test meals were similar in values because all contained equal 50g digestible carbohydrates determined and calculated in the laboratory prior to ingestion by the volunteers. The weights of the test meals containing the 50g carbohydrates are shown in Table 9 below.

Pounded yam has the least fiber content (2.11g) while Amala has the highest fiber content (3.38g). The difference in their values which was statistically significant when compared to the unprocessed yam (0.6g) contributes to the different glycemic indices obtained in Amala and Pounded yam (Table 5 above) as it is generally accepted that presence of fiber slows down digestion thus reducing GI value.

The ash content of the processed test meals showed varying degree of reduction except for the fried yam whose ash content value (1.57g) was slightly higher than the unprocessed yam meal (1.2g). The differences in the value obtained in the entire processed meals compared to the raw meal were statistically significant ($p < 0.05$).

The calories of the processed meals are expressed in Table 11 below. Despite the closeness of their calorific content, fried yam displayed the highest energy content (383.81kcal) revealing the contributory effect of lipid to calories of food. In comparison with unprocessed raw food, the calories of the entire processed foods were highly greater and their differences statistically significant. Amala has the lowest calories content.

4.3.3 GROSS ENERGY COMPARISON BETWEEN PLAIN AND MIXED MEALS

Table 14 below showed the difference in the gross energy content of the plain meals and the mixed meals i.e. test meals with sauce and meat. The difference in their values which was statistically significant ($p < 0.05$) reflects the contributory effect of ingredients to the total calories of foods. However, throughout the study, same type of soup (Ewedu with stew) was served which was prepared from local 'Ewedu' vegetable (*Corchorus Olithorus*) and tomato sauce. The food types (test meals) are eaten in Western Nigeria culture with a bowl of soup and so it was necessary to serve the meals with soup prepared in a standard and uniform way to avoid the introduction of possible variables that may affect the results. The boiled beef meat per serving (25g) was equal in weight throughout the study.

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TABLE 12

FOOD COMPOSITION (PROXIMATES AND CALORIES) ANALYSIS OF WHITE YAM BEFORE PROCESSING AND PREPARATION IN TERMS OF 100g EDIBLE PORTION (DRY WEIGHT %)

Food energy (kcal)	Moisture (%)	Protein (g)	Fat (g)	Carbohydrate (g)	Fiber (g)	Ash (g)
71±0.00	80.8±0.01	1.5±0.00	0.1±0.01	16.4±0.00	0.6±0.01	1.2±0.00

Values are means ± standard errors of means (SEM) of three (3) determinations.

TABLE 13

FOOD COMPOSITION (PROXIMATES AND CALORIES) ANALYSIS OF VARIOUS PREPARED/PROCESSED TEST (YAM) MEALS IN TERMS OF 100g EDIBLE PORTION (DRY WEIGHT %)

Test meals	Food energy (kcal)	Moisture (%)	Protein (g)	Fat (g)	Carbohydrate (g)	Fiber (g)	Ash (g)
Pounded Yam	381.19±0.01	65.52±0.00	7.23± 0.01	0.36±0.00	84.26±0.01	2.11±0.00	1.14±0.02
Amala (Yam Flour Paste)	375.71±0.00	77.08±0.01	5.47±0.00	0.00±0.00	84.12±0-01	3.38±0.02	0.74±0.01
Boiled Yam	380.71±0.02	61.01±0.01	7.66±0.00	0.56±0.01	83.06±0.02	2.41±0.01	0.82±0.02
Roasted Yam	379.52±0.02	51.39±0.00	9.41±0.01	0.59±0.01	80.40±0.02	3.27±0.01	0.51±0.01
Fried Yam	383.81±0.11	54.33±0.01	8.10±0.01	2.20±0.01	79.07±0.00	2.67±0.01	0.57±0.00

Values are means ± standard errors of means (SEM) of three (3) determinations.

TABLE 14

GROSS ENERGY (Kcal) PER 100g OF EDIBLE PORTION OF PLAIN AND MIXED MEALS

Food Types (Test Meals)	Energy (Kcal)	
	Plain Meal (without Soup/sauce and meat)	Mixed Meal (with soup/sauce and meat)
Pounded Yam	381.19±0.01 ^a	485.24±0.12 ^b
Amala (Yam Flour Paste)	375.71±0.00 ^a	419.52±0.00 ^b
Boiled Yam	380.71±0.02 ^a	437.86±0.10 ^b
Roasted Yam	379.52±0.02 ^a	436.67±0.11 ^b
Fried Yam	383.81±0.11 ^a	497.14±0.21 ^b

Values are means ± standard errors of means (SEM) of three (3) determinations.

Means with the different letter (superscripts) within the same row are significantly different at P value < 0.05.

STUDY PHASE II:

COMPARISON OF LOW- AND HIGH-GI DIETS (ANIMAL STUDY)

4.4 BODY AND ORGAN WEIGHTS

4.4.1 BODY WEIGHT

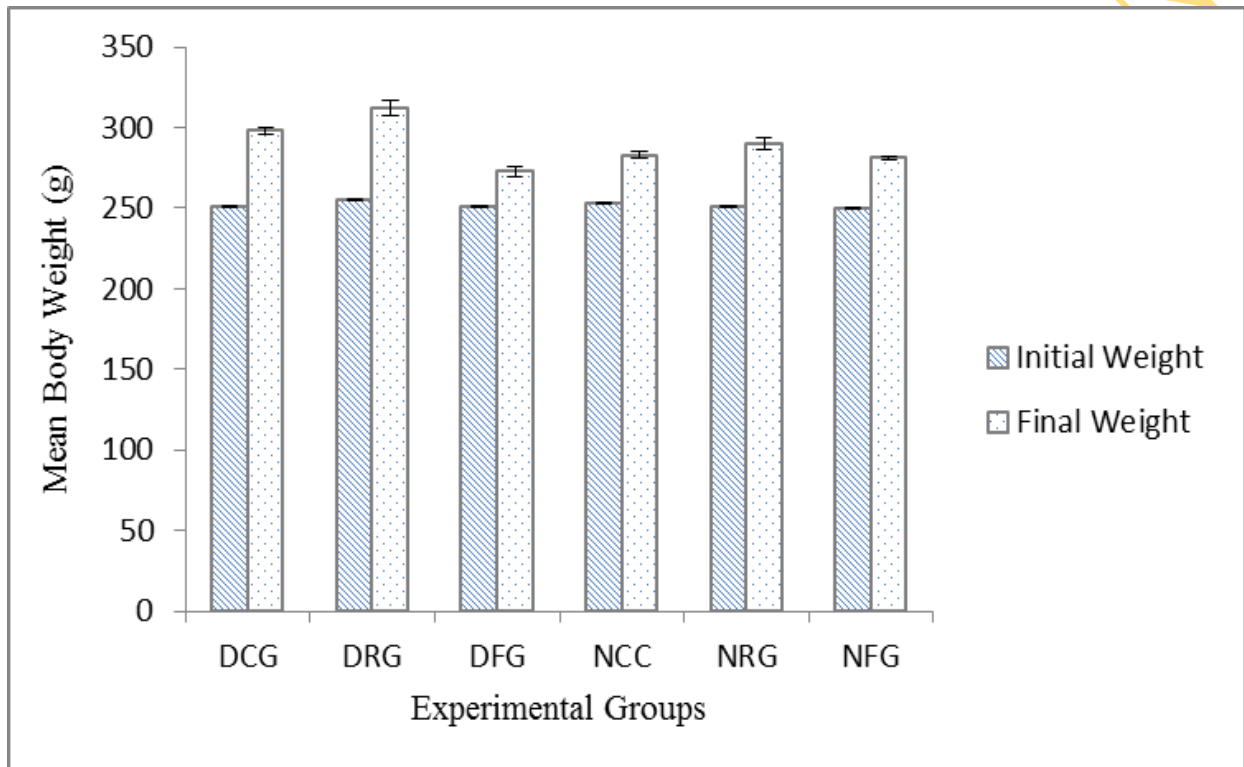
The duration of this study lasted for 12 weeks. As shown in Table 15, the mean body weights were almost the same (~250 g) in all groups at the start of the study. At the time of sacrifice, mean body weight was significantly highest ($p = 0.001$) in diabetic roasted yam-fed (DRG) group ($312.33 \pm 5.85\text{g}$) and lowest in diabetic fried yam-fed (DFG) group (273.17 ± 1.72) compared to the normal and diabetic controls. Both low- and high-GI diets intake had significant effects ($p < 0.05$) on body weight however, the high-GI diets (Roasted yam) had more effect on body weights compared to low-GI diets (Fried yam). Rats fed with roasted yam gained weight more than the rats fed with normal or fried yam. Figure 23 below reflects the graphical representation of the effect of the various diets on the mean body weights of the experimental rats before and after 6 weeks period.

TABLE 15
Effect of Control, Low- and High-GI Diets on Body and Organ Weights of
Experimental Rats.
(n = 6)

	Experimental Animal Categories					
Body Weight (g)	DCG	DRG	DFG	NCG	NRG	NFG
Initial	251.00±1.61 ^a	254.67±2.46 ^a	250.67±1.12 ^a	252.67±2.17 ^a	251.33±1.19 ^a	250.33±0.62 ^a
Final	297.67±6.23 ^a	312.33±5.85 ^b	273.17±1.72 ^{ab}	282.50±1.34 ^a	289.83±1.82 ^a	281.17±2.72 ^a
Organ Weights (g)						
SPLEEN	0.78±0.00	0.73±0.00	0.75±0.02	0.67±0.00	0.65±0.01	0.65±0.02
KIDNEYS	1.46±0.00	1.39±0.00	1.42±0.02	1.23±0.00	1.28±0.02	1.21±0.03
LUNGS	1.18±0.00	1.47±0.00	1.3±0.06	0.92±0.00	1.07±0.12	1.21±0.07
HEART	0.49±0.00	0.58±0.02	0.53±0.02	0.66±0.00	0.66±0.00	0.67±0.03
LIVER	5.56±0.00	5.52±0.04	5.54±0.04	5.56±0.02	5.24±0.15	4.9±0.00
TESTES	2.12±0.01	2.44±0.06	2.32±0.10	2.16±0.01	2.2±0.03	2.25±0.02

Values are expressed in mean±SEM

NFG = Non-Diabetic Group fed with fried yam (Low-GI); NCG = Non-Diabetic Group fed with normal feed; NRG = Non-Diabetic Group fed on roasted yam (High-GI); DFG = Diabetic Group fed with fried yam (Low-GI); DCG = Diabetic Group fed with normal standard rat feed; DRG = Diabetic Group fed on roasted yam (High- GI).



Values are expressed in mean±SEM

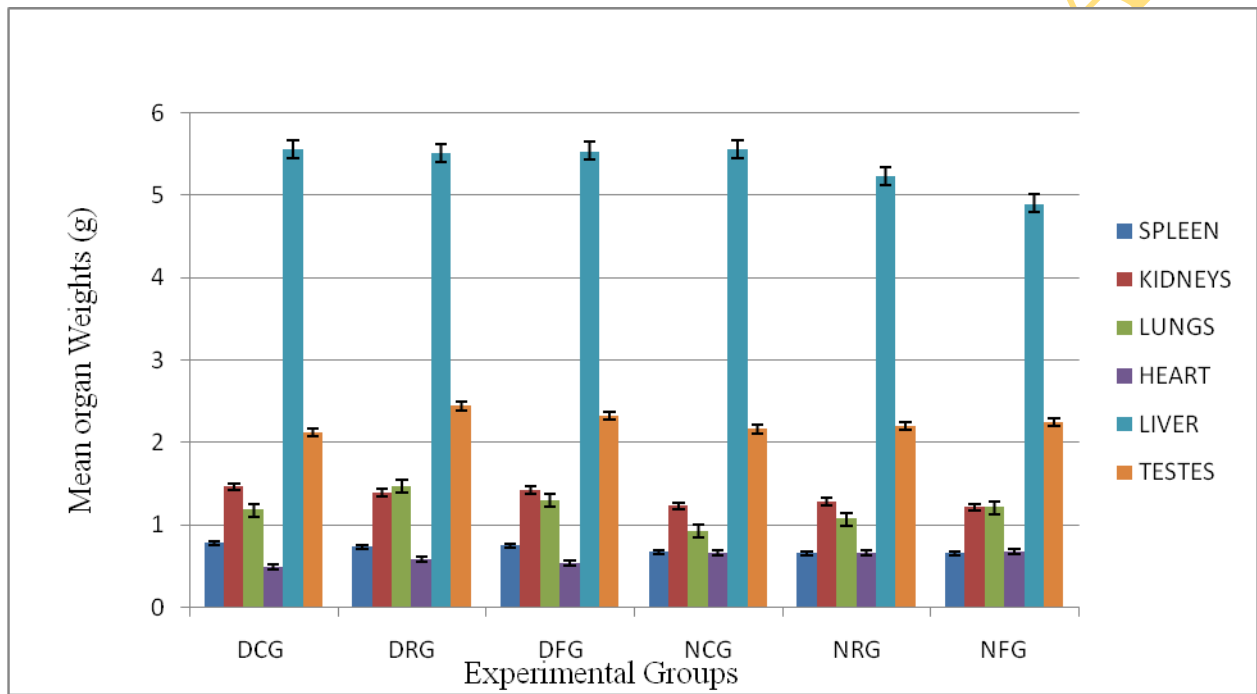
NFG = Non-Diabetic Group fed with fried yam (Low-GI); NCG = Non-Diabetic Group fed with normal feed; NRG = Non-Diabetic Group fed on roasted yam (High-GI); DFG = Diabetic Group fed with fried yam (Low-GI); DCG = Diabetic Group fed with normal standard rat feed; DRG = Diabetic Group fed on roasted yam (High- GI).

Figure 23: Effect of low and high-GI diets on body weights of experimental rats after 6 weeks.

4.4.2 ORGAN WEIGHTS

The relative weights of organs such as liver, heart, kidney, lung, spleen and testes (Table 13 above) were not significantly affected by any of the diets used in this study. The values obtained in normal and diabetic rats fed with roasted and fried yams were not significantly different ($p > 0.05$) compared to the normal and diabetic controls fed with standard rat. Organ weight measurement is important to assess general toxicity because any change in organ weight is a sensitive indicator of toxicity. This finding is similar to the report of other studies (Geetha et al, 2011). In theory, organ weight will be affected by the suppression of body weight as described by Marshall (2000). In this study, the Low- and the High-GI diets did not give any significant change in the organs' relative weights of rats in normal and diabetic groups fed with roasted and fried yams compared to the normal and diabetic control groups fed with standard rat feed.

The diagrammatic representation of the effect of the test (Low- and High-GI yam meal) and standard (normal rat feed) diets on relative organ weights is shown below in figure 24.



Values are expressed in mean±SEM. NFG = Non-Diabetic Group fed with fried yam (Low-GI); NCG = Non-Diabetic Group fed with normal feed; NRG = Non-Diabetic Group fed on roasted yam (High-GI); DFG = Diabetic Group fed with fried yam (Low-GI); DCG = Diabetic Group fed with normal standard rat feed; DRG = Diabetic Group fed on roasted yam (High- GI).

Figure 24: Effect of standard, low- and high-GI diets on organ weights of experimental rats.

4.4.3 ORAL GLUCOSE TOLERANCE TEST (GLYCEMIC RESPONSE)

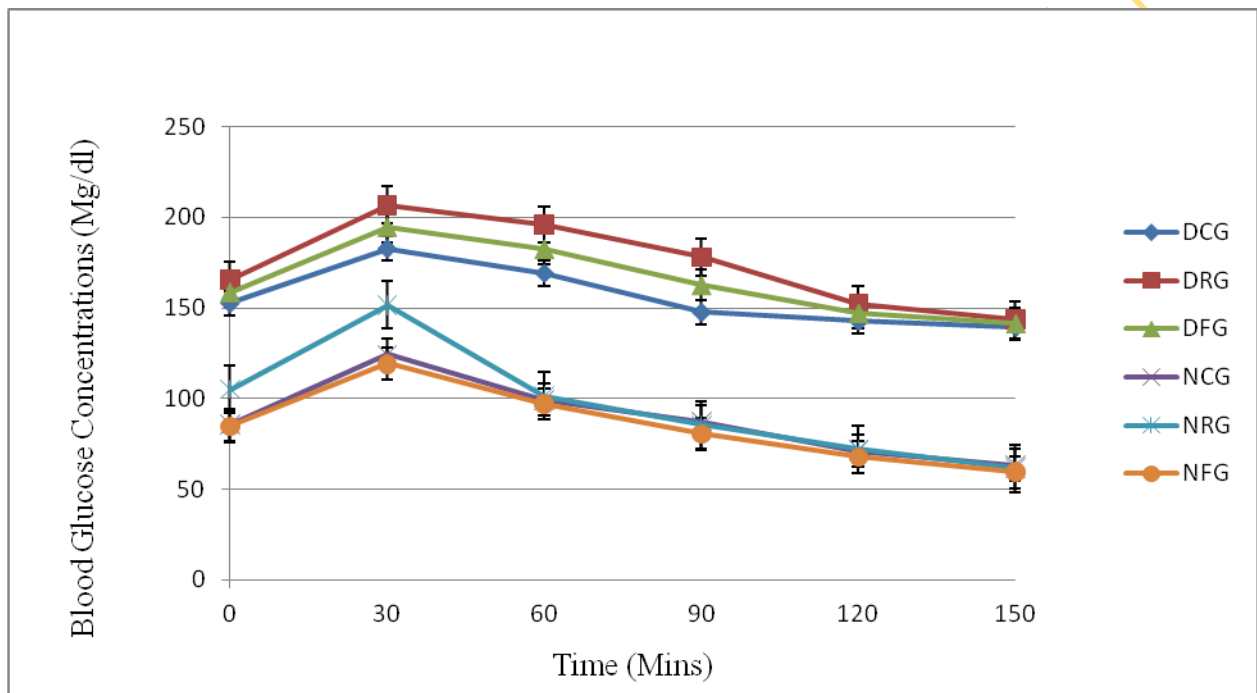
The response of the experimental rats to oral glucose challenge is depicted by the glycemic response curves of figure 25 below. The picture of the glycemic response to roasted and fried yam meals in the experimental rats of both diabetic and non-diabetic groups is similar to that observed in the human subject study. The high-GI diet (roasted yam) fed rats (DRG and NRG groups) displayed significant quicker and higher glycemic responses to oral glucose challenge as compared to the Low-GI diet (fried yam) fed diabetic rats (DFG) and Non-diabetic rats (NFG) which displayed significant delayed, slower and decreased glycemic responses to oral glucose challenge. Both DCG and NCG groups (standard diet fed diabetic and normal rats) displayed relative normoglycemic response curves.

This observation agrees with the findings of other studies which reported significant decrease glycemic response and insulin release to diets that are high in fiber and oil in comparison to more traditional grain-based feeds (Williams et al., 2001) and also for grain-based feeds that are top-dressed with oil (Pagan et al., 1999).

The reduction in glycemic response to low-GI diet feeding is partly due to the lower starch and sugar content and also likely to be influenced by the presence of oil in a feed. Fat delays the peak but not the total glucose response (Marion, 2000 and Gannon et al, 2001). The more fat or acid a carbohydrate food contains, (or, the more fat or acid in the stomach, during digestion) the slower the carbohydrate food is converted to glucose and absorbed into the bloodstream. The presence of fat and/or acid retards the emptying of the stomach.

Figure 24 below displayed the collective representative picture of the glycemic response curves pattern to oral glucose in all experimental rats. The mean incremental areas under the glycemic response curves for the rats fed with high-GI diets are significantly higher than those fed with low-GI diets. The diabetic and normal controls rats (DCG and NCG) displayed the least incremental areas under the glycemic response curves.

The glycemic response curves in the experimental rats peaked at 30 minutes of oral glucose administration in the entire rats in different groups. This is different from that observed in the human subjects which occurred at 60 minutes of oral glucose ingestion.



NFG = Non-Diabetic Group fed with fried yam (Low-GI); NCG = Non-Diabetic control Group fed with normal feed; NRG = Non-Diabetic Group fed on roasted yam (High-GI); DFG = Diabetic Group fed with fried yam (Low-GI); DCG = Diabetic control Group fed with normal standard rat feed; DRG = Diabetic Group fed on roasted yam (High- GI).

Figure 25: Mean incremental blood glucose responses in experimental rats (glycemic tolerance).

4.5 LIPID PROFILE

The results of the lipid profile as seen in Tables 14 and 15 below clearly showed that fried yam (low-GI diet) fed groups whether diabetic or non-diabetic rats showed higher significant increase ($p > 0.05$) in total cholesterol (TC) level compared to roasted yam fed groups. Similarly, triglycerides (TG) and LDL show significant increase in fried yam fed groups than roasted yam fed groups. The reverse is the case in HDL level which was observed to be lower in rats fed with fried yam as compared to roasted and control groups. After 6 weeks of diet, levels of the constituting parameters of the lipid profile increased in all groups but much more significant in fried yam fed groups than roasted yam fed groups except for HDL.

The main rationale for providing a high carbohydrate intake has been the possibility of decreasing dietary fat and cholesterol intake, since diabetics who have their carbohydrate intake restricted consume greater proportion of fat. Such high fat intake has been associated with raised blood lipids and an increased risk of cardiovascular diseases (Jenkins et al, 1980).

With respect to the above statement, similar observation of elevated blood lipids was made in this study which corresponds with the findings of other studies (Jenkin et al, 2000).

However, very high carbohydrate diet has been observed to result in a rise in fasting triglycerides in hyperlipidemic patients, in diabetics and in normal subjects (MacDonald, 1978). Such carbohydrate-induced lipidemia has been linked to the high insulin levels stimulated by the high carbohydrate diet (Olefsky et al, 1974).

The low-GI diets in this study which promotes good glycemic response is fried yam, however caution should be taken to avoid long term consumption of fat enriched carbohydrates because of the effects on lipid profile with associated increased risk of cardiovascular diseases.

Table 16
Effect of low- and high-GI Diets on lipid profile of Diabetic Experimental Rats Groups
(n= 6)

Time (weeks)	Experimental groups		
	DFG (Low-GI)	DCG (Diabetic control)	DRG (High-GI)
	Total cholesterol (TC) mg/dl		
Baseline	42.00±1.47	41.20±3.05	41.23±2.25
6	60.50± 1.50 ^b	47.22±6.85	45.40 ±2.60
	Triacylglycerol (TG) mg/dl		
Baseline	21.05±1.30	20.45±2.00	20.05±2.30
6	52.24±1.00 ^b	28.34±2.90	29.54±2.70
	High density lipoprotein cholesterol (HDL- C) mg/dl		
Baseline	8.85 ±1.85	8.75 ±1.05	8.95 ±1.75
6	8.35±2.50	10.46±2.34 ^b	11.00±2.80 ^b
	Low density lipoprotein cholesterol (LDL- C) mg/dl		
Baseline	34.05±7.25	35.00±6.30	35.05±6.25
6	54.32± 2.65 ^b	39.72±1.20	38.50±1.00

Means with the same letter in same row are not significantly different. P value < 0.05 is significant.
DFG = Diabetic Group fed with fried yam (Low GI); DCG = Diabetic Group fed with normal standard rat feed; DRG = Diabetic Group fed on roasted yam;

Table 17
Effect of low- and high-GI Diets on lipid profile of Non-Diabetic Experimental Rats Groups
(n= 6)

Time (weeks)	Experimental groups		
	NRG (High-GI)	NFG (Low-GI)	NCG (Control Group)
	Total cholesterol (TC) mg/dl		
Baseline	41.20±3.05	40.00±3.25	41.23±2.25
6	42.22±2.51	57.05±1.95 ^b	43.40 ±2.60
	Triacylglycerol (TG) mg/dl		
Baseline	20.45±2.00	21.00±2.35	20.05±2.30
6	29.34±2.90 ^b	49.44±3.00 ^b	28.54±2.70 ^b
	High density lipoprotein cholesterol (HDL- C) mg/dl		
Baseline	5.75 ±1.05	6.05 ±1.75	5.95 ±1.75
6	10.46±2.34 ^b	8.00±1.85 ^b	11.00±2.80 ^b
	Low density lipoprotein cholesterol (LDL- C) mg/dl		
Baseline	35.00±1.00	36.05±1.30	35.05±1.25
6	36.45±0.78	56.15±1.25 ^b	37.11±0.75

Means with the same letter in same row are not significantly different. P value < 0.05 is significant.
NFG = Non-Diabetic Group fed with fried yam (Low GI); NCG = Non-Diabetic Group fed with normal standard rat feed; NRG = Non-Diabetic Group fed on roasted yam;

4.6 HISTOLOGICAL ANALYSIS.

Under high power magnification light microscopic examination, the photomicrographs of the pancreas of the diabetic and normal rats fed with low- and high-GI diets were closely examined (PLATE 1 – 3 below).

Pancreatic islet cells of rats in NRG, NFG and NCG (normal control) groups exhibited normal histoarchitecture, namely clearly visible darkly stained serous acini containing centroacinar cells while the diabetic rats in DCG, DRG and DFG groups exhibited degenerated islets with degranulation and vacuolization of β -cells. However, the histoarchitecture of the rats in the DFG group fed with low-GI diet showed some visible regeneration of the β cells displaying the advantageous effect of low-GI diets over the high-GI diets in diabetic control through regeneration of the pancreatic islets tissue and improved glycemic response.

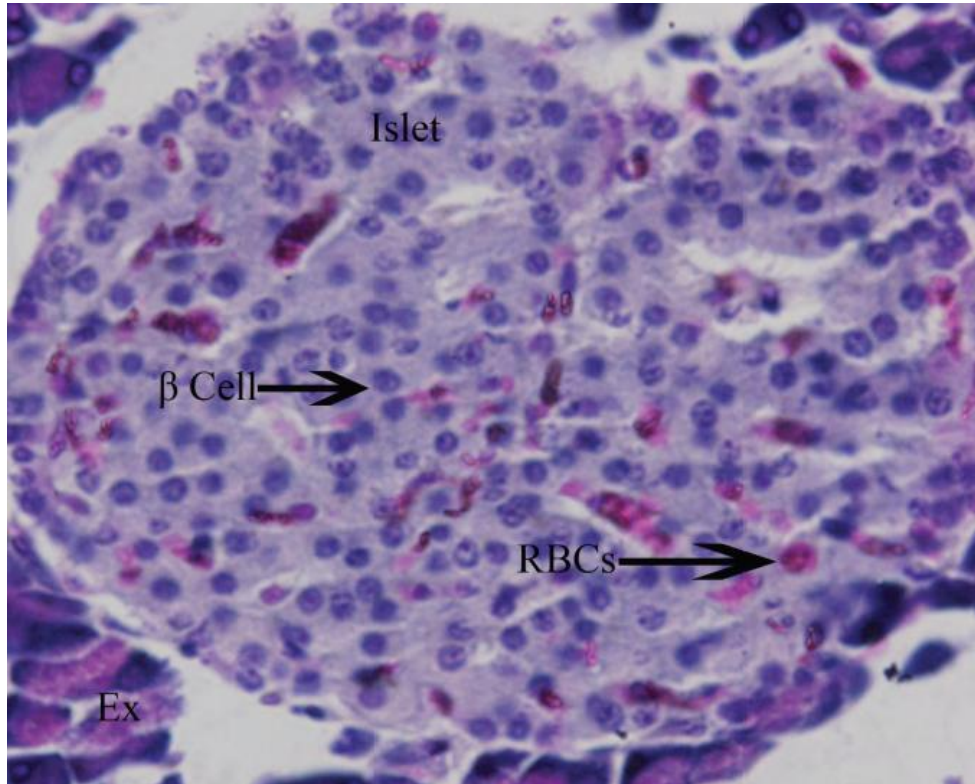


PLATE 1: Photomicrograph of the pancreas from NCG (normal control) rat demonstrating normal histoarchitecture. Blood capillaries are surrounded by centroacinar cells containing serous acini (hematoxylin and eosin; original magnification x400). EX: Exocrine pancreas, β cell: beta cells, RBCs: red blood cells.

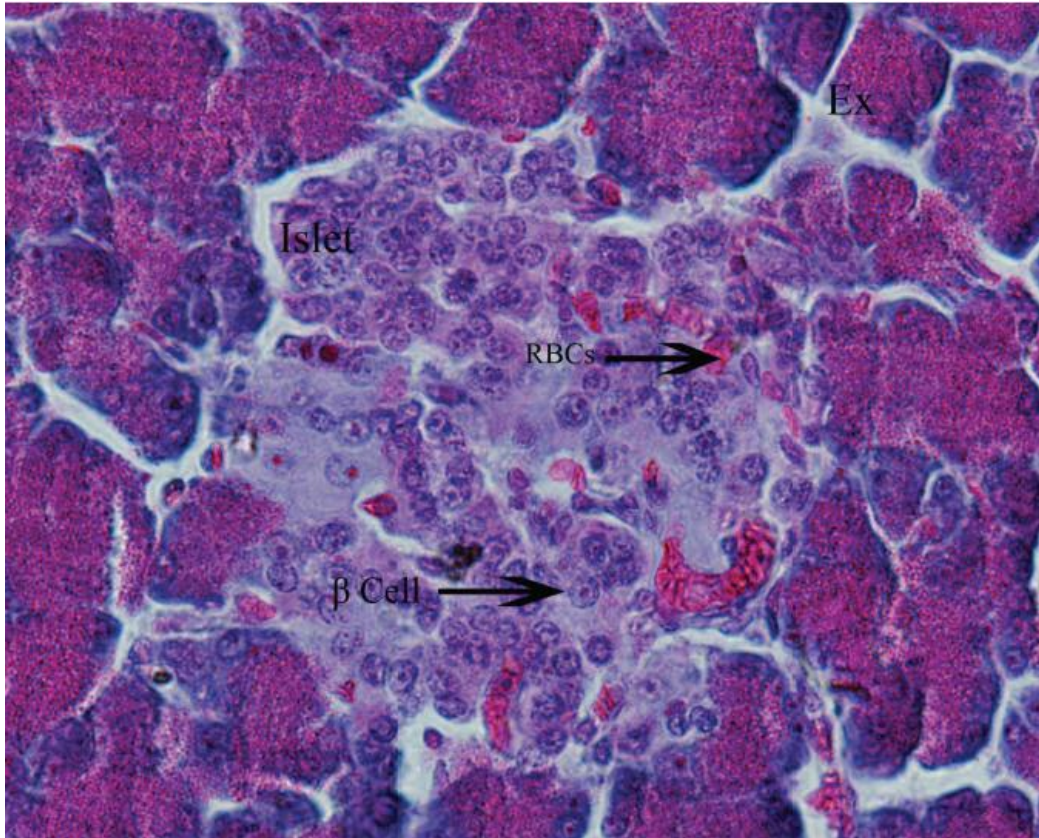


PLATE 2:Photomicrograph of the pancreas from DRG group rat (diabetic rat on high-GI diets) showing the exocrine region and islets of Langerhans with damaged β cells due to necrosis (degranulation and degeneration) and a decreased number of β cells. (hematoxylin and eosin; original magnification x400)
EX: Exocrine pancreas, β cell: beta cells, RBCs: red blood cells.

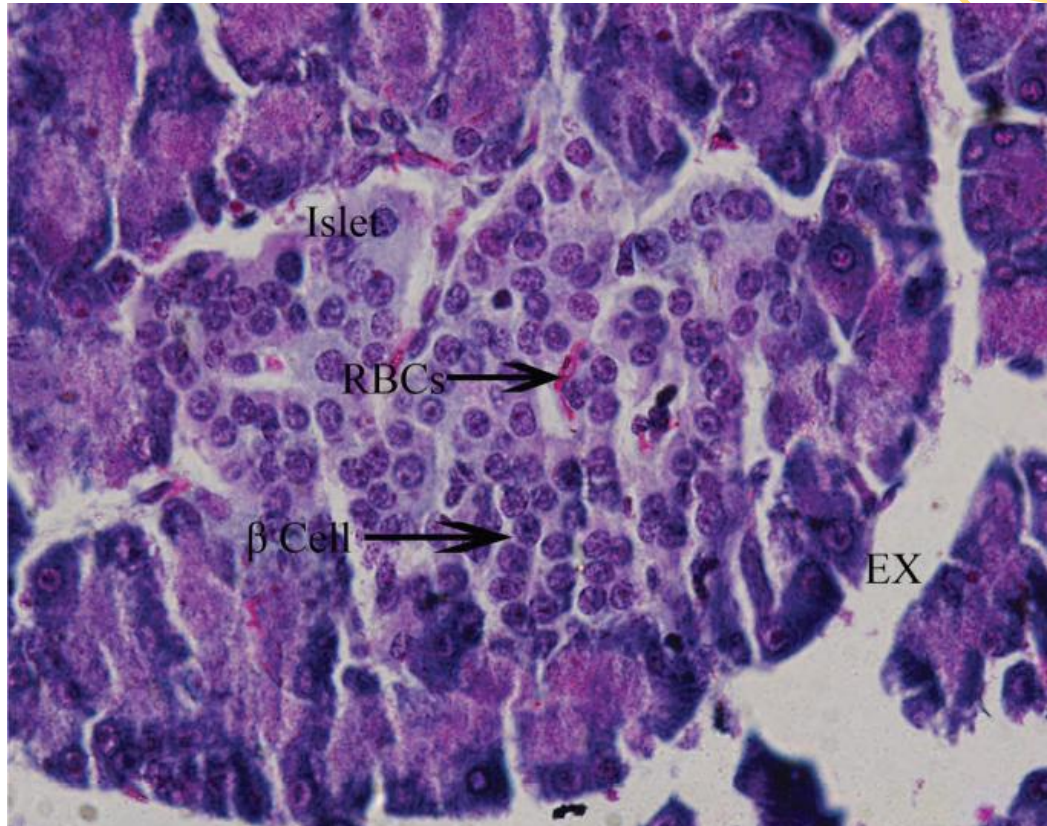


PLATE 3: Photomicrograph of the pancreas from a diabetic rat on low-GI diet (DFG group) showing degenerated serous acini, although some regeneration of β cells is also visible (hematoxylin and eosin; original magnification x400). EX: Exocrine pancreas, β cell: beta cells, RBCs: red blood cells;

CHAPTER FIVE

5. 1 DISCUSSION

Human diet contains many types of carbohydrates each of which contributes to different physiologic responses (Cumming et al, 1997). Starch is considered by many to be digested slowly, resulting in a modest glycemic response. However the rate and extent to which starch is digested and absorbed, and the resulting glucose and insulin responses, vary considerably depending on the source and food processing (Englyst et al, 1986, 1987; Jenkins et al, 1990; Snow et al, 1981; Heaton et al, 1988 and Larsen et al, 1996),

Recent knowledge of the variation in glycemic response to carbohydrate-containing foods comes largely from measurements of glycemic index (GI). In principle, GI is calculated as the measured glycemic response to a portion of test food that contains 50g of “available” carbohydrate expressed as a percentage of the glycemic response to the same amount of “available” carbohydrate from a standard food eaten by the same subject (Jenkins et al, 1981 and Wolever et al, 1991).

GI values have been published for a wide range of foods (Foster-Powell et al, 2002) and have been used in several studies to design low-glycemic load diets for diabetic subjects (Brand et al, 1991; Wolever et al, 1992; Fontvielle et al, 1992 and Frost et al, 1994). The methods for defining GI are not standardized with values having large inter- and intra- individual variability as demonstrated also in this study as well as other several studies (Rasmussen et al, 1990, 1993 and Riccardi et al, 2003)

The accuracy of GI values determined is influenced particularly by several factors which include: method of defining the amount of tested food which contains 50g of hyperglycemic (i.e absorbable, digestible) carbohydrates, method of calculating the incremental area under the curve (IUAC), subjects variability, time of the day when the test is carried out and day to day glycemic

variability (Berger, 1995). In this study, great care was taken to eliminate the sources of undue variability, as strict adherence to standard protocol was observed throughout the study.

This research study carried out in two phases using human subjects and animal model (rats), determined the effect of food processing on the nutrient composition of foods, the glycemic response and the GI of yam-based meals prepared by different processing methods. It also compared the effects of the low-GI and high-GI diets prepared on body and organ weights, glycemic tolerance, lipid profile and the pancreas histoarchitecture. GI values obtained for various prepared foods in the study were also compared between the diabetic and non-diabetic healthy subjects. Observations made in this study, showed that method of food processing by way of alteration in the micronutrients (proximate) and physical properties of the carbohydrate food (Table 13) affected the rate of starch digestion (quantified by the glycemic responses) and subsequent GI values (Table 7) obtained for different processed foods derived from the same carbohydrate source (white yam – a commonly found species of the genus *Dioscorea*). This observation is in agreement with the works of other researchers (Jenkins et al, 2002; Foster-Powel et al, 2002) who studied the effect of processing on different food types.

Five different methods of processing were employed in the preparation of the test foods and their nutrients composition determined in the laboratory to see the extent and the degree of effect of processing on their proximate values which was compared with the unprocessed raw food proximate values (Tables 12 and 13). Their GI values (Table 7 above) obtained in both study groups were as follows:

In diabetic subjects, Roasted yam has the highest GI (93.34%) while Fried yam has the lowest GI (36.16%). Others include Boiled yam (GI=65%), Pounded yam (GI=70.75%) and Yam Flour Paste, Amala (50.09%). Similarly in non-diabetic healthy subjects, roasted yam has

the highest GI (64%) while Fried yam has the lowest GI (24.5%). Others fall within this range: Boiled yam (GI=52.32%); Pounded yam 53.74%) and Amala (GI=36.12).

In both groups, method of roasting resulted in highest values of GI while frying with vegetable oil further reduced the GI value to the lowest as shown above. GI values differences obtained for the test meals within each group were statistically significant ($P < 0.05$) when compared with one another. This supported the hypothesis that processing of foods affects the glycemic response (GR) and the GI of the food through alteration of the carbohydrate micronutrient content (proximate) and bioavailability (Foster- Powel et al, 2002), rate and extent of carbohydrate digestion and absorption (Jenkins et al, 1980; Snow et al, 1981; Englyst et al, 1985-87 and Hearton et al, 1988) and distinct changes or disruption in the physicochemical structures of the carbohydrates ((Boohher et. al, 1951). However, 60% (3) of the GI values of the test meals showed statistically significant differences when compared between the Diabetic and Non-diabetic Healthy subjects as shown in Table 7. This also supported the view that GI value is affected by the presence of metabolic dysfunction and disorder as revealed in the study of Donduran et al (1999).

Diabetes is fundamentally a disorder of glucose metabolism. Therefore, influence of dietary carbohydrate type on the course and management of Diabetes Mellitus (DM) should be critically considered.

Subjects with DM, as would be expected, had a slower rate of glucose clearance in the blood hence, the peculiar nature of the GR curves obtained with the blood glucose variability (figure 10). However, the healthy subjects had a better glucose clearance following ingestion of meals rich in carbohydrates. The soup served with each meal (as it would be expected in the culture to serve the meal with a bowl of soup) had the same content and was prepared in the

same expected standard manner. Therefore, this should not have contributed to the variability of the observed GI to any appreciable extent.

Modern methods of food processing such as extrusion cooking, explosion puffing and instantization appear to make starch in this food more readily digested (Granfeldt et al, 1995). Conditions which are known to increase the digestibility of starches are those which produce obvious hydration of the granules (gelatinization), distinct changes in chemical nature or disruption of the organized granule structure (Booher et al, 1951).

Yam pounding in a mortar with pestle, which is a traditional method of preparing special yam delicacy (Pounded yam, Iyan or fufu) in our environment, results in the disruption of the organized granule structure of starch in the pounded yam (Ralet et al, 1993) thus causing increase in the availability of starch to amylase enzyme digestion. As a result, more glucose is rapidly absorbed into the system.

Altering the physical form of carbohydrates changes the postprandial glucose and insulin response profile (O'Dea et al, 1980, 1981 and Wong et al, 1983). Thus, pounding of boiled yam increased the postprandial glycaemic response as observed in this study (Tables 5 and 10) which agrees with the findings of O'Dea et al (1983) where the postprandial glycaemic response of grounded rice was shown to be higher than that of the ungrounded rice in diabetic and healthy subjects.

Fried yam was observed to have the lowest GI value among all other test foods in both diabetics and healthy subjects. This may result from the effect of fat in gastric emptying and rate of intestinal absorption. Fried yam has the highest lipid content after processing compared with the other test foods (Table 13). Marion (2000) and Gannon et al (2001), demonstrated the above fact in their studies which revealed delay in peak postprandial GR to test diet. This effect was

more pronounced for the rapidly absorbed carbohydrates as also observed in this study with diabetic and healthy subjects (Table 6 and Table 11 respectively).

The role of food on gastric emptying rate of starchy meals and the correlation with the blood glucose response was studied by Tosdottir et al (1984) on healthy and diabetic subjects. A high correlation was found between the difference in gastric emptying and the difference in blood glucose and insulin responses (rice meal versus mashed potato meal – the time was greater for rice). The high correlation observed may imply that digestion of starchy foods is not the sole limiting factor for glucose absorption as observed in the case of mashed potato versus mashed white bean which displayed same gastric emptying rate but different glucose and insulin response (Wursh et al, 1986).

The above statements may be responsible for the observed flattening of the GR curve and the reduced GI of the fried yam (Figure 19 and Table 7).

Processing of Yam Flour meal was more laborious than the other test meal but however, it has lower GI than others except fried yam. This was in contrast to several studies view which state that: ‘the more processed a food is, the higher the digestion rate and subsequently the glycemic and insulin responses’ (Booher et al, 1951; Thorne et al, 1983 and Jenkins et al, 2003).

The explanation for the above difference may stem from some factors affecting the readily availability of starch in yam flour meal. During process of boiling yam in water, gelatinization of the starch molecules occurs which increases the availability of starch to the amylolytic enzymes digestion (Granfeldt et al, 1995). This is the case for boiled yam when eaten directly as well as pounded yam without further processing. However, in yam flour meal preparation, the par-boiled yam is sun-dried for about 48-72 hours, losing almost all of its water content with a progressive re-association of the starch molecules (retrogradation) (Akingbala et al, 1995 and WHO/FAO 1997). This association reduces the starch molecules digestibility.

The processing undergone by the parboiled yam may also increase the fiber content as observed in this study during the proximate analysis of the test food where yam flour meal displayed the highest fiber content value (3.38g) among others. It is well known that fiber of yam flour is greater than that of the raw tuber (Plat, 1982). Fiber viscosity effect on postprandial glycemic response to food has been demonstrated by several studies (Jenkins et al, 1977 and Morgan et al, 1978). Other factor that may contribute to the reduction of the GI of Amala may be due to reduction in exposure time to heat as compared to roasting process during preparation for consumption which may affect its availability for digestion (Collins et al, 1981). Another factor is the escape of the chewing process during consumption which normally enhances digestion. Normally, yam flour paste (Amala) is mostly swallowed in our culture and not chewed. A study reported that swallowing of food reduced the in-vivo postprandial glycemia of meals (Read et al, 1986). The above observations may be largely responsible for the low GI of Amala despite the rigorous processing.

Roasted yam has the highest GI value in this study (Table 7). This may be as a result of extent and degree of exposure to heat during roasting processing and preparation. This obviously increase the readily availability of starch to digestion as previously described. Such condition is often seen in factory processing. Eating of roasted yam in our environment generally requires drinking of much water which may enhance the hydration of the starch granules thus increasing availability and rate of digestion. Moreover, roasted yam is well masticated before swallowing which further enhances the digestion. These observations may contribute to the high GI value observed.

The GI approach was used in this study to allow the comparison of the results between different individuals. This was possible since each subject was standardized with a reference food (in this case glucose).

Based on the GI concept (Jenkins et al, 1981), foods may be classified into three groups. Foods with $GI \leq 55\%$ are regarded as Low-GI while those with $GI \geq 70\%$ as High-GI foods. GI of foods falling between 55% and 69% are called intermediate or medium - GI. A glycemic index classification of some Nigerian diets was attempted to provide an indication of the rate of digestion of different starchy foods (Anyakudo and Fasanmade, 2007) and it was hoped that the selection of foods with lower GI as recommended by WHO/FAO through epidemiological and intervention studies would contribute to the nutrients absorption and possibly improve glycemic profile in diabetics.

Recently, focus on GI of Nigerian foods has attracted attention by some nutrition researchers to enable the development of data based nutritional table with GI values of Nigerian foods (Anyakudo and Fasanmade, 2007). This would help in providing information on dietary guidelines and recommendations by the dieticians in the dietary management of diabetics in our environment.

Generally, the GI values of foods in young, normal weight non-diabetic volunteers has been observed to agree well with those of middle-aged overweight diabetic patients (Jenkins et al, 1983). However, the agreement between GI values between the above has been called to question in this study whose finding is not in agreement with the statement as also observed by other studies (Jannet et al, 1984; Jenkins et al, 2003; Anyakudo and Fasanmade, 2007). In this regard, it is very important to use the results obtained using diabetic volunteers for the dietary plan management of diabetes rather than extrapolating the results obtained using non-diabetic healthy subjects or animal studies.

In support of the above statement, the wide variability of GI observed by several studies as well as in this study, shows that other factors yet to be determined may be at play in the response of individuals to carbohydrate ingestion. Therefore, in this wise, one should be careful

in interpreting the results of GI determination of foods, especially when such results are used as the sole basis for therapeutic recommendations

The result of this study supports the view that the more processed a food is, the higher the GR it will produce and the higher the GI subsequently (Thorne et al, 1983). However, this view is in contrast to the finding obtained for Amala (Yam Flour Meal) where processing reduced the glycemic impact as also observed in pasta by other researchers (Jenkins et. al, 2003 and Anyakudo and Fasanmade, 2007). The study on yam flour has been conducted by some researchers and similar observations were made; the reason which could be due to those aforementioned factors above.

Frying processing favoured low-GI of yam meal products as observed in this study. However, caution should be taken in diabetic with concurrent hypertension when advocating process of frying because hyperlipidaemia complicates hypertension which in turn, may accelerate the progress of diabetic complication. Frying processing is better in non-hypertensive diabetics than hypertensive diabetics. The observation made in the animal study on the effect of fried yam meal on lipid profile supports the above statement. The total cholesterol, TG and LDL values were significantly on the high side compared with the rest of the test diets.

Presently, Amala-based meals are encouraged and should be used more generously to supply the 55-75% of the recommended daily calories from carbohydrates (Anyakudo and Fasanmade, 2007) while boiled yam should be eaten occasionally.

Healthy subjects especially those with positive family history of type 2 DM (NIDDM) can also be advised to follow this recommendation above.

Roasted and pounded yam should be discouraged in diabetic individuals.

The results of this study should guide dieticians in our environment and researchers in the field of nutrition in appropriate dietary recommendations to diabetic patients and to subjects desiring low caloric food intervention such as overweight and obese individuals.

Further studies on the effect of other type of food processing on carbohydrates especially yam and yam-based meals with other tubers are necessary to establish a data-based nutritional table of GI of local Nigerian foods thus ascertaining their suitability or otherwise in their incorporation into the recommended menu in the dietary management of diabetes.

In summary, relative to the degree, extent and nature of processing, not all yam-based food products should be encouraged in diabetic patients. In addition, dietary caution should be indicated when recommending dietary pattern to diabetics.

In the animal study, the high-GI diet (roasted yam) significantly increases the total body weight more than the low-GI diets (fried yam) while the effect of both on the organ weights were insignificant. At the time of sacrifice, mean body weight of experimental animals was significantly higher ($p < 0.05$) in all the groups: highest in DRG group ($312.33 \pm 5.85\text{g}$) and lowest in DFG group ($273.17 \pm 1.72\text{g}$). High-GI diet had much more effect on body weight as compared with low-GI diet. This observed increase in the body weight is in line with the findings of few studies which suggested that total dietary fat intake and overconsumption of high-GI foods are linked to an increased risk of obesity (Astrup et al, 2008) and diabetes (Ma Y. et al, 2006). Due to increasing obesity and altered dietary habits in both western and developing countries, the prevalence of type 2 diabetes mellitus is growing at an exponential rate (Zimmet et al, 1996), thus dietary precaution should be taken when recommending dietary menu to the diabetic in our environment.

Some studies showed that glycemic response and insulin release is significantly lower in response to diets that are high in fiber and oil in comparison to more traditional grain-based feeds

(Williams et al., 2001) and also for grain-based feeds that are top-dressed with oil (Pagan et al., 1999). The reduction in glycemic response to feeding is partly due to the lower starch and sugar content of these former feeds but is also likely to be influenced by the presence of oil in the latter.

This statement above supports the observation made in this study on the glycemic tolerance of fried yam-fed rats to oral glucose challenge as compared to the roasted yam-fed rats (Figure 24). The fried yam fed diabetic rats (DFG) displayed significant higher delayed glycemic response to oral glucose challenge with a flatter GR curve while the roasted yam fed diabetic rats (DRG) displayed quick and faster GR to oral glucose challenge. This observed effect is similar to that experienced with human subjects strongly supporting the facts that diets with high GI values stimulate higher GR than those with low GI values.

This observation agrees with the findings of other studies which reported significant decrease glycemic response and insulin release to diets that are high in fiber and oil in comparison to more traditional grain-based feeds (Williams et al., 2001; Gannon et al, 2001 and Marion, 2000) and also for grain-based feeds that are top-dressed with oil (Pagan et al., 1999). It also reflects the effect of diabetes mellitus on dietary glucose handling.

Pancreas is an important organ of glucose metabolism. Damage to the pancreatic islets especially of β cells imposes assault on the metabolic function of the organ in glucose metabolism. Certain metabolic and diet-related disorders have been implicated in the disruption of the architecture of the pancreas such as diabetes mellitus, obesity and dyslipidaemia (Vessby 1994).

The photomicrograph of the pancreas of the diabetic and normal rats was closely examined. Under high power magnification light microscopic examination, pancreatic islet cells from NFG, NRG and NCG (normal control) rat exhibited normal histoarchitecture, namely

clearly visible darkly stained serous acini containing centroacinar cells. Degenerated islets with degranulation and vacuolization of β -cells were observed in pancreatic tissue from Diabetic rats fed with standard, low- and high-GI diets as seen in figures 25 – 27. These observations comply with the finding of other studies using alloxan to induce diabetes mellitus in animal models (Viana et al., 2004).

Regeneration of some islets cells observed in the photomicrograph of the pancreas in the DFG group raises some hope of improving the course of diabetes with good dietary recommendations (figure 27).

The lipid profile is a group of tests often ordered to determine the risk of coronary heart disease and other dietary lipid related disorder. They have proved to be good cardiovascular indicators of detecting onset of heart attack or stroke caused by vasocclusion and atherosclerosis.

In this study, results of the lipid profile as seen in Table 16 and 17 above clearly show that fried yam diet fed groups showed significant increase in total cholesterol (TC) level. Similarly, triglycerides (TG) and LDL show significant increase. Decrease in HDL level was observed as compared to control groups

The aim of diet therapy is to achieve normoglycemia and maintain ideal body weight. Dietary advice is primarily given in diabetes mellitus to avert symptoms of hyper- and hypoglycemia and to eliminate or postpone secondary complications, which may arise e.g. atherosclerotic cardiovascular disease due to micro- and macroangiopathy complication (Vessby, 1994). Thus, dietary recommendations in diabetes aim at normalizing blood glucose concentrations, serum insulin concentrations, blood lipid abnormalities and blood pressure

Dietary modification and restriction still remains a chief cornerstone in the prevention and management of DM as upsurge in the incidence and prevalence of diabetes worldwide is

noted to be on the high side (IDF, 2003). Therefore, while urgent action in the adoption of appropriate dietary management is considered, effect of processing on the glycemic impact should be borne in mind.

5.2 CONCLUSION

The main findings of this study are summarized below.

- i. Processing and the methods of preparation affected the nutrient composition of food with subsequent impact on the glycemic response to and the glycemic index of the processed foods
- ii. Processing of white yam-based diets increased calories, protein and fiber content of food significantly while effect on ash and moisture contents depends on the degree and extent of exposure to heat and method of processing.
- iii. Amala has the highest fiber and lowest lipid and calories and protein contents while fried yam has the highest calories, lipid and ash contents. However, roasted yam has the highest protein and lowest ash contents while Pounded Yam has the highest total carbohydrate content.
- iv. Processing and method of preparation delayed or improve the glycemic response with consequent increase or decrease in the glycemic index of carbohydrate-based foods.
- v. The pattern of glycemic responses to carbohydrate-based meal differs within and between the individuals as seen by the different incremental areas under the GR curves in diabetics and non-diabetic healthy subjects
- vi. The peak and the mean peak blood glucose responses were delayed and higher in the diabetics than the non-diabetic healthy subjects.

- vii. The glycemic indices of differently processed and prepared yam-based meals differ significantly with diabetics having higher values than the non-diabetic healthy subjects
- viii. In Diabetic and Non-diabetic healthy subjects, fried yam has the lowest GI while Roasted yam has the highest GI.
- ix. In Healthy subjects, all food items tested (except roasted yam) fall under Low-GI ($GI \leq 55\%$) foods according to Jenkins classification whereas in Diabetic subjects, Fried yam and Amala satisfied Low-GI classification while the rest fall under High-GI ($GI \geq 70\%$) foods.
- x. The GIs obtained showed wide inter- and intra- individual variability.
- xi. Food processing generally (except in few as observed in Amala) increases the GI and digestibility with subsequent rapid GR as evidenced by the incremental area and peak blood glucose concentration.
- xii. High-GI diets and Dietary fat intake contributes significantly to increase in total body weight thus establishing relationship between weight and dietary fat and high carbohydrate-rich foods
- xiii. There is no significant change in organ weight with respect to low- and high-GI diets.
- xiv. There is a reduction in glycemic tolerance and delayed glycemic response in low-GI diets fed rats as compared to the normal and high-GI fed rats.
- xv. In the diabetic rats there is disruption of the normal architecture of the pancreatic islets photomicrograph as compared to the normal non-diabetic rats. Although, some regenerations of the islets cells were observed in the diabetic rats on low-GI diets.
- xvi. In this study, results of the lipid profile clearly show that fried yam fed groups showed significant increase in total cholesterol (TC) level. Similarly, triglycerides (TG) and LDL

show significant increase. Decrease in HDL level was observed as compared to control groups

On the basis of the above findings and observations, it can be concluded that food processing and preparation favourably or adversely affect the glycemic response and the glycemic index with inter- and intra- individual variability through alteration of the nutrient composition and physical structure of the food during processing. In addition, Low-GI diets poses little risk for obesity and raises some hope of improving the course of diabetes with good dietary recommendations although prolonged consumption of fatty food should be discouraged because of the long term detrimental effect on blood lipid profile.

Statistically significant differences in the GIs of the test meals between the Diabetic and the Non-diabetic Healthy subjects showed that dietary caution should be indicated when recommending appropriate dietary management plan for the Diabetics.

Relative to the degree, extent and nature of processing, not all yam-based food products should be encouraged in diabetic patients. Those with Low-GI should be encouraged while those with High-GI should be discouraged according to the recommendation of WHO/FAO.

Dietary advice regarding the content and type of dietary carbohydrate in the diabetic diet has to be individualized depending on the degree and characteristics of the metabolic dysfunction. Although, low-GI foods have proved favorably in the dietary management and control of type 2 diabetes mellitus as shown and supported by different research findings.

Dietary modification and restriction still remains a chief cornerstone in the prevention and management of DM as upsurge in the incidence and prevalence of diabetes worldwide is noted to be on the high side (IDF, 2003). Therefore, while urgent action in the adoption of

appropriate dietary management is considered, effect of processing on the glycemic impact should be regarded.

Increase interest in further studies on the effect of other types of food processing on carbohydrates especially yam and yam-based meals with other tubers may be expected as they are necessary to establish a data-based nutritional table of GI of local Nigerian foods thus ascertaining their suitability or otherwise in their incorporation into the recommended menu for the dietary management of diabetes. Therefore, adequate facilities and equipments should be made available to fulfill such demands.

5.3 LIMITATIONS

The cost and time of carrying out the study was highly demanding and expensive. Thus, other processing methods employed conventionally could not be applied in this study. Other studies such as insulin assay and glycated haemoglobin concentration measurement could not be carried out due to lack of fund and other logistic reasons.

5.4 RECOMMENDATIONS

To facilitate the choice of the most suitable processing and preparatory method(s) favouring low-glycemic load diets for the diabetic patients, more data is needed regarding other food types and the effect of various processing methods on the glycemic response and the glycemic index of foods in our environment.

This should include information on the total carbohydrate content, calories supplied and the glycemic index values.

Fried yam and Amala may be usefully incorporated into diets of type 2 diabetic patients. Boiled yam should be occasionally considered while Roasted yam and Pounded yam should be

discouraged on regular daily meal as revealed by the rapid glyceimic response with resultant hyperglycemia. In the case of hypertensive diabetics, caution should be taken when consuming fatty foods.

Adding of acidic extra such as lemon juice or a little original olive oil (which delays or retards gastric emptying) to diet may help in reducing the GI value of a meal thus improving the glyceimic profile.

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APPENDIX A: UCH/UI IRC ETHICAL APPROVAL



INSTITUTE FOR ADVANCED MEDICAL RESEARCH AND TRAINING (IMRAT)

COLLEGE OF MEDICINE, UNIVERSITY OF IBADAN. IBADAN, NIGERIA.

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Ag. Director : Prof F. A. A. Adeniyi



UI/UCH IRC Registration Number: **Pending**
: Date: **17/12/2007**

NOTICE OF FULL APPROVAL AFTER FULL COMMITTEE REVIEW

RE: Effect of Food Processing on the Glycemic Response and the Glycemic Index of Foods in Diabetic and Non-Diabetic Nigerian Subjects: A Study of Yam (*Dioscorea rotundata*).

UI/UCH Ethics Committee assigned number: UI/EC/07/0092

Name of Principal Investigator: Dr. M. M. C. Anyakudo

Address of Principal Investigator: Department of Physiology,
University of Ibadan, Ibadan

Date of receipt of valid application: 03/09/2007

Date of meeting when final determination of research was made: 19/11/2007

This is to inform you that the research described in the submitted protocol, the consent forms, and other participant information materials have been reviewed and *given full approval by the UI/UCH Ethics Committee.*

This approval dates from 17/12/2007 to 16/12/2008. If there is delay in starting the research, please inform the UI/UCH EC so that the dates of approval can be adjusted accordingly. Note that no participant accrual or activity related to this research may be conducted outside of these dates. *All informed consent forms used in this study must carry the UI/UCH EC assigned number and duration of UI/UCH EC approval of the study.* In multiyear research, endeavour to submit your annual report to the UI/UCH EC early in order to obtain renewal of your approval and avoid disruption of your research.

The National Code for Health Research Ethics requires you to comply with all institutional guidelines, rules and regulations and with the tenets of the Code including ensuring that all adverse events are reported promptly to the UI/UCH EC. No changes are permitted in the research without prior approval by the UI/UCH EC except in circumstances outlined in the Code. The UI/UCH EC reserves the right to conduct compliance visit to your research site without previous notification.



Prof. C. A. Adebamowo
Chairman, UI/UCH EC
E-mail: uiuchirc@yahoo.com

Research Units: ■Genetics & Bioethics ■Malaria ■Environmental Sciences ■Epidemiology Research & Service

APPENDIX B: INFORMED CONSENT FORM

INFORMED CONSENT FORM (UI/EC/07/0092)

My name is Dr. Michael Magnus Anyakudo . I am a postgraduate student of the Department of Physiology, College of Medicine University of Ibadan.

I am conducting a research to determine the effect of food processing on the glycemic response and the glycemic index of foods such that the information derived may help in: (i) identifying processing method(s) that favours good diabetic control via dietary therapy (ii) providing a database of glycemic index of common Nigerian foods and (iii) contributing to the existing knowledge on the above thesis.

Please note that your name will not be written on the form. You will only be given a number. The information obtained at the end of this research will be provided to the diabetologists and dieticians to help in the dietary management of diabetes.

During this exercise, you will be requested to participate in the ingestion of various food provided after overnight fasting. There after, four blood samples (about a drop) will be taken at interval of 30 minutes by finger prick using fine disposable lancet. The process of taking the specimen will not cause you any harm or injury. These specimens will be used to determine the glycemic index of the various foods provided.

Your full participation will help to conduct this research with minimal error. You are free to refuse to take part in this programme. You have a right to withdraw at any given time if you choose to.

I will greatly appreciate your help in taking part in this study.

CONSENT

Now that the study has been well explained to me and I fully understand the content of the study process, I will be willing to take part in the programme.

Signature/Thumbprint of Participant/Date

Signature of Interviewer/Date