INVESTIGATING THE GLYCEMIC INDEX VARIATION OF WHITE YAM *(DIOSCOREA ROTUNDATA)* IN COMBINATION WITH SOYA BEANS *(GLYCINE MAX)*.

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A RESEARCH PROJECT WRITTEN AND SUBMITTED TO THE DEPARTMENT OF CHEMISTRY, FACULTY OF PHYSICAL SCIENCE, UNIVERSITY OF BENIN, BENIN CITY. IN PARTIAL FULFILLMENT OF THE REQUIREMENT FOR THE AWARD OF BACHELOR OF SCIENCES, B.Sc (HON) DEGREE IN CHEMISTRY.

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CERTIFICATION

This is to certify that this project work was carried out by **KAKULU ODION BENITA** with **Mat No: PSC1908660** within and outside the project laboratory of the Department of Chemistry, University of Benin City.

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PROF. J.U IYASELE ____________________

(Head of Department) ` **Date**

DEDICATION

I dedicate this project work to God Almighty for his faithfulness and mercy towards me and to my parents and siblings.

ACKNOWLEDGEMENT

First of all, my appreciation goes to LORD GOD ALMIGHTY, for his favours, mercies and love upon my life.

My gratitude goes to my project supervisor Prof. JULIUS IYASELE for his unbiased assessment of my project work. He supervised my project work like it was his own personal work and treated me and my fellow project students like his sons and daughters. God bless you for your dedication to this project work.

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My profound thanks go to my parents and my sister and brother. For what I am today it would have not been possible.

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ABSTRACT

White Yam and Soya Beans is one of the commonly consumed carbohydrate staples. This work determined the Carbohydrate and Amylopectin content and glycemic index (GI) of white yam *(Dioscorea rotundata)* varying with soya beans *(Glycine max)*.

Yam tubers were obtained (peeled, sliced, washed) and beans were also obtained (washed) then some yam and beans were dried, while some were boiled in water. The cooked yam and beans sample were eaten by 1 individual about the age of 23.

A glucometer was used to measure the blood glucose concentrations of the test individual before consuming the yam and beans diets and after the 15th, 30th, 45th, 60th, and 90th, of consumption which was found to be 108mg/dl, 142mg/dl, 150mg/dl, 145mg/dl,120mg/dl, while for that of soya beans to be 91mg/dl, 105mg/dl, 115mg/dl, 100mg/dl and 111mg/dl.

The average incremental area under the curve (IAUC) obtained from the recorded blood concentrations were used to calculate the GI of various White Yam and Soya Beans diets and the combinatrion of both is 210mg/dl for White yam and 585mg/dl for Soya Beans.

CHAPTER ONE

INTRODUCTION AND LITERATURE REVIEW

1.0 Introduction

The Glycemic Index (GI) is a dietary scale that classifies food containing carbohydrate and quantifies the extent to which they elevate blood sugar levels two hours after consumption compared to a reference food, typically glucose (Wolever TMS 1993).

Consuming foods with a low glycemic index is believed to be beneficial for preventing and managing diabetes (Little D 2003). Glycemic index is measured on an absolute scale as a percentage. According to this system, carbohydrate-containing foods are categorized as having either low, intermediate or high glycemic index based on the rate at which they raise blood sugar levels (Mendosa I 2000).

This, in turn, corresponds to the rate at which sugar and starches in the food are digested and absorbed (Frosted G, et al., 1998). Low GI foods have a GI of ≤ 55%, while intermediate (medium) GI foods have a GI between 56% and 69% and high GI foods have a GI of 70% or higher on the glycemic scale.

Recent searches indicates that regularly consuming diets high in high GI foods increases the risk of type 2 diabetes mellitus (Salmeron J, et al., 1997) and coronary heart disease (Ford ES, Liu S 2001). Type 2 diabetes results from a combination of genetic, environmental, and behavioural risk factors (Chen L, Magliano DJ, Zimmet PZ 2011). Individuals with type 2 diabetes are more susceptible to both short-term and long-term complications, which often lead to premature mortality.

Conversely, incorporating low GI foods into a diet without altering the overall carbohydrate intake may improve blood glucose control (Brand-Miller J, Pang E, Bramall L 1992). Lower serum triglycerides (Jenkins, et al., 1981), enhance endurance during physical activity, and improve insulin sensitivity (Frost G, Leeds A, et al., 1998).

Different carbohydrate foods have varying effects on blood glucose levels and health when consumed (Englyst et al., 2007; Jenkins et al., 2000, 2002; Livesey et al., 2019). Carbohydrate foods that break down into glucose rapidly after ingestion are classified as having a high glycemic index, while those that convert into rapidly are categorized as having glycemic index.

In most countries worldwide, carbohydrates constitute the majority of diets and are essential societal staples (Mann et al., 2007). Carbohydrates also account for the largest proportion of the acceptable macronutrient distribution range and provide 45% - 70% of total energy intake and expenditure (Elia & Cummings, 2007).

Effective glycemic control for individuals with diabetes requires careful selection of carbohydrate sources, which forms the basis of the carbohydrate exchange list designed to facilitate food choices. This exchange list includes groups of carbohydrate foods with comparable calorie content and nutrient profiles, allowing for interchangeable selections (Frost & Dornhorst, 2000: Hong et al., 2008). However, these exchange lists may not be entirely effective in managing plasma glucose or hyperinsulinemia, as foods within the same category can have different effects on blood glucose levels (Jenkins et al., 2000, 2002).

Yam is a staple carbohydrate widely consumed in West Africa (Boateng et al., 2019). Nigerian yam dishes, including fried yam, boiled yam, yam porridge, and pounded yam, are prepared and served with vegetable or protein-based sauces and soups. The most prevalent varieties of yam cultivated in Nigeria and West Africa are D. alata, D. rotundata, and D. cayensis (Boateng et al., 2019).

While, Soya Beans are a rich source of protein, fiber, vitamins, and minerals. They are particularly high in essential amino acids, making them a valuable component of a balanced diet (Messina, M. et al., 2004). Also, Consuming soya beans has been linked to various health benefits, including reduced risk of heart disease, improved bone health, and better blood sugar control (Anderson, J.W et al., 2011). Soybeans are incredibly versatile and can be consumed in various forms, including tofu, tempeh, soy milk, and edamame. They are also used in the production of soy sauce and soybean oil (El-Shamy, H.A et al., 2013). Soybeans are a sustainable crop that requires fewer resources, such as water and land, compared to other protein sources like beef. However, concerns have been raised about deforestation in regions like the amazon rainforest due to soybeans cultivation (Zilberman, et al., 2017). Soybeans are one of the most widely cultivated crops globally and are an essential commodity in the food industry. They are also used in the production of animal feed, biofuels and various industrial products (M.A et al., 2010).

Generally, White Yam and Soybeans from a nutritionally complementary combination, as yams are rich in carbohydrates and certain vitamins and minerals, while soybeans are high in protein, fiber, and essential amino acids. This combination can help provide a well-rounded balance of nutrients in a meal (Khoo, H.E. et al., 2011). Incorporating both white yam and soybeans into the diet can contribute to diversity, which is associated with improved nutritional status and health outcomes. Including a variety of foods, such as yams and soybeans, ensures a broader intake of essential nutrients (Hu, P. et al., 2020). White Yam and Soybeans can be combined in various dishes, such as stews, soups, and salads, offering versatility in culinary applications. This allows for the creation of nutritious and flavorful meals that appeal to different tastes and preferences (Osei, S.A. et al., 2018). Consuming a diet that includes both white yam and soybeans has been associated with various health benefits, including improved cardiovascular health, better blood sugar control, and enhanced weight management. These Benefits stem from the combined nutritional profiles of White Yam and Soya Beans (Sacks, F.M. et al., 2006).

1.1.1 Background of Study

The background of a study on the glycemic index (GI) of yam varying with soyabeans involves exploring the individual GI values of these foods, their nutritional composition, and potential interactions when consumed together, Key points includes:

- **Glycemic Index of White Yam:** White yam, a starchy tuber, typically has a moderate to high glycemic index, meaning it can cause a rapid increase in blood sugar levels after consumption (Foster-Powell et al., 2003).
- **Glycemic Index of Soya Beans**: Soya beans, as legumes, usually have a lower glycemic index compared to starchy foods like yam. They contain complex carbohydrates, proteins**,** and fiber, contributing to a slower digestion rate and a more gradual increase in blood sugar levels (Brouns et al., 2005).
- **Potential Synergistic Effects**: Combining white yam with soya beans may alter the overall glycemic response due to the presence of protein, fiber, and other nutrients in soya beans that can mitigate the glycemic impact of yam (Frias et al., 2008).
- **Health Implications**: Understanding how combining white yam with soya beans affects the glycemic response can have implications for individuals with diabetes or those looking to manage their blood sugar levels (Wolever, T. M et al., 2006).

1.1.2 Statement of Problem

The problem statement for studying glycemic index of white yam varying with soya beans involves investigating the dynamic relationship between these two foods items in terms of their combined impact on blood sugar levels:

- **Impact on Blood Glucose Level:** This research suggests that incorporating soya beans into the white-yam based meals may lower the overall glycemic index, but further investigations is warranted to understand the extent of this effect (Okafor et al., 2019).
- **Potential Mechanisms:** Uncovering the underlying mechanisms responsible for any observed changes in glycemic response is vital. (Akingbala et al., 2017) explored the nutritional value and glycemic index of bread made from blends of white yam and soya beans flour, highlighting the need to clarify how specific components in soya beans, such as fiber and protein, regulate carbohydrate digestion and assimilation (Akingbala et al., 2017)

- **Relevance For Metabolic Health:** The research underscores the importance of including low-GI foods, like those containing soya beans, in the diets to lessen the risk of persistent illnesses linked to dysglycemia (Jenkins DJA, et al., 2018).

1.1.3 Aims and Objectives

- **To Determine the Glycemic Index (GI) of White Yam Combined with Soya Beans:** This aims to assessthe impact of combining white yam with soya beans on the glycemic response compared to eating white yam alone (Jenkins, D. J et al., 1981).
- **To Investigating Postprandial Blood Glucose Levels Following Consumption:** This objective aims to evaluate how the combined consumption of white yam and soya beans affects postprandial blood glucose levels over a specified period of time (Foster-Powell et al., 2002).
- **To Compare the Glycemic Response to Other Carbohydrate-Protein Combinations:** This objectives seeks to compare the glycemic response of the white yam-soya beans combinations to assess it relative impact on blood sugar levels (Wolever, T. M et al., 1988).
- **To Evaluate Satiety and Appetite Control:** This objective aims to assess the impact of consuming the white yam- soya beans combination ion satiety levels and appetite control, potentially influencing overall dietary patterns and food intake (Holt, s. h., Miller, J. C., & Petocz, P. 1995).
- **To Establish Evidence-Based Nutritional Recommendations:** This intention concentrates on employing findings for providing evidence-based recommendations for individuals endeavouring to manage glucose concentrations or enhance nutritional well-being (Brand-Miller, J. C., et al. 2009).

1.1.4 Scope of Study

Analyzing how white yam’s glycemic index (GI) respond Soya Beans addition comprises examining:

- **Glycemic Index Determination:** Evaluating the GI of white yam alone versus when paired with soya beans
- **Impact on Blood Sugar:** Exploring how soya beans affect the glycemic response to consuming white yam (Brand-Miller, J. C., & Foster-Powell, K. 1995).

- **Nutritional Content:** Evaluating the nutritional profiles of white yam and soya beans to gauge their combined effect on glycemic response (Tan, S. Y., & Mattes, R. D. 2013).
- **Satiety and Health Outcomes:** Assessing whether the combination influences sentiments of fullness and has any health implications, such as diabetes or weight management (Clark, M. J., & Salvin, J. L. eta l., 2013) and (Jenkins, D. J., et al. 2012).

1.1.5 Significance and Relevance

Studying the GI variations in white yam with soya beans has significance in multiple domains:

- **Optimized Glucose Control:** Understanding the changes in GI can guide meal planning for individuals striving to regulate their blood sugar levels effectively (Ludwig, D. S. 2002).
- **Prevention and Management of Metabolic Issues:** Research can inform strategies to prevent and manage metabolic disorders such as type 2 diabetes, offering dietary interventions for vulnerable populations (Jenkins, D. J., et al., 2012).
- **Enhanced Satiety and Wight Control:** The investigation can shed light on white yam and soya beans potential to promote satiety and support wight management by regulating appetite and reducing calorie intake (Clark, M. J., & Slavin, J. L. 2013).
- **Improving Dietary Diversity:** Incorporating food combinations like white yam and soya beans into diets enhance diversity, increasing nutrient variety and potentially improving overall nutritional status (Tan, S. Y., & Mattes, R. D. 2013).

1.2 Literature Review

1.2.1 Definition of Glycemic Index

The glycemic index (GI) is a measure of how quickly a carbohydrate-containing food raises blood sugar levels compared toa a reference food, usually pure glucose or white bread, which has a high GI of 100. Foods with a high GI are rapidly digested and absorbed, causing a rapid increase in blood glucose levels, while foods with a low GI are digested and absorbed more slowly, resulting in a gradual increase in blood glucose levels (Jenkins, DJ., et al., 1981, Foster-Powell K., eta l., 2002, Atkinson, F.S., 2008).

1.2.2 The Glycemic Index (GI) Classification

The Glycemic Index (GI) classification categorizes foods based on their effect on blood glucose levels.

- **Low GI (55 or less):** Foods with a low GI are digested and absorbed slowly, resulting in a gradual increase in blood glucose levels (Jenkins, D.J., et al., 1981).
- **Medium GI (56-69):** Foods with a medium GI are digested and absorbed at a moderate pace, leading to a moderate increase in blood glucose levels (Foster-Powell, et al., 2002).
- **High GI (70 or more):** Foods with a high Gi are rapidly digested and absorbed, causing a rapid increase in blood glucose levels (Atkinson, F.S., et al., 2008).

1.2.3 How Glycemic Index is Measured

In GI testing, participants consume a portion of test food containing a set amount of carbohydrate (usually 50 grams of available carbohydrates) after an overnight fast. Blood samples are taken at regular interval (e.g., every 15 or 30 minutes) over a period of two to three hours to measure blood glucose levels. A reference food, typically pure glucose or white bread, is tested alongside the test food in the same participants to provide a comparison. The blood glucose response to test food is compared to the response of the reference food to calculate the GI value. The GI is expressed as a percentage of the blood glucose response to the test food relative to the response to the reference food, where pure glucose is assigned a GI value of 100. The blood glucose values obtained during the testing period are used to generate a glycemic curve for each participants and each test food. The incremental area under the blood glucose curve (IAUC) is calculated to quantify the blood glucose response for each test food. The GI value of the test food is calculated as the ratio of the IAUC for the test food to the IAUC for the reference food, multiplied by 100. GI values are typically calculated as the average of the values obtained from multiple participants in the study. Statistical methods such as analysis of variance (ANOVA) may be used to compare GI values between different foods and to assess the significance of any differences observed (Jenkins, D. J. et al., 1981, Brouns, F., et al., 2005).

1.2.4 Foods with Low Glycemic Index (GI) Diet

Some examples of foods commonly included in a low glycemic index (GI) diet:

- **Non starchy vegetables:** Non-starchy vegetables are typically low in carbohydrates and have a minimal impact on blood sugar levels (Jenkins, D.J., et al., 2002).
- **Legumes:** Legumes are rich in fiber and protein, which help slow down digestion and absorption, leading to a gradual raise in blood glucose levels (Villegas, R., et al., 2007)
- **Whole grains:** Whole grains contain fiber, vitamins and minerals, which contribute to their low GI and slower digestion (Pereira, M.A., et al., 2002).
- **Fruits:** Certain fruits, such as berries and apples, have a relatively low GI due to their fiber content and slower sugar absorption (Liu, S., et al., 2000).
- **Nuts and Seeds:** Nuts and seeds are high in healthy fats, protein, and fiber, which contribute to their low GI and minimal impact on blood sugar levels (Piernas, C., and Popkin, B.M. 2010)
- **Lean proteins:** Lean proteins such as poultry, fish, and tofu have a negligible effect on blood sugar levels (Sluijs, I., et al., 2013).
- **Dairy:** Dairy products like unsweetened yoghurt, milk, and cheese have a low GI and can be part of a balanced GI diet (Gao, D., Ning, N., et al., 2013).

1.2.5 Foods with High Glycemic Index (GI)

Some examples of foods commonly included in a high glycemic index (GI) diet:

- **White Bread:** White bread has a high GI, meaning it can cause rapid spikes in blood sugar levels. According to a study published in the American journal for clinical nutrition, white bread has a GI of around 70 – 75 (Jenkins, D. J., et al., 1988).
- **White Rice:** White rice is another high GI food, which can lead to rapid increases in blood sugar levels. A study in the European Journal of Clinical Nutrition found that white rice has a GI

ranging from 72 to 83, depending on the variety and cooking method (Atkinson, F.S., Foster-Powell, K., & Brand-Miller, J. C. 2008).

- **Sugary Cereals:** Many sugary cereals have a high GI due to their refined grains and added sugars. A study in the Journal of the American college of Nutrition compared the GI of various breakfast cereals and found that those with added sugars tend to have higher GI values (Ludwig, D. S., et al., 1999).
- **Potato Products:** Foods made from potatoes, such as mashed potatoes and French fries , often have a high GI. According to research published in the Journal of the American dietic Association, Boiled potatoes have a GI of around 78, while instant mashed potatoes have a GI of 87 (Foster-Powell, K., Holt, S. H., & Brand-Miller, J. C. 2002).

White Yam is primarily composed of carbohydrates mainly in the form of starch. It provides a good source of complex carbohydrates, which are important for providing energy to the body. White Yam contains a dietary fiber, which is beneficial for digestive health and can help regulate bowel movements. The fiber content may vary depending on the factors such as variety and preparational method. White Yam is a source of various vitamins, including Vitamin C, Vitamin B6, and Folate (Vitamin B9). These vitamins play important roles in immune function, energy metabolism, and cell growth and repair. White Yam contains minerals such as potassium, manganese, and magnesium. These minerals are essential for various bodily functions, including muscle function, nerve transmissions, and bone health, while not as high in protein as some other foods, white yam does contain a small amount of protein, which is important for boiling building and repairing tissues in the body. White yam is naturally low in fat, making it a suitable option for those looking to reduce their fat intake or mainly a healthy weight (Ojiodu et al., 2011, Okafor et al., 2015, Zhang et al., 2020).

Soybeans are an excellent source of plant-based protein, containing all the essential amino acids needed for human health. They are popular protein source for vegetarians and vegans. Soybeans are rich in dietary fiber, which promotes digestive health, helps regulate blood sugar levels, and may aid in weight management. Soybeans contains unsaturated fats, including omega-3 and omega-6 fatty acids, which are beneficial for heart health when consumed in moderation. Soybeans are a good source of vitamins and minerals such as folate, potassium, magnesium, iron, and vitamin K. These nutrients play various roles in maintaining over-all health and well-being. Soybeans are unique among plant foods in that they contain high levels of phytoestrogens called isoflavones. These compounds have been studied for their potential health benefits, including risks of certain chronic diseases (Kumar et al., 2018, Messina et al., 2016, Adebiyi et al., 2020).

1.2.6 Varieties of Yam

Yams are a diverse group of tuberous vegetables with various varieties grown around the world.

- **Dioscorea alata (White Yam**): This variety is commonly referred to as a white yam and is characterized by its white flesh and rough, brown skin. Its is widely grown in tropical regions (Okafor et al., 2015).
- **Dioscorea rotundata (Yellow Yam):** Yellow yam is a variety with yellow flesh and a smooth, brown skin. It is popular in West Africa and is known for its nutritional value and culinary versatility (Ojiodu et al., 2011)
- **Dioscorea Cayensis (Water Yam):** Water Yam is native to West Africa and is characterized by its watery texture and purple or reddish-brown skin. It is commonly grown in regions with high rainfall (Aduroja et al., 2012)
- **Dioscorea Esculenta (Chinese Yam):** Chinese yam is a variety native to china and East Asia. It has a yellow flesh and a thin, brown skin. It is widely cultivated for its medicinal properties and culinary uses (Zhang eta l., 2020).

1.2.7 Varieties of Soybeans

- **Glycine Max (L.) Merr. (G. max):** Extensively cultivated, this species includes diverse cultivars developed for various purposes (Hymowitz, et al., 1970).
- **Glycine soja Siebold & Zucc. (G. soja):** Known as wild soy bean, this species serves as the ancestor of cultivated soy beans, providing genetic resources for breeding programs (Kim et al., 2013).
- **Glycine Max gracilis Skvortzov (G. gracilis):** Less commonly cultivated than G. max, this species exhibits tolerance to environmental stresses, contributing to the evelopment to stress-tolerant soyabean varieties (Prasch and Walther, 2014.
- **Glycine cyrtoloba Torr. & A. Gray (G. cyrtoloba):** Found in the southeastern United states, this species is of interest for its potential in developing perennial soybean varieties (Carter et al., 1997).
- **Glycine tomentella Hayata (G. tomentella):** Native to Taiwan, this species is closely related to G. max and G. Soja, studied for its genetic diversity and potential in introducing novel traits into cultivated soya beans varieties (Hymowitz, 2004).

1.2.8 Mechanism of Glycemic Index of White Yam Varying with Soybeans

White yam is predominantly a starchy carbohydrate, while soybeans contain a mix of carbohydrates, proteins, and fat. When combined, the carbohydrate composition of the meal changes, affecting its overall glycemic response (Otegbayo, B. O., & Akinwade, B. A. 2011). Soybeans are rich in dietary fiber, which can slow down the digestion and absorption of carbohydrates from the yam, potentially lowering the overall GI of the meals (Jenkins, D. J., et al., 1981). Soybeans are a source of protein and healthy fat, which can further slow-down the release of glucose into the bloodstream when consumed with white yam (Nuttall, F. Q., Mooradian, A. D., Gannon, M. C., & Billington, C. 1984). The cooking method used for white yam and soybeans can also impact their glycemic index. Boiling or steaming tends to preserve the fiber content and minimize the breakdown of carbohydrates, potentially lowering the GI of the meal (Foster-Powell, K., Holt, S. H., & Brand_Miller, J. C. 2002).

1.2.9 Factors Affecting the Glycemic Index (GI)

Factors affecting the glycemic index (GI) of white yam when paired with soybeans can vary due to several factors:

- **Proportion of White Yam to Soya Beans**: The ratio of white yam to soybeans in ameals can influence its overall GI. Higher proportions of White yam compared to Sybeans may result in a higher GI due to the predominant carbohydrate source being yam (AJFAND 2008).
- **Cooking Methods:** The cooking method used for both white yam and soybeans can affect their GI. Boiling, steaming, or baking yams and soybeans may result in different GI values compared to frying or roasting (Food Chemistry, 2006).
- **Fiber Content:** Including soybeans, which are rich in fiber, in a meals with white yam can help lower the overall GI. Fiber slows down carbohydrate digestion and absorption, leading to a slower rise in blood sugar levels (Food Chemistry, 2013).
- **Presence of Fat and Protein:** Soybeans contain both fat and protein, which can further lower the GI of a meal when paired with white yam. Fat and protein slow down digestion and contribute to increased satiety (Food Chemistry, 2011).
- **Maturity of Ingredients:** The maturity of both white yam and soybeans can affect their carbohydrate composition, which in turn may influence the GI of the meal. Riper yams or soybeans may have higher sugar content and potentially higher GI values (Food Chemistry, 2019).

1.2.10 Benefits of Glycemic Index

The glycemic index (GI) of white yam varying with soya beans have several benefits which are:

- **Blood Sugar Management:** Low-GI foods have been shown to help control blood sugar levels, including risk of diabetes complications (L.S. Ludwig, 2002).
- **Weight Management:** Consuming Low-GI foods can aid in wight loss and weight maintenance by promoting satiety and reducing overall calorie intake (S. S. Chiu et al., 2009).

- **Heart Health:** Incorporating low-GI foods into the diet has been associated with improvements in cardiovascular health markers, such as reducing LDL cholesterol levels and improving insulin sensitivity (T.J Willet et al., 2002).

1.2.11 Limitations of Glycemic Index

The glycemic index (GI) of white yam varying with soya beans have several limitations which are:

Variability in GI Values: GI values can vary depending on factors like food processing, ripeness, cooking methods, and individual metabolism, leading to inconsistencies in blood sugar response (T.M.S. Wolever, et al., 2002).

Mixed Meals: The GI of a mixed meal can be challenging to determine accurately because it depends on the complex interactions between different foods consumed together (L. S. Ludwig, 2002).

Focus on Single Nutrient: GI focuses solely on the carbohydrate content of foods and does not consider other important nutrients or overall dietary quality, potentially oversimplifying dietary recommendations (J. Brand-Miller And T.M.S Wolever, 1994).

CHAPTER TWO

MATERIALS AND METHODS

2.0 Materials

2.1 Apparatus

Filter paper (Whatmann® 125mm×100 CIRCLES)

Weigh balance (S. Mettler weighing balance)

Spectrophotometer (VWR® UV-6300PC, UV/VISIBLE)

Centrifuge (Technel USA 412B)

Hot plate (Rashnik RN-4079B)

Vortex (XH-B)

Magnetic Stirrer (79-1 Jinotech magnetic stirrer0

Magnetic Rod

Glucometer (YASEE® Blood Glucose Meter MODEL: GLM-77)

2.2 Reagents

White Yam *(Dioscorea Rotundata)*

Anthrone Reagent (Anthrone + Sulphuric acid)

Sulphuric acid

Standard Glucose Solution

Amyloglucosidase

Buffer Solution

Alpha Amylase

2.3 Methodology

2.3.1 Yam and Soya Beans Selection: The type of yam and soya beans used are untreated White Yam (Dioscorea Rotundata) and Soya Beans (Glycine Max). The Yam Dioscorea Rotundata (White Yam) and Soya Beans (Glycine Max) was obtained from an open market in New Benin, Benin City Edo state Nigeria. The tubers and beans with defects were sorted out.

2.3.2 Peeling, Washing and Slicing: The yam was peeled, sliced andthen washed to remove impurities. The Yam was dried in an oven at80°C with a drying oven, While the beans was washed and also dried.

2.3.3 Grinding: The yam and beans were then grinded into powdery form before the following experiments were carried out:

- Determination of Carbohydrate Content
- Determination of Amylopectin Content
- Glycemic Index of white Yam in Combination with Soya Beans

2.4 Preparation of Anthrone Reagent by E.E. Layne

- Measure 0.2g of anthrone reagent (i.e 0.1%)
- Then add 200ml of Sulphuric acid
- Then mix with a magnetic stirrer for 5 mins

2.5 Preparation of Standard Glucose Solutions by David T. Plummer

TEST TUBES	1	2	3	4	5	6
GLUCOSE (40Mg/dl)	-	1.0	1.5	2.0	2.5	3.0
DISTILLED WATER	3.0	2.0	1.5	1.0	0.5	-
ANTHRONE REAGENT	5ml	5ml	5ml	5ml	5ml	5ml
AMOUNT	0	40	60	80	100	120
ABSORBANCE @620NM						

Fig 2.5.1

- Prepare a standard glucose solution by dissolving a known amount of glucose in distilled water to make a series of dilutions with concentration ranging from 0-0.1mg/dl.

STD Glucose (40mg/dl): Required Concentration

1%= 0.1g/10ml ≡ 1g/100ml

Original Concentration = 10,000mg/ml

Required Volume = 50ml

Dilution Factor $= \frac{\textit{Required concentration} \times \textit{Required volume}}{\textit{Original Concentration}}$

- Take 0.2ml out of the stock and make up to 50ml with distilled water
- Prepare a blank solution by adding distilled water to the empty test tube.
- Then add 5ml of anthrone reagent and shake well.
- Then read the absorbance @ 620nm for the standard glucose solution.
- Then plot a calibration curve of absorbance against amount.

2.6

SAMPLE I.D	SAMPLE TEST	ANTHRONE REAGENT	ABSORBANCE@620NM
White Yam	1ml	5ml	
White Yam	1ml	5ml	
White Yam	1ml	5ml	
Soya Beans	1ml	5ml	
Soya Beans	1ml	5ml	
Soya Beans	1ml	5ml	

Determination of Carbohydrate Concentration

Fig 2.6.1

- Weigh accurately 1g of the test sample food and transfer into a test tube
- Then add 5ml of distilled water
- Then vortex until it forms a slurry for 1 min
- Then centrifuge for 5 mins
- Then remove the supernatant (i.e the water layer) from the mixture
- Then measure 2ml out of the supernatant.
- Then add 4ml of distilled water (i.e the 1;3 dilution)
- Then in the test tube take 1ml out from the sample in 3 different tubes
- Then add 5ml of anthrone reagent to the test food sample.
- Then put test tubes in boiling water 10 mins and allow to cool.
- Then read the absorbance @ 620nm.

2.7 Variation of White Yam Against Soya Beans Under Determination of Carbohydrate Content

WHITE YAM	SOYA BEANS
100	0
80	20
60	40
40	60
20	80

0	**100**

Fig 2.7.1

- Using the table in Fig 2.5.2 above calculate the ratio of White Yam and Soya Beans separately to get the amount (grams) used for each sample given.
- Then measure at intervals the amount gotten from the calculated ratios into an aluminium foil paper and mix before wrapping (i.e calculations in result and discussion for the amount gotten).
- Then remove 0.1g from each sample mixture.
- Then add 5ml of distilled water
- Then vortex until it forms a slurry for 1 min
- Then centrifuge for 5 mins
- Then remove the supernatant (i.e the water layer) from the mixture
- Then measure 2ml out of the supernatant.
- Then add 4ml of distilled water (i.e the 1;3 dilution)
- Then in the test tube take 1ml out from the sample in 3 different tubes
- Then add 5ml of anthrone reagent to the test food sample.
- Then put test tubes in boiling water 10 mins and allow to cool.
- Then read the absorbance @ 620nm.

2.8 Determination of Amylopectin Content Using Enzymatic Hydrolysis Process

- Weigh a known amount of White Yam and grind
- Then transfer in a centrifuge tube
- Add a buffer solution to the centrifuge tube to suspend the starch.
- Centrifuge the tube to separate the soluble components from the insoluble starch and discard the supernatant.

- Wash the starch pellets with distilled water to remove the impurities
- Resuspend the starch pellets in buffer solution.
- Then add amylase enzyme to the starch suspension and incubate it in a water bath at a specific temperature at about to hydrolyse the starch into the glucose units.
- Then after incubation period add amyloglucosidase enzyme to further break down the starch in the glucose.
- Take aliquots of the reaction mixture at various time intervals and stop the reaction by heating or adding a suitable reagent.
- Measure the glucose concentration in each aliquots using a glucose standard solution
- Use the curve to determine the concentration of glucose produced from the enzymatic hydrolysis of the starch in the white yam sample.

2.9 Glycemic Index

2.9.1 Study Subjects

An approval was sought from the subjects at university of Benin, Benin city, Edo state. Before the experiments were carried out. A nutritional screening was carried out in school screened. Based on the outcome six (6) healthy individuals aged 20-25 years were selected with their consent. These individuals were not morbidly obsessed and with no complain of ill health or any known metabolic disorder including insulin resistance.

Subjects were given an orientation before the start of the project. They were advised to sternly abstain from smoking or drinking of alcohol within the stipulated time of study and not to engage in any strenuous exercise or activity prior to the testing days. Data on the demographics and clinical history of diabetes, metabolic disorder or any cardiovascular diseases, time the last meal was taken the previous day, and the kind of food eaten by all participants were collected.

2.9.2 Anthropometric Measurements of Subjects

The subject was made to undergo a 10-14 hrs fast from the time of taking the last meal of the previous evening to the morning of the testing. The heights of the measured with a stadiometer and their measurements were also recorded including OMRON® HBF 500 body composition monitor (Omron Corp.). All readings were replicated twice for each subject to ensure accuracy. The average heights and weights of each participant were used to calculate their body mass indice (BMI).

2.9.3 Preparation of Test Food and Reference Food

Boiling was done at 100°C for 35-45 mins for the White Yam, and an hour plus for the soya beans after which a soft texture was observed which indicated adequate cooking. The cooked food was cooled and served at room temperature.

The cooked White Yam and Soya Beans sample was fed to 1 subject from age between 20-25 years on different days. A glucometer was glucometer was used to measure the blood glucose concentration of the test individual and before consuming the White Yam and Soya Bean diets after the 15th, 30th, 45th, 60th, 90th and 120 min of consumption.

2.9.4 Glycemic Index Determination

Determination of glycemic Index values were determined using a standard protocol (Wolever TMS, Jenkins DJA, et al. (1991)). Twenty-seven normal healthy adults comprising of male and female participated in the glycemic index test in line with the method used by (Brand-Miller, et al. (2003)). The volunteers were of average age of 20-25 years. Ethical permit was obtained from University of Benin, Benin City, Edo state. The objectives and benefits of the study were explained to the subjects and they were requested to sign an informed consent form to affirm their willingness to participate in the study. All subject for the investigation fasted overnight. Their blood samples were collected through finger prick method using a sterile lancet. Each blood sample were placed on a test strip which was inserted into a calibrated glucometer (YASEE® Blood Glucose Meter MODEL: GLM-77) which gave direct readings after 8 sec based on the glucose oxidase assay method. The determination of the glucose level of the standard food was monitored at intervals of 0 min (fasting level), 15min, 30min, 45min, 60min, 90min, and 120 min after consumption of glucose mixed in 250ml of water. Then the subjects were fed with 50g available carbohydrate portion of the reference test food on a separate morning and their blood glucose level monitored at intervals of 0 mins (fasting levels), 15min, 30min, 45min, 60min, 90min, and 120 min. The blood glucose levels after consumption of the white yam diets/treatments were determined after a minimum of two (2) days allowed between the consumption of the White Yam and Soya beans, and the combination of both. To ensure there was no residual effect of one treatment over the other. The same participants were used for all the test carried out. Glycemic Response curves were plotted from the average blood glucose concentrations of all participants against time. The incremental area under the glucose response curve (IAUC) were calculated using the trapezoid rule as recommended. The area under the fasting baseline was ignored in the calculation. All GIs that were 2 standard deviations above or below the mean GI value for a given test were ignored as an outlier. The glycemic index of the test food samples was calculated as:

$$\mathrm{GI} = \frac{\mathrm{IAUC\ OF\ TEST\ FOOD}}{\mathrm{IAUC\ OF\ REF.\ FOOD}}\ \mathrm{X}\ 100$$

The average of the two measures for each subject was taken as the GI for that White Yam for the subject. The GI for each White Yam diet was finally calculated as the mean of the average of the GIs in ten subjects in the group (Okorie Chinasa (2021)).

CHAPTER THREE

RESULTS AND DISCUSSION

3.0 Tables and Calculations
Table 3.1 Standard Glucose Table

TEST TUBES	1	2	3	4	5	6
GLUCOSE (40Mg/dl)	-	1.0	1.5	2.0	2.5	3.0
DISTILLED WATER	3.0	2.0	1.5	1.0	0.5	-
ANTHRONE REAGENT	5ml	5ml	5ml	5ml	5ml	5ml
AMOUNT	0	40	60	80	100	120
ABSORBANCE @620NM	0.000	0.173	0.482	0.547	0.593	0.768

0.2g of glucose was dissolved in 200ml of distilled water (1% stock solution with the dilution factor 2ml was taken from the glucose stock and made up to 500ml.

0, 1.0, 1.5, 2.0, 2.5, 3.0 was diluted in 3.0, 2.0, 1.5, 1.0, 0.5, 0 in separate test tubes respectively.

Then an equal amount of anthrone reagent was added to each test tube, thereby giving us the resulting absorbance in the above table.

STANDARD GLUCOSE CALIBRATION CURVE

The above graph was derived from the standard glucose table plotting the amount (y-axis) in mg/dl and absorbance (x-axis) in nm.

Table 3.2 Carbohydrate Content Table

SAMPLE I.D	SAMPLE TEST	ANTHRONE REAGENT	ABSORBANCE@620NM
White Yam	**1ml**	**5ml**	**1.576**
White Yam	**1ml**	**5ml**	**1.552**
White Yam	**1ml**	**5ml**	**1.636**
Soya Beans	**1ml**	**5ml**	**1.265**
Soya Beans	**1ml**	**5ml**	**1.087**
Soya Beans	**1ml**	**5ml**	**0.991**

- **For White Yam**

Total Average Absorbance = $\frac{1.576+1.552+1.636}{3} = 1.588$

- **For Soya Beans**

Total Average Absorbance = $\frac{1.576+1.552+1.636}{3} = 1.588$

Table 3.3 Using the Standard Glucose Calibration Curve

- **For White Yam**

READINGS	ABSORBANCE	AVERAGE ABSORBANCE	TOTAL CARB CONTENT
1st Reading	**1.576**		
2nd Reading	**1.552**	**1.588**	**240**
3rd Reading	**1.636**		

Total Average Absorbance = $\frac{1.576+1.552+1.636}{3} = 1.588$

$$\frac{1.588}{3} = 0.529$$

In the Graph

80x3 = 240mg/ml

SINCE FOR 1g of White Yam we have 240mg/ml of carbohydrate (0.24g/ml)

Therefore, 100g of White Yam will give 24g of carbohydrate.

- **For Soya Beans**

READINGS	ABSORBANCE	AVERAGE ABSORBANCE	TOTAL CARB CONTENT
1^{st} Reading	1.265		
2^{nd} Reading	1.087	1.114	174
3^{rd} Reading	0.991		

Total Average Absorbance = $\frac{1.265+1.087+0.991}{3} = 1.114$

$$\frac{1.114}{3} = 0.371$$

$$\frac{1.114}{3} = 0.371$$

In the Graph

58x3 = 174mg/ml

SINCE FOR 1g of Soya Beans we have 174mg/ml of carbohydrate (0.17g/ml)

Therefore, 100g of Soya Beans will give 17g of carbohydrate.

Table 3.4 Variation of White Yam to Soya Beans Calculation Using Fig 2.7.1

WHITE YAM	SOYA BEANS
$\frac{100}{100} X\,5 = 5$	$\frac{0}{100} 0\,X\,5 = 0$
$\frac{80}{100} X\,5 = 4$	$\frac{20}{100} X\,5 = 1$
$\frac{60}{100} X\,5 = 3$	$\frac{40}{100} X\,5 = 2$
$\frac{40}{100} X\,5 = 2$	$\frac{60}{100} X\,5 = 3$
$\frac{20}{100} X\,5 = 1$	$\frac{80}{100} X\,5 = 4$
$\frac{0}{100} X\,5 = 0$	$\frac{100}{100} X\,5 = 5$

From the above table the following value 100g, 80g, 60g, 40g, 20g, and 0g of White Yam was varied with soya Beans of 0g, 20g, 40g, 80g, 100g respectively. Were modified to give table below to meet adequate laboratory and experimental methods. The calculation modifications are input in the table above.

WHITE YAM	SOYA BEANS
5	0
4	1
3	2
2	3
1	4
0	5

From the table above exactly 5g, 4g, 3g, 2g, 1g, 0g of White Yam was varied with 0g, 1g, 2g, 3g, 4g, 5g of Soya Beans.

Table 3.5 Variation of White Yam to Soya Beans Absorbance Table

SAMPLE I.D	SAMPLE TEST	ANTHRONE REAGENT	ABSORBANCE@620NM
Y + S 1: 1	1ml	5ml	1.576
Y + S 1: 2	1ml	5ml	1.552
Y + S 1: 3	1ml	5ml	1.636
Y + S 2: 1	1ml	5ml	0.769
Y + S 2: 2	1ml	5ml	0.584
Y + S 2: 3	1ml	5ml	0.518
Y + S 3: 1	1ml	5ml	1.119
Y + S 3: 2	1ml	5ml	0.825
Y + S 3: 3	1ml	5ml	0.844
Y + S 4: 1	1ml	5ml	0.788
Y + S 4: 2	1ml	5ml	0.715
Y + S 4: 3	1ml	5ml	0.754
Y + S 5: 1	1ml	5ml	0.765
Y + S 5: 2	1ml	5ml	1.174
Y + S 5: 3	1ml	5ml	0.964
Y + S 6: 1	1ml	5ml	1.265
Y + S 6: 2	1ml	5ml	1.087
Y + S 6: 3	1ml	5ml	0.991

From this table exactly three test tube of dilution was prepared from each sample variation from the variation table. And 1ml was removed from each sample. 5ml of anthrone was added to each test tube and the absorbance was read at 620nm.

Table 3.6 Amylopectin Content of White Yam

ABSORBANCE	AVERAGE ABSORBANCE	CARBOHYDRATE CONCENTRATION (AMYLOSE)
0.315		
0.310	0.318	48
0.327		

16 x 3 = 48 (Amylose)

Carb content = Amylopectin + Amylose

Amylopectin = Carb content (174) – Amylose (48) = 126

Amylopectin content of White Yam = 126 in 174 of Total Carb Content.

3.1 Glycemic Index

3.1.1 Characteristics of test subjects

The test subjects were of average age, weight, and height of 22±2 years, 62.21±6.13kg and 1.64±0.08m, respectively, the mean fasting blood sugar (FBS) OF 4.45±0.43 mmol/L.

Table 3.1.2 Glycemic Response Table for White for White Yam

TIME	AVG BLOOD GLUCOSE LEVEL OF 6 PARTICIPANTS
Normal Blood Glucose	81mg/ml
15mins after food intake	108mg/ml
30 mins after food intake	142mg/ml
45 mins after food intake	150mg/ml
60 mins after food intake	145mg/ml
90 mins after food intake	120mg/ml

Table 3.1.3 Glycemic Response Curve for White Yam

The above curve and table, was derived by first measuring the blood glucose level of the fasting blood sugar, which was an average of 81mg/dl and afterwards the blood glucose level of the six participants were measured at 15, 30, 45, 60, and 90 mins after ingesting the food. To give 108, 142, 150, 145, and 120mg/dl respectively.

Table 3.1.4 Glycemic Response Table and Curve for Soya Beans

TIME	AVG BLOOD GLUCOSE LEVEL OF 6 PARTICIPANTS
Normal Blood Glucose	83mg/ml
15mins after food intake	91mg/ml
30 mins after food intake	105mg/ml
45 mins after food intake	115mg/ml
60 mins after food intake	100mg/ml
90 mins after food intake	111mg/ml

Glycemic Response Curve for Soya Beans

The above curve and table, was derived by first measuring the blood glucose level of the fasting blood sugar, which was an average of 83mg/dl and afterwards the blood glucose level of the six participants were measured at 15, 30, 45, 60 and 90 mins after ingesting the food to give 91, 105, 115, 100, and 111mg/dl respectively.

3.5.7 Glycemic Response Curve of Soya Beans Against White Yam

WHITE YAM	SOYA BEANS	IAUC		G.I
80g	20g	YAM	SOYA BEANS	35.1
		210	585	
60g	40g	200	590	33.9
40g	60g	190	600	31.7
20g	80g	170	620	27.4

Table 3.1.6 Improve Glycemic Response of White Yam by Integrating with Soya Beans

FOOD ITEM	IAUC	GI REF TO GLUCOSE	INTEGRATION OF WHITE YAM WITH SOYA BEANS
WHITE YAM	210	44.26	
			35
SOYA BEANS	585	14	

From the above table the IAUC and Glycemic Index of White Yam with Soya Beans was determined by the following calculations.

So, therefore the IAUC of Soya Beans is = 120 + 210 + 150 + (-225) + 330 = 585mg/dl

Therefore, the glycemic index of White Yam (TEST FOOD) upon incorporation with Soya Beans (REFERENCE FOOD).

The Glycemic Index of White Yam

$$\mathbf{GI} = \frac{\textbf{IAUC OF WHITE YAM}}{\textbf{IAUC OF SOYA BEANS}}\ \mathbf{X\ 100}$$

$$\frac{\mathbf{210}}{\mathbf{585}}\ \mathbf{X\ 100 = 35.1}$$

Therefore, The Glycemic Index of White Yam is lower/improved from 51 to 35.1 upon variation with soya beans.

CONCLUSION

After conducting an investigation into the glycemic index of white yam when paired with varying amounts of soya beans, it is evident that the combination influences the overall glycemic response. The findings suggest that incorporating soya beans with white yam may potentially reduce or modulate its glycemic index, impacting blood sugar levels differently. Further research could delve deeper into the specific mechanisms behind this interaction and its implications for dietary recommendations and managing blood sugar levels effectively.

APPENDIX

To calculate the dilution factor in carbohydrate content

STD Glucose (40mg/dl): Required Concentration

1%= 0.1g/10ml ≡ 1g/100ml

Original Concentration = 10,000mg/ml

Required Volume = 50ml

Dilution Factor = $\frac{Required\ concentration \times Required\ volume}{Original\ Concentration}$

$$\mathrm{D.F} = \frac{40 \text{ x } 50}{10{,}000} = 0.2\mathrm{g}$$

For White Yam,

0 to 15 mins

Blood sugar increased by

108 – 81 = 27mg/ml

Time Interval 15 – 0 min = 15 mins

Area of Interval = 27 x 15 = 465mg.min/dl

15 to 30 mins

Blood sugar increased by

142 – 108 = 34mg/ml

Time Interval 30 – 15 min = 15 mins

Area of Interval = 34 x 15 = 510mg.min/dl

30 to 45 mins

Blood sugar increased by

150 – 142 = 2mg/ml

Time Interval 45– 30 min = 15 mins

Area of Interval = 8 x 15 = 120mg.min/dl

45 to 60 mins

Blood sugar increased by

145 – 150 = 15mg/ml

Time Interval 60 – 45 min = 15 mins

Area of Interval = -5 x 15 = -75mg.min/dl

60 to 90 mins

Blood sugar increased by

120 – 145 = -25mg/ml

Time Interval 90 – 60 min = 30 mins

Area of Interval = -25 x 15 = -750mg.min/dl

For Soya Beans,

0 to 15 mins

Blood sugar increased by

91 – 83 = 8mg/ml

Time Interval 15 – 0 min = 15 mins

Area of Interval = 8 x 15 = 120mg.min/dl

15 to 30 mins

Blood sugar increased by

105 – 91 = 14mg/ml

Time Interval 30 – 15 min = 15 mins

Area of Interval = 15 x 14 = 210mg.min/dl

30 to 45 mins

Blood sugar increased by

105 – 115 = -15mg/ml

Time Interval 45– 30 min = 15 mins

Area of Interval = 10 x 15 = 150mg.min/dl

45 to 60 mins

Blood sugar increased by

105 – 115 = -15mg/ml

Time Interval 60 – 45 min = 15 mins

Area of Interval = -15 x 15 = -225mg.min/dl

60 to 90 mins

Blood sugar increased by

111 – 100 = 11mg/ml

Time Interval 90 – 60 min = 30 mins

Area of Interval = 11 x 30 = 330mg.min/dl

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