MODULATORY ROLE OF CABBAGE (*Brassica olaeracae*) SUPPLEMENT ON BLOOD GLUCOSE AND SOME PHYSIOLOGICAL PARAMETERS IN ALLOXAN – INDUCED DIABETIC WISTAR RATS

BY

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A THESIS SUBMITTED TO THE DEPARTMENT OF HUMAN PHYSIOLOGY FACULTY OF MEDICINE, AHMADU BELLO UNIVERSITY ZARIA IN PARTIAL FULFILLMENT OF THE AWARD OF MASTER OF SCIENCE DEGREE IN HUMAN PHYSIOLOGY

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DECLARATION

I, Muhammad Abdulmalik declare that the work in this thesis entitled: Modulatory Role of Cabbage (*Brassica olaeracae*) Supplement on Blood Glucose and Some Physiological Parameters in Alloxan Induced Diabetic Wistar Rats was carried out by me in the Department of Human Physiology, Faculty of Medicine, Ahmadu Bello University, Zaria, under the supervision of Dr. Aliyu Mohammed and Dr. Yusuf Tanko. The information derived from the literature has been duly acknowledged in the text and a list of references provided. No part of this thesis was previously presented for another degree or diploma at this or any other institution.

_____ _____

Muhammad Abdulmalik Date

CERTIFCATION

This thesis titled "**Modulatory Role of Cabbage (*Brassica olaeracae*) Supplement On Blood Glucose and Some Physiological Profiles in Alloxan Induced Diabetic Wistar Rat"** by Muhammad Abdulmalik, meets the regulation governing the award of Master of Science in Human Physiology of Ahmadu Bello University, Zaria and is approved for its contribution to knowledge and literal presentation.

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DEDICATION

This work is dedicated to Almighty Allah, for his guidance and mercy throughout the course of the programme and to my beloved parents and family.

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ABSTRACT

Diabetes mellitus is a global health problem affecting with more people in developing than developed countries. Insulin and oral hypoglycemic drugs have remained the corner stone for the management of diabetes mellitus. Unfortunately, apart from having a number of side effects, none of the oral synthetic hypoglycemic agents has been successful in maintaining euglycaemia. The use of medicinal plants for the treatment of diabetes mellitus has gained recognition and recommendation by the World Health Organization especially in developing countries where access to the conventional treatment is expensive and not readily accessible. Various plants and plant extracts have been found to play an important role in the treatment of diabetes and these plants were believed to have hypoglycemic properties. Cabbage is one of such medicinal plants, whose therapeutic application has a folkloric background. The plant enjoys widespread reputation as a remedy for various ailments. Most of the research work done on cabbage has been on extract. Hence, a scientific verification of its use as a supplement in food would be important in establishing a pharmacological basis for some of the claimed ethnomedicinal uses of the plant. This scientific verification forms the basis of the present investigation using animal models. The aim of the study is to determine the modulatory role of cabbage supplement on blood glucose levels and some physiological parameters in alloxan induced diabetic Wistar rats. While the objectives were to determine the effects of cabbage on serum glucose levels, to determine the effects of cabbage on lipid profile and also to determine the effects of cabbage on serum liver enzymes activities on alloxan induced diabetic Wistar rats respectively. The study was designed to investigate the effect of cabbage supplement on blood glucose, lipid profile and serum liver enzymes on alloxan induced diabetic Wistar rats. A total of twenty five Wistar rats of both sexes weighing 100 – 150 g were used. They were randomly allocated into five groups of five rats (n = 5 rats/group). Group one were diabetic rats given distilled water and served as the negative

control. Group two were diabetic rats that received 5 mg/kg b/w of glibenclamide orally and served as positive control. While, groups three, four and five were diabetic rats that received 10, 25 and 50% cabbage supplement, respectively. All groups were treated for thirty days. Blood glucose and some physiological parameters including lipid profile, aspartate aminotransferase (AST), alanine aminotransferase (ALT), and alkaline phosphatase (ALP), were measured in all rats. Blood glucose level was significantly ($p< 0.05$) reduced in treated diabetic rats in comparison to the diabetic control rats. In addition, serum total cholesterol, triglyceride, and low-density lipoprotein (LDL) were significantly decreased ($p<0.05$) while high-density lipoprotein (HDL) was increased ($p<0.05$) in treated than in diabetic control groups. Furthermore, feed supplementation with cabbage in caused a significant increase ($p<0.05$) in serum levels of AST and ALT but decrease ($p<0.05$) in ALP enzymes. The results of this study suggest that cabbage when given as a supplement has hypoglycaemic and antilipidaemic properties and thus its beneficial effect in the management of diabetes mellitus may be considered.

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LIST OF ABBREVATION

ALP	Alkaline transferase
ALT	Alanine aminotransferase
ANG II	Angiotensin II
ANOVA	Analysis of variance
ApoB	Apolipoprotein B
AST	Aspartate aminotransferase
ATP	Adenosine triphosphate
CETP	Cholesterol ester transport protein
cGMP	Cyclic guanosine monophosphate
CRP	C-reactive protein
CVD	Cardiovascular disease
DM	Diabetes Mellitus
DSPN	Distal symmetrical polyneuropathy
ED	Erectile dysfunction
FFA	Free fatty acids
GDM	Gestational diabetes mellitus
GPX	Glutathione peroxidise
HDL	High density lipoprotein cholesterol
HLA	Human leukocyte antigen
IL-6	Interleukin-6
LDL	Low density lipoprotein cholesterol
MetS	Metabolic syndrome
MODY	Maturity onset of diabetes of the young
NDM	Neonatal diabetes mellitus

NO	Nitric oxide
PAI-1	Plasminogen activator inhibitor-1
PNDM	Permanent neonatal diabetes mellitus
SACS	S-allyl cystein sulfoxide
SNS	Sympathetic nervous system
SOD	Superoxide dismutase
SUR	Sulfonylurea receptors
T1DM	Type 1 diabetes mellitus
TC	Total cholesterol
TGF-β	Transforming growth factor-β
TGs	Triglycerides
TNDM	Transient neonatal diabetes mellitus
TNFα	Tumour necrosis factor alpha
TZDs	Thiazolidinediones
VEGF	Vascular endothelial growth factor
VLDL	Very low density lipoprotein
WHO	World Health Organization

CHAPTER ONE

1.0 INTRODUCTION

Diabetes is a disease which affects the metabolism of carbohydrates, proteins, and fat due to absolute or relative deficiency of insulin secretion with or without varying degree of insulin resistance (Asadujjaman *et al*., 2011). The number of individuals with diabetes has been increasing due to population growth, aging, urbanization, and increasing prevalence of obesity and physical inactivity (Sarah *et al*., 2004). The World Health Organization (WHO) estimated the diabetic population to increase up to 300 million or more by the year 2025 (Patel *et al.*, 2012). The most important distinctive feature of diabetes is an elevated blood glucose concentration, but this abnormality is just one of a number of biochemical and physiological changes that occur (Olaitan, 2012). Hypercholesterolemia and hyper triglyceridemia are common complications of diabetes mellitus (Akhtar *et al*., 2007). The treatment of diabetes mainly involves the use of hypoglycaemic drugs in addition to insulin but the unwanted side effects of these drugs prompted a demand for new compounds for the treatment of diabetes (Asadujjaman *et al*., 2011). The drive for change from orthodox to herbal medicines is to an extent due to the adverse reactions, undesirable side effects of synthetic drugs, the cost of buying modern antidiabetic drugs, which is beyond the reach of the lower class citizens and the belief that natural products are safer to the biological systems (Mohammed *et al*., 2007). It has now become necessary to search for new compounds in order to overcome these problems, and several traditional medicines are now used to manage diabetes mellitus in different societies all over the continents (Raju *et al*., 2011).

Cabbage (*Brassica oleracea*) is locally called *Kabeji* in Hausa language and *Akojopo* or *Jaleji* in Yoruba language. It is an important vegetable crop of the *Brassicaceae* family consumed all

over the world. It is popular probably due to its low price and availability at local markets, richness in phytochemicals such as polyphenolics, glucosinolates, carotenoids, and vitamin C. It has demonstrated antioxidant, anticancer and potential anti-obesity properties (Dunja *et al.*, 2013). It consists of a wide range of important vegetable and fodder crops which are excellent sources of fibers that help prevents constipation, reduces the risk of colorectal cancer and helps to reduce blood sugar and blood cholesterol levels, thereby reducing the risk of heart disease and diabetes (Puddephat *et al,*1996; Enas and Atif 2010). Ethanolic extract of cabbage, has demonstrated significant hepatoprotective activity which justifies its use as a hepato protective agent as a result of the presence of biologically active phytoconstituents (Subramanian, 2011).

1.1 Statement of Research Problem

Diabetes mellitus is a global health problem which affects an estimated population of 135 million in 1995 and the number being expected to rise to about 300 million in the year 2025 with more people in developing than developed countries (Baltazar *et al.*, 2004). Insulin and oral hypoglycemic drugs such as sulfonylureas and biguanides have remained the corner stone for the management of diabetes mellitus. Unfortunately, apart from having a number of side effects, none of the oral synthetic hypoglycemic agents has been successful in maintaining euglycaemia. The use of medicinal plants for the treatment of diabetes mellitus has gained recognition and recommendation by the World Health Organization (WHO), especially in developing countries where access to the conventional treatment are expensive and not readily accessible (Akhtar, 2007).

1.2 Justification

More than four hundred different plant and plant extracts have been found to play an important role in the treatment of diabetes and most of these plants were believed to have hypoglycaemic properties (Bailey and Caroline, 1989). Cabbage is one of such medicinal plants, whose therapeutic application has a folkloric background. The plant enjoys widespread reputation as a remedy for peptic ulcer disease, hypocholeterolaemic, anti-cancer, antimicrobial and anti-inflammatory properties (Sathya, 2012). Most of the research work done on cabbage has been on extract. Hence a scientific verification of its use as a supplement in food would be important in establishing a pharmacological basis for some of the claimed ethnomedicinal uses of the plant. This scientific verification forms the basis of the present investigation using animal models.

1.3 Research Hypothesis

1. Cabbage supplement have no effect on blood glucose level and some physiological profiles on alloxan induced diabetic Wistar rats.

1.4. General Aim

To determine the modulatory role of cabbage supplement on blood glucose lipid profile and liver enzymes activities on alloxan induced diabetic Wistar rats.

1.4.1 Objectives of the Study

1. Determine the effects of cabbage supplement on serum glucose levels on alloxan induced diabetic Wistar rats.
2. Determine the effects cabbage supplement on lipid profile namely; total cholesterol, high density lipoprotein, low density lipoprotein and total triglycerides on alloxan induced diabetic Wistar rats.
3. To determine the effects of cabbage supplement on serum liver enzymes activities: alanine aminotransferase (ALT), aspartate aminotransferse (AST) and alkaline aminotransferase (ALP) on alloxan induced diabetic Wistar rats.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 INTRODUCTION

Diabetes Mellitus (DM) is a global health care menace that may reach pandemic levels by 2030 (Abougalambou *et al.*, 2010). About 80% of the total adult diabetics are in developing countries and the greatest concern is the growing incidence of Type 2 Diabetes at a younger age including some obese children even before puberty affecting the productive years of their lives (Tabish, 2007). Considerable evidence has seen diabetes changing into an epidemic in many developing countries with an estimated prevalence of diabetes of 1% in rural areas of Africa and prevalence in Nigeria ranging from 0.65% in rural Mangu in the North to 11% in urban Lagos in the Southern part of the country (Chinenye and Young 2011; Mutlu, *et al.*, 2014). Diabetes mellitus is a disease that is waging war against the wellbeing of humans and may probably be due to drastic lifestyle changes accompanying urbanization and westernization in developing countries (Harande, 2011). Individuals with diabetes are more likely to be hospitalized with cardiovascular disease, end stage renal disease and most frequently, non traumatic lower limb amputation compared to the general population (Cheng, 2013).

2.2 Diabetes Mellitus

Diabetes mellitus is a chronic and complex metabolic disease that has deleterious effects on multiple organ system (Bhuva and Addepalli, 2011). It is an endocrine-metabolic disorder characterized by chronic hyperglycaemia secondary to relative or absolute lack of insulin giving rise to the risk of microvascular and macrovascular damage, with associated reduced life expectancy and diminished quality of life (Chinenye and Young, 2011). Several disease

mechanisms are involved in the development of diabetes mellitus ranging from autoimmune destruction of the β-cells of the pancreas to abnormalities that result in resistance to insulin action. However of recent, strong evidence has shown that both type-1 diabetes and type-2 diabetes are now caused by the formation of tiny toxic clumps of the hormone amylin, produced by the beta cells of the pancreas which in turn destroys the pancreatic beta cells leading to the development of diabetes **(** Zhang *et al.,* 2014).

2.3 Types of Diabetes Mellitus

2.3.1 Type 1 diabetes mellitus

Type 1 diabetes mellitus (T1DM) is present in about 5–10% of people having diabetes with a strong genetic linkage inherited mainly through the HLA complex (Daneman, 2006). An autoimmune disease mediated by T lymphocytes which cause a progressive destruction of the pancreatic beta cells (Dip and Gomez, 2009). A large number people with T1DM are youths, amounting to ≥85% of all diabetes cases in youth < 20 years of age and individuals diagnosed with T1DM when they are adults have been referred to as having latent autoimmune diabetes of adults (Maahs *et al.,* 2011). Those with T1DM have increased risk of other autoimmune disorders like autoimmune thyroid and celiac diseases while complications like nephropathy may result into hypercalciuria which can alter vitamin D metabolism leading to vitamin D deficiency and consequently osteoporosis (Dhaon and Shah, 2014). The genetic basis of T1DM is not yet fully understood. However, genetic determinants such as alleles of the major histocompatibility locus (HLA) at the HLA-DRB1 and DQB1 loci and it has been discovered that HLA-B*39 locus account for some 40–50% of the familial clustering of T1DM (Forbes and Cooper, 2013).

2.3.2 Type 2 diabetes mellitus

Type 2 Diabetes Mellitus is a chronic metabolic disease caused by impaired glucose tolerance due to insulin resistance and consequential islet β-cell exhaustion, resulting to insulin deficiency (Badawi *et al.,* 2010). It is the most common type of diabetes affecting about 85-90% of all people with older adults usually affected. Younger populations are also now increasingly being diagnosed with Type 2 diabetes, having about 45% of new cases of Type 2 diabetes mellitus in the paediatric population **(**Jordan and Jordan, 2012). Genetic and environmental factors play a role in the aetiology of Type 2 diabetes and the risk is greatly increased with change in lifestyle factors such as high blood pressure, obesity, lack of exercise and poor diet (Hu *et al.,*2001). It is still not established whether genetic factors or aging can explain the rapid increase in the prevalence of Type 2 Diabetes (Forbes and Cooper, 2013).

2.3.3 Maturity onset of diabetes of the young (MODY)

Maturity onset of diabetes of the young (MODY) is a form of diabetes that is caused by mutations in a number of different genes. It has now been discovered that there are at least six forms of MODY (MODY 1- 6), each with mutation in a different gene that is directly involved with beta cell function (Winter, 2003).Those who carry MODY2 mutations have a very mild form of the disease, while those who carry MODY1 and MODY3 variants have a much more severe expression that is associated with long-term complications (Pearson *et al.,* 2004). MODY, is inherited as an autosomal dominant trait resulting from mutations in glucokinase gene on chromosome 7p.This condition is diagnosed as hyperglycemia before the age of twenty-five years and treatable for over five years without insulin in cases where islet cell antibodies are negative and HLA-DR3 and DR4 are heterozygous. MODY is more common in blacks and

Indians seen in more than 10% of diabetics but rare in Caucasians having a prevalence of less than 1% (Ozougwu *et al.,* 2013).

2.3.4 Gestational diabetes

Gestational diabetes mellitus (GDM) is defined by glucose intolerance diagnosed first at any time during pregnancy.This condition is found to be associated with various prenatal and maternal complications ranging from neonatal hypoglycemia, macrosomia, jaundice, hypocalcaemia and polycythemia with an increased frequency of maternal hypertensive disorders leading to an increased rate of caesarean delivery (Linsay, 2009).

2.3.5 Neonatal diabetes

Neonatal diabetes mellitus (NDM) is defined as hyperglycaemia requiring insulin treatment occurring in the first 6 months of life (Kataria, 2014). Neonatal diabetes mellitus is considered a rare disease affecting one in 300,000 to 400,000 newborns (Polak and Cavé , 2007 ; Grulich-Henn *et al.,* 2010). It is classified as transient NDM (TNDM) and permanent NDM (PNDM) with TNDM, accounting for 50% to 60% of all NDM cases, requires initial insulin treatment but appears to resolve spontaneously by a median of 12 weeks of age, only to relapse years later (Kataria, 2014). Permanent NDM is less common and is also characterized by early hyperglycaemia. However, unlike TNDM, PNDM has no period of remission and must be treated lifelong. Unlike autoimmune diabetes, which is extremely rare before 6 months of age, NDM is a monogenic form of diabetes, with insulinopenia resulting from abnormal pancreatic islet development, decreased β -cell mass, or -cell β dysfunction (Aguilar-Bryan and Bryan, 2008).

2.3.6 Pathogenesis of type 2 diabetes mellitus

Under normal physiological conditions, plasma glucose concentrations are maintained within a narrow range, despite wide fluctuations in supply and demand, through a tightly regulated and dynamic interaction between tissue sensitivity to insulin (especially in the liver) and insulin secretion (DeFronzo, 1988). In type 2 diabetes these mechanisms break down, with the consequence that the two main pathophysiological defects in type 2 diabetes are impaired insulin secretion through a dysfunction of the pancreatic β-cell, and impaired insulin action through insulin resistance. Insulin resistance is now regarded as being synonymous with a reduced rate of whole-body insulin-mediated glucose disposal in insulin-sensitive tissues (DeFronzo and Ferrannini, 1991; Moller and Flier, 1991). This definition is too narrow, and insulin resistance may be better defined as existing when normal insulin concentrations fail to produce a normal biological response (Kahn, 1978).

The main advantage of the latter definition is that it does not restrict consideration of insulin action to a solitary aspect of intermediary metabolism. The importance of insulin actions on other aspects of intermediary metabolism, including lipid and protein metabolism, has now been appreciated. In 1988, Reaven described syndrome X (now commonly known as the metabolic syndrome or insulin resistance syndrome) as the association between several cardiovascular risk factors, including hypertension, dyslipidaemia and glucose intolerance, and he proposed that insulin resistance was the underlying cause (Reaven, 1988). Since his original description, it has been recognised that a significant proportion of patients with the metabolic syndrome do not have insulin resistance (Ford *et al*, 2002). The definition of the metabolic syndrome has therefore

been modified: most notably, central obesity has been added as a core feature. This is important, as it is increasingly recognised that central obesity is fundamental to the origin of this disorder (Maison *et al,* 2001). Abnormalities in β-cell function are found early in the natural history of type 2 diabetes and in first-degree relatives of people with type 2 diabetes (Pratley and Weyer, 2001), suggesting that they are an integral component of the pathogenesis of type 2 diabetes.

2.4 Metabolic Syndrome

Metabolic syndrome (MetS) is defined by a group of interconnected physiological, biochemical, clinical, and metabolic factors that directly increases the risk of atherosclerotic cardiovascular disease (Grundy *et al.*, 2005; Kaur, 2014). Worldwide prevalence of MetS ranges from <10% to as much as 84%, depending on the region, urban or rural environment, composition of the population studied, and the definition of the syndrome used (Kaur, 2014). MetS is a state of chronic low grade inflammation as a consequence of complex interplay between genetic and environmental factors. Insulin resistance, visceral adiposity, atherogenic dyslipidemia, endothelial dysfunction, genetic susceptibility, elevated blood pressure, hypercoagulable state, and chronic stress are the several factors which constitute the syndrome.

2.5 Abdominal Obesity

Obesity is principally driven by an increased consumption calorie-dense food and reduced physical activity. Adipose tissue is a heterogeneous mix of adipocytes, stromal preadipocytes, immune cells, and endothelium, and it can respond rapidly and dynamically to alterations in nutrient excess through adipocytes hypertrophy and hyperplasia (Halberg *et al.*, 2008). Progressive adipocytes enlargement with obesity may lead to depletion of blood supply to the

adipocytes which may consequently lead to hypoxia (Cinti *et al.,* 2005). Hypoxia has been proposed to be an inciting etiology of necrosis and macrophage infiltration into adipose tissue that leads to an overproduction of biologically active metabolites known as adipocytokines which includes glycerol, free fatty acids (FFA), proinflammatory mediators (tumour necrosis factor alpha (TNFα) and interleukin-6 (IL-6)), plasminogen activator inhibitor-1 (PAI-1), and C-reactive protein (CRP) (Lau *et al.,* 2005). This result in a localized inflammation in adipose tissue that propagates an overall systemic inflammation associated with the development of obesity related comorbidities (Trayhurn and Wood, 2004). Adipocytokines integrate the endocrine, autocrine, and paracrine signals to mediate the multiple processes including insulin sensitivity, oxidant stress, energy metabolism, blood coagulation, and inflammatory responses which are thought to accelerate atherosclerosis, plaque rupture, and atherothrombosis (Jacobs *et al.,* 2009; Saleem *et al.,* 2009; Tsimikas *et al.,* 2009). This shows that the adipose tissue is not only specialized in the storage and mobilization of lipids but it is also a remarkable endocrine organ releasing the numerous cytokines.

2.6 Free Fatty Acid

The upper trunk subcutaneous adipocytes generate a majority of circulating Free Fatty Acid (FFA). In addition, an intra-abdominal fat content has been positively correlated with the splanchnic FFA levels which may a contributory factor to the liver fat accumulation commonly found in abdominal obesity (Miles and Jensen, 2005). An acute exposure of skeletal muscle to the elevated levels of FFA induces insulin resistance by inhibiting the insulin-mediated glucose uptake, while, a chronic exposure of the pancreas to the elevated FFA impairs a pancreatic β-cell function (Boden *at al.,* 2001).

2.7 C - Reactive Protein

Elevated level of C- reactive protein (CRP) is likely to be seen in obese insulin-resistant, but, not in obese insulin-sensitive subjects (McLaughlin *et al.,* 2002). In addition, it has been demonstrated that regardless of the presence or degree of the MetS in an individual, CRP levels independently predicts the occurrence of future cardiovascular disease (CVD) events (Ridker *et al.,* 2003). Because the MetS has been linked with a greater chance of future CVD events, CRP levels may be an important independent predictor of unfavourable outcomes in the MetS (Clearfield, 2005).

2.8 Leptin

It is an adipokine involved in the regulation of satiety and food intake (Lau *et al.,* 2005). Levels of leptin in the plasma increase during the development of obesity and decline during the weight loss. Leptin receptors are located mostly in the hypothalamus and the brainstem and signals through these receptors controls satiety, energy expenditure, and neuroendocrine function. Most overweight and obese individuals have an elevated level of leptin that do not suppress appetite (leptin resistance). Leptin resistance is thought to be a fundamental pathology in obesity (Hutley and Prins, 2005). Besides its effect on appetite and metabolism, leptin acts in the hypothalamus to increase the blood pressure through activation of the sympathetic nervous system (SNS) (Carlyle *et al.,* 2003). High circulating levels of leptin are reported to explain much of the increase in the renal sympathetic tone observed in obese human subjects (Eikelis *et al.,* 2003). Leptin is a nitric oxide (NO) dependent vasodilator but also increases the peripheral vascular resistance and the sympathetic nerve activity (Shirasaka, 2003). The concentration of plasma

leptin is correlated with adiposity, and hyperleptinemia is indeed considered an independent cardiovascular disease risk factor (Kaur, 2014).

2.9 Tumour Necrosis Factor α

It is a paracrine mediator in adipocytes and appears to act locally to reduce the insulin sensitivity of adipocytes (Lau *et al.,* 2005). Evidence suggests that Tumour Necrosis Factor α (TNF-α) induces adipocytes apoptosis and promotes insulin resistance by the inhibition of the insulin receptor substrate 1 signalling pathway (Hotamisligil *et al.,* 1996 ; Xydakis *et al.,* 2004). The paracrine action would further tend to increase the FFA release, inducing an atherogenic dyslipidaemia. Plasma TNFα is positively associated with the body weight, and triglycerides (TGs), while, a negative association exists between the plasma TNFα and High density lipoprotein (HDL) (Krauss, 2004).

2.10 Adiponectin

It regulates the lipid and glucose metabolism, increases insulin sensitivity, regulates food intake and body weight, and protects against a chronic inflammation. It inhibits hepatic gluconeogenic enzymes and the rate of an endogenous glucose production in the liver. It increases glucose transport in muscles and enhances fatty acid oxidation (Eckel *et al.,* 2005). It has a multifactorial antiatherogenic action which includes an inhibition of endothelial activation, a reduced conversion of macrophages to foam cells, and inhibition of the smooth muscle proliferation and arterial remodelling that characterizes the development of the mature atherosclerotic plaque (Eckel *et al.,* 2005). Adiponectin is inversely associated with CVD risk factors such as blood pressure, low density lipoprotein cholesterol (LDL), and Triglycerides (Krauss, 2004). It has

been reported that hypoadiponectinemia is associated with insulin resistance, hyperinsulinemia, and the possibility of developing T2DM, independent of fat mass (Fumeron *et al.,* 2004). Adiponectin is seen to be "protective," not only in its inverse relationship with the features of MetS but also through its antagonism of TNFα action.

2.11 Dyslipidemia

This dyslipidemia is characterised by a spectrum of qualitative lipid abnormalities reflecting perturbations in the structure, metabolism, and biological activities of both atherogenic lipoproteins and antiatherogenic high density lipoprotein (HDL) which includes an elevation of lipoproteins containing apolipoprotein B (apoB), elevated TGs, increased levels of small particles of LDL, and low levels of HDL. Insulin resistance leads to an atherogenic dyslipidemia in several ways. First, insulin normally suppresses lipolysis in adipocytes, so an impaired insulin signalling increases lipolysis, resulting in increased FFA levels. In the liver, FFAs serve as a substrate for the synthesis of TGs. FFAs also stabilize the production of apoB, the major lipoprotein of very low density lipoprotein (VLDL) particles, resulting in a more VLDL production. Second, insulin normally degrades apoB through PI3K-dependent pathways, so an insulin resistance directly increases VLDL production. Third, insulin regulates the activity of lipoprotein lipase, the rate-limiting and major mediator of VLDL clearance. Thus, hypertriglyceridemia in insulin resistance is the result of both an increase in VLDL production and a decrease in VLDL clearance. VLDL is metabolized to remnant lipoproteins and small dense LDL, both of which can promote an atheroma formation. The TGs in VLDL are transferred to HDL by the cholesterol ester transport protein (CETP) in exchange for cholesteryl esters, resulting in the TG-enriched HDL and cholesteryl ester enriched VLDL particles. Further,

the TG-enriched HDL is a better substrate for hepatic lipase, so it is cleared rapidly from the circulation, leaving a fewer HDL particles to participate in a reverse cholesterol transport from the vasculature. Thus, in the liver of insulin-resistant patients, FFA flux is high, TGs synthesis and storage are increased, and excess TG is secreted as VLDL (Hu *et al.,* 2001). For the most part, it is believed that the dyslipidaemia associated with insulin resistance is a direct consequence of increased VLDL secretion by the liver .These anomalies are closely associated with an increased oxidative stress and an endothelial dysfunction, thereby reinforcing the proinflammatory nature of macrovascular atherosclerotic disease (Bastaki, *et al.,* 2005).

2.12 Hypertension

Hypertension is frequently common among patients with diabetes (Matheus *et al.,* 2013).The coexistence of these two conditions increase the risk of developing macrovascular complications and also microvascular complications and close monitoring and treatment of hypertension may reduce the progression of these complications(Matheus *et al.,* 2013). Hypertension affects majority of individuals with diabetes mellitus especially those with type 2 diabetes mellitus and its pathogenesis is complex, involving interactions between genetic predisposition and a range of environmental factors (Gilbert *et al.,* 2013). Hypertension commonly occurs without abnormal renal function and insulin resistance precipitate hypertension by stimulating the sympathetic nervous system and the rennin – angiotensin system, promoting sodium retention (Lago *et al.,* 2007).

2.12 Complications of Diabetes Mellitus

Diabetes is associated with a number of complications. These complications are wide ranging and are due at least in part to chronic elevation of blood glucose levels, which leads to damage of blood vessels (angiopathy). In diabetes, the resulting complications are grouped under microvascular disease and macrovascular disease. Microvascular complications include retinopathy, nephropathy, and neural damage or neuropathy. The major macrovascular complications include accelerated cardiovascular disease resulting in myocardial infarction and cerebrovascular disease manifesting as strokes. Other chronic complications of diabetes include depression, (Nouwen *et al.,* 2011), dementia (Cukierman *et al.,* 2005), and sexual dysfunction (Adeniyi *et al.,* 2011; Thorve *et al.,* 2011).

2.12.1 Diabetic Nephropathy

Diabetes is one of the leading causes of end-stage renal failure. The pathophysiologic event that takes place in diabetic nephropathy is damage to the basement membrane with associated renal damage. There is a progressive thickening of the basement membrane and damage to mesangial and vascular cells leading to the passage of macromolecules which may activate inflammatory pathways that contribute to secondary damage (Evans and Capell, 2000). Activation of the transforming growth factor-β (TGF-β) system is also said to be linked in the pathogenesis of diabetic nephropathy, based on the properties of TGF-β, as well as the observation that levels of TGF-β mRNA and protein are significantly increased in the glomeruli and tubulointerstitium in human diabetes and in animal models (Wendt *et al.,* 2013). Some of the risk factors for the development of diabetic nephropathy include hypertension, hyperglycaemia smoking and high protein diet. Treatment intervention may include screening for the earliest stages of renal damage and aggressively controlling blood glucose and blood pressure which can help prevent further renal damage (Evans and Capell, 2000; Ayodele *et al.,* 2004).

2.12.2 Diabetic Neuropathy

The most common complication forms of diabetic neuropathy are autonomic neuropathy and distal symmetrical polyneuropathy (DSPN) with DSPN being the most common manifestation, but many patterns of nerve injury can also occur (Callaghan *et al.,* 2012; Jimenez-Cohl *et al.,* 2012). The risk factor is hyperglycaemia. Sensorimotor neuropathy is associated with pain, sensory loss and paresthesia. Gastroparesis and Genitourinary dysfunctions are the main complications of diabetic neuropathy. The pathology involves oxidative stress, advanced glycation end products, polyol pathway flux and protein kinase C activation which contribute to nerve dysfunction and microvascular disease (Duby *et al.,* 2004). In addition to motor neuron dysfunction, the autonomic nervous system is also influenced by diabetes. One common abnormality in autonomic function seen in individuals with diabetes is orthostatic hypotension, due to an inability to adjust heart rate and vascular tone to maintain blood flow to the brain. The autonomic nerves innervating the gastrointestinal tract are also affected leading to gastroparesis, nausea, bloating, and diarrhoea, which can also alter the efficacy of oral medications. In particular, delayed gastric emptying can dramatically affect glycaemia control by delaying the absorption of key nutrients, as well as antidiabetic agents leading to imbalances in glucose homeostasis. The wide variety of clinical manifestations seen with neuropathy, in addition to impaired wound healing, erectile dysfunction, and cardiovascular disease, can severely impede quality of life. Indeed, autonomic markers can predict which diabetic individuals have the poorest prognosis following myocardial infarction (Barthel *et al.,* 2011). Consistent with other complications, the duration of diabetes and lack of glycaemia control are the major risk factors for neuropathy in both major forms of diabetes (Forbes and Cooper, 2013). Other than

optimization of glycaemia control and management of neuropathic pain, there are no major therapies approved in either Europe or the United States for the treatment of diabetic neuropathy. In addition, as is seen with other complications, the mechanisms leading to diabetic neuropathy are poorly understood. At present, treatment generally focuses on alleviation of pain, but the process is generally progressive.

2.12.3 Diabetic Retinopathy

Diabetic retinopathy develops over many years, and almost all patients with type 1 diabetes (Roy *et al.,* 2004; Hirai *et al.,* 2011) and most having type 2 diabetes (Kempen *et al.,* 2004), exhibit some retinal lesions after 20 years of disease. Furthermore, whereas in type 1 diabetes the major vision threatening retinal disorder appears to be proliferative retinopathy (Klein *et al.,* 2008), in type 2 diabetes there is a higher incidence of macula oedema. Nevertheless, only a minority of such patients will have progression resulting in impaired vision. In addition to maintenance of blood pressure and glycaemia control, there are a number of treatments for diabetic retinopathy that have efficacy in reducing vision loss. These three treatments include laser photocoagulation, injection of the steroid triamcinolone, and more recently vascular endothelial growth factor (VEGF) antagonists into the eye, and vitrectomy, to remove the vitreous. However, there is no agreed medical approach to slow disease progression before the use of these rather invasive treatments.

2.12.4 Cardiovascular Disease

There is increased risk of cardiovascular disease (CVD) in diabetes, such that an individual with diabetes has a risk of myocardial infarction equivalent to that of non diabetic individuals who have previously had a myocardial infarction (Haffner *et al.,* 1998). CVD accounts for more than half of the mortality seen in the diabetic population (Haffner *et al.,* 1998 ; Laing *et al.,* 2003) and diabetes equates to an approximately threefold increased risk of myocardial infarction compared with the general population (Domanski *et al.,* 2002). Atherosclerosis is a complex process involving numerous cell types and important cell-to-cell interactions that ultimately lead to progression from the "fatty streak" to formation of more complex atherosclerotic plaques. These complex atherosclerotic plaques may then destabilize and rupture, resulting in myocardial infarction, unstable angina, or strokes. The precise mechanism is unknown. However, dysfunction within the endothelium is thought to be an important early contributor. The endothelium is crucial for maintenance of vascular homeostasis, ensuring that a balance remains between vasoactive factors controlling its permeability, adhesiveness, and integrity such as Angiotensin II (ANG II) and nitric oxide, but this balance appears compromised by diabetes (Okon, 2005). Localized abnormalities drive atherogenesis, where immune cells including macrophages and T cells can bind to the vessel wall (O'Brien, 1996). This initiates movement of low-density lipoprotein into the subendothelial space leading to foam cell and fatty streak formation (Glass, 2001) which are commonly seen at sites of turbulent flow such as bifurcations, branches, and curves (Ross, 1995). Ultimately, proliferation of smooth muscle cells and matrix deposition, often with associated necrosis, result in the formation of a complex atherosclerotic plaque, which may occlude the blood vessel at the site of formation such as in the coronary or femoral circulation or become an embolus occluding blood vessels at distant sites, commonly originating from within carotid vessels and reaching the cerebral circulation.

2.12.5 Erectile Dysfunction

Erectile dysfunction (ED) affects approximately 34 % to 45 % of men with diabetes and has a negative impact on quality of life (Eardley *et al.*, 2007). Studies have shown that alteration of the cyclic guanosinemonophosphate (cGMP /nitric acid (NO) pathway among men with diabetes with impaired vascular relaxation is related to endothelial dysfunction (Angulo *et al.,* 2006 ; Angulo *et al.,* 2009 ; Angulo *et.al.,* 2010). Among individuals with diabetes, the risk factors that may lead to ED include increasing age, poor glycaemic control, hypertension, cigarette smoking dyslipidemia, androgen deficiency states and cardiovascular disease (CVD) (Brock, and Harper, 2013).

2.13 Management of Diabetes Mellitus

2.13.1 Insulin

Insulin is lifesaving pharmacological therapy for people with type 1 diabetes. This hormone is primarily produced by recombinant DNA technology and is formulated either as structurally identical to human insulin or as insulin analogues (McGibbon *et al.*, 2013). Insulin regimens should be tailored to the individual's treatment goals, lifestyle, diet, age, general health, motivation, hypoglycaemia awareness status and ability for self-management. Social and financial aspects also should be considered. While fixed-dose regimens once were common and still may be used in some circumstances, they are not encouraged. The intensive insulin treatment of type 1 diabetes significantly delays the onset and slows the progression of microvascular and macrovascular complications (Nathan *et al.,* 2005; McGibbon *et al.,* 2013). The most successful protocols for type 1 diabetes rely on basal-bolus regimens that are used as a

component of intensive diabetes therapy. Basal insulin is provided by intermediate-acting insulin or a long-acting insulin analogue once or twice daily. Bolus insulin is provided by short-acting insulin or a rapid-acting insulin analogue given at each meal. Such protocols attempt to duplicate normal pancreatic insulin secretion. Prandial insulin dose must take into account the carbohydrate content and glycaemia index of the carbohydrate consumed, exercise around meal time. Regular insulin should ideally be administered 30 to 45 minutes prior to a meal. However, it has been known that pre-prandial injections achieve better postprandial control and, possibly, better overall glycaemia control (Jovanovic *et al.*, 2004; Schernthaner *et al.*, 2004; Garg *et al.*, 2005). Complications of insulin therapy include weight gain and hypoglycaemia. Hypoglycaemia may result from an inappropriately large dose, from mismatch between the peak delivery of insulin and food intake.

2.13.2 Oral hypoglycemic agents:

2.13.2.1 *Sulfonylureas*

The first generation of sulfonylureas includes tolbutamide, acetohexamide, tolazamide, and chlorpropamide. A second generation of sulfonylureas has emerged and includes glibenclamide, glipizide, gliclazide, and glimepiride .They are more potent than the first generation agents.

Sulfonylureas act by stimulating insulin release from pancreatic ß-cells by binding to sulfonylurea receptors (SUR) on the ß-cell plasma membrane, causing closure of adenosine triphosphate (ATP) sensitive potassium channels, leading to depolarization of the cell membrane. This in turn opens voltage-gated channels, allowing influx of calcium ions and subsequent secretion of preformed insulin granules (Bastaki, 2005).

2.13.2.2 *Metformin*

Metformin has a very low rate of causing lactic acidosis. When given alone or in combination with a sulfonylurea improves glycaemia control and lipid concentrations in patients who respond poorly to diet or to a sulfonylurea alone . Studies have shown that metformin improves insulin resistance in the liver, skeletal muscle, and adipose tissue, a major pathogenic component of type 2 diabetes (Bailey, 1992). The mechanism of action of metformin is not fully understood. It has been shown to increase peripheral uptake of glucose and to reduce hepatic glucose output by approximately 20-30% when given orally. Metformin has also been shown to decrease serum triglycerides and fatty acid concentrations and slows the rate of lipid oxidation, actions that indirectly inhibit gluconeogenesis (Wu *et al.,* 1990).

2.13.2.3 *Thiazolidinediones*

Thiazolidinediones (TZDs) are chemically and functionally unrelated to the other classes of oral antidiabetic agents. Two compounds in this class are currently in use. Rosiglitazone (Avandia) and pioglitazone (Actos) are the two thiazolidinediones in use. Thiazolidinediones are found in key target tissues for insulin action such as adipose tissue, skeletal muscle and liver. They exert their principal action by lowering insulin resistance in peripheral tissue, and also by lowering glucose production by the liver1 (Park et al., 1998).Thiazolidinediones increase glucose transport into muscle and adipose tissue by enhancing the synthesis and translocation of specific forms of the glucose transporter proteins.

2.13.2.4 *Meglitinide analogues*

The meglitinide analogues are a new class of drugs developed to improve early-phase insulin secretion, which is one of the earliest pathophysiolocal manifestations of type 2 DM. These are

derived from the meglitinide portion of sulfonylureas. The meglitinide analogues act on ß-cell receptors to stimulate insulin secretion by binding to the sulfonylurea receptor subunit and closing the K+ ATP channel. Probably at a site distinct from that of the sulfonylurea receptor Closure of the potassium channel leads to depolarization of ß-cell plasma membrane, which promotes influx of calcium ions through voltage-gated calcium channels, resulting in exocytosis of insulin granules (Bastaki et *al.,* 2005).

2.13.2.5 *α- Glucosidase inhibitors*

α-Glucosidase inhibitors have been developed specifically to delay the digestion of complex carbohydrates and decrease the postprandial rise in plasma glucose. α-Glucosidase inhibitors competitively block small intestine brush border enzymes that are necessary to hydrolyze oligosaccharides and polysaccharides to monosaccharides (Bischoff, 1995). Inhibition of this enzyme slows the absorption of carbohydrates; the postprandial rise in plasma glucose is blunted in both normal and diabetic subjects (Rabasa-Lhoret and Chiasson 1998). Examples of α-glucosidase inhibitors include acarbose, miglitol, and voglibose and all have similar pharmacological profiles.

2.14 Anti Diabetic Medicinal Plants

2.14.1 Cabbage

Cabbage is one of the most important crop plants of the species *(Brassica oleracea)*. It is a herbaceous, biennial, dicotyledonous flowering plant with leaves forming a characteristic compact head. The cabbage is differentiated into white head cabbage *(Brassica oleraceavar capitatasub.varalba)* and red head cabbage *(Brassica oleraceavar. capitata sub. var. rubra)*. It is widely consumed probably due to the its acceptable price, consumer preferences and availability at local markets The influence of cabbage consumption on human health is evident and is, in addition to being a source of vitamins and fibre, connected with secondary metabolites called glucosinolates, which are known to possess anticarcinogenic properties (Sarikamiş *et al.,* 2009). Cabbage is rich in phytochemicals such as polyphenolics, glucosinolates, carotenoids and vitamin C that have showed antioxidant, anticancer (Podsędek, 2007) and potential anti-obesity properties (Williams *et al.,* 2013). Raw cabbage juice can potentially heal an ulcer within 14 days of treatment. It is believed that the high level of glutamine is responsible for this healing effect. The ulcer healing properties of cabbage juice was verified in the 1950s in clinical trials both in United States and Europe (Priya, 2012). It has also been known that taking the combination of cabbage and Broccoli juice daily for 12 weeks significantly reduce the blood levels of low – density lipoproteins (Takai *et al.,* 2003). Epidemiological studies have shown cabbage to reduce the risk of lung, stomach, colorectal, breast, bladder and prostate cancer. It is postulated that this ability comes from glucosinolate proficiency for inhibiting cell division and inducing apoptosis (Priya, 2012). Cabbage has high level of anti – oxidants, such as vitamin C which has an immune boosting effect (Singh *et al*., 2006). Cabbage extract intake reduce serum cholesterol level and enhance faecal bile acid excretion and cholesterol 7 alpha hydroxylase activity, the rate limiting enzyme for bile acid biosynthesis in the microsomal fraction liver (Priya, 2012).

2.14.2 *Brassica juncea* (*Brassicaceae*)

The aqueous extract of Brassica juncea seeds in diabetic rats at a dose of 250, 350 and 450 mg/kg p.o. significantly reduced the blood glucose level. Further its showed insulinomimetic activity in the tested group signifying its anti diabetic effect (Thirumalai *et al.*, 2011).

2.14.3 *Azadirachta indica* (*Neem*)

Hydroalcoholic extracts of this plant showed antihyperglycaemic activity in streptozotocin treated rats and this effect is because of increase in glucose uptake and glycogen deposition in isolated rat hemidiaphragm. Apart from having anti-diabetic activity, this plant also has anti-bacterial, antimalarial, antifertility, hepatoprotective and antioxidant effects (Dheer and Bhatnagar, 2010).

2.14.4 *Zea mays* (*Corn Silk, Indian corn*)

Cornsilk is a proven diuretic. It is plentiful, cheap, and commonly used to treat cystitis, pyelitis, oliguria, and edema. It possesses hypoglycemic, antimicrobial, cholinergic and hypotensive properties. Cornsilk has more or less confirmed oral hypoglycemic activity. In one study the herb produced a constant hypoglycemic effect in starving rabbits. The active principle is not known (Amreen *et al.*, 2012).

2.14.5 *Allium sativum* (*garlic*)

This is a perennial herb cultivated throughout India. Allicin, a sulfur-containing compound is responsible for its pungent odour and it has been shown to have significant hypoglycemic activity. This effect is thought to be due to increased hepatic metabolism, increased insulin release from pancreatic beta cells and/or insulin sparing effect. Aqueous homogenate of garlic (10 ml/kg/d) administered orally to sucrose fed rabbits (10 g/kg/d in water for two months) significantly increased hepatic glycogen and free amino acid content, decreased fasting blood glucose, and triglyceride levels in serum in comparison to sucrose controls. S-allyl cystein sulfoxide (SACS), the precursor of allicin and garlic oil, is a sulfur containing amino acid, which controlled lipid peroxidation better than glibenclamide and insulin (Ravikumar *et al.,* 2010).

2.14.6 *Vernonia amygdalina* (*Bitter-tea vernonia*)

The anti-diabetic activity of the various combinations of metformin (50 mg/kg) and aqueous extracts of the leaves of *Vernonia amygdalina* (Asteraceae) (100 mg/kg) in diabetic rats were investigated. Extract and metformin at the ratios of 1:1 and 2:1 were given to both normoglycemic and diabetic. From the data it was found that, blood glucose level was decreased more significantly by the drug combination compared to the single treatment of drug in the diabetic rats (Amreen *et al.,* 2012).

2.14.7 *Sphenostylis stenocarpa* (Africa yam bean, Wild yam bean)

The methanolic extract of seeds of Sphenostylis stenocarpa (Leguminoseae) in diabetic rats at doses of 200, 400 and 600 mg/kg, p.o., significantly reduces the blood glucose level. Moreover, 600 mg/kg, p.o. was found to be more significant compared to other tested dose level (Okokon *et al.,* 2009).

2.14.8 *Phoenix dactylifera* (Date Palm, True Date, Medjool Palm, Medjool Date Palm)

The phoenix dactylifera (P. dactylifera) (Arecaceae) leaf extract in diabetes wistar rats at 100, 200, and 400 mg/kg, p.o. and its fractions at 50, 100, and 200 mg/kg, p.o. for 14 d treatment, significantly reduced blood glucose, TC, TG level and water intake and increase plasma insulin level significantly compare to control group. The data obtained from experiment showed that P. dactylifera have anti-diabetic potential (Mard *et al.,* 2010).

2.14.9 *Juglans regia* (walnut)

Anti-diabetic effects of methanolic extract of *Juglans regia* (*J. regia*) (*Juglandaceae*) leaves was estimated in diabetic male wistar rats at 250 mg/kg, p.o. for three weeks. J.ragia significantly decreases the blood glucose, TG and total cholesterol (TC) level. Further it increased gluthione peroxidise (GPX), super oxide dismuthase (SOD) and cell antibody level significantly and therefore signified its anti-diabetic potential (Teimoori *et al.,* 2010).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Materials

3.1.1 Chemicals

All chemicals and drugs used (Alloxan monohydrate, chloroform, ethanol, formalin, Glibenclamide and alloxan were purchased from (Sigma chemical Company St. Louis U.S.A.).

3.1.2 Equipment

Materials used were digital glucometer for blood glucose determination (Accu-check advantage, Roche Diagnostic, Company), weighing balance (GF 2000), dissecting set, syringes and needle.

3.1.3 Plant Material

Fresh cabbages were purchased in April 2014 from Kubani farm, Zaria, Kaduna. Authentication and identification was done by the Taxonomist in the Herbarium section of Department of Biological sciences, Ahmadu Bello University, Zaria and a voucher number (43382) was given and future reference.

3.1.4 Animals

A total of twenty five albino Wistar rats of both sexes weighing 100-150 g were used for this study. The animals were obtained from the Animal house of Department of Human Physiology, Ahmadu Bello University, Zaria. They were randomized into experimental and control groups and were kept in polypropylene cages. Standard animal feed made of pellets from grower's mash were provided to the animals. The rats were allowed access to drinking water *ad libitum* throughout the period of the study.

3.2 Methods

3.2.1 Preparation of cabbage supplement

Fresh leaves of cabbage was shed dried, ground, weighed and added to the rat feed (grower's mash) in 10, 25 and 50% percentage proportions.

3.2.2 Experimental induction of diabetes mellitus

Diabetes was induced by single intraperitoneal injection of alloxan monohydrate (Sigma St. Louis, M.S., U.S.A.) at a dose of 150 mg/kg body weight dissolved in 0.9% cold normal saline solution. The rats were fasted for 16 - 18 h before induction (Katsumata *et al*., 1999). Since alloxan is capable of producing fatal hypoglycaemia as a result of massive pancreatic insulin release, rats were treated with 20% glucose solution orally for 6 hrs after induction. The rats were then kept for the next 24 hrs on 5% glucose solution bottles in their cages to prevent hypoglycaemia (Dhandapani *et al*., 2002). After 72 hrs of alloxan treatment, venous blood was collected from the tail of the rats. Rats having fasting blood glucose level greater than 200 mg/dl were considered to be diabetic.

3.2.3 Experimental design

The study was carried out on alloxan induced Wistar rats. The animals were fasted for 16-18 hrs with free access to water prior to the induction of diabetes. After induction, the animals were randomly divided into 5 groups of 5 animals as follows:

Group 1: (Negative control): Diabetic rats which were given distilled water for four weeks.

Group 2: (Positive control): Diabetic rats that received 5mg/kg b/w of glibenclamide orally daily for 4 weeks.

Group 3: Diabetic rats that received 10% cabbage supplement for 4 weeks.

Group 4: Diabetic rats that received 25% cabbage supplement for 4 weeks.

Group 5: Diabetic rats that received 50% cabbage supplement for 4 weeks.

3.2.4 Determination of blood glucose levels

All venous blood samples were collected from the tail of the rats at weekly intervals for 4 weeks respectively. Fasting blood glucose levels was determined by using glucose oxidase method (Beach and Turner, 1958) using a digital glucometer (Accu-Chek Advantage, Roche Diagnostic, Germany). The results were expressed in mg/dl (Rheney and Kirk, 2000).

3.2.5 Collection and preparation of sera samples for lipid profile analysis

Four weeks after the experimental period, all animals were sacrificed after overnight fasting (16 – 18 hrs) and were exposed to chloroform soaked cotton wool placed in anaesthetic box covered with lid. Blood samples were drawn from the heart of each animal from all groups by cardiac puncture. The blood samples were collected in Eppendorf tubes, allowed to clot and the serum separated by centrifugation using Denley BS400 centrifuge (England) at 3000 g for 10 minutes. The supernatant (serum) collected was then used for lipid profile analysis.

3.2.6 Estimation of serum lipid profile

Serum lipid profile was determined spectrophotometrically, using enzymatic colometric assay kits (Randox, Northern Ireland) as follows:

3.2.6.1 Determination of serum total cholesterol

The serum level of total cholesterol (TC) was quantified after enzymatic hydrolysis and oxidation of the sample as described by method of Stein, 1987. 1000µl of the reagent was added to each of the sample and standard. It was then incubated for 10 minutes at 20-25 ^{0}C after mixing and the absorbance of the sample (A_{sample}) and standard ($A_{standard}$) were measured against the reagent blank within 30 minutes at 546 nm. The value of TC present in serum was expressed in mg/dl.

TC concentration = A *sample* /A *standard x* 196.86 mg/dl.

3.2.6.2 Determination of serum triglyceride

The serum triglyceride level was determined after enzymatic hydrolysis of the sample with lipases as described by method of Tietz, 1990. 1000µl of the reagent was added to each of the sample and standard. This was incubated for 10 minutes at 20-25 ^{0}C after mixing and the absorbance of the sample (A_{sample}) and standard ($A_{standard}$) was measured against the reagent blank within 30 minutes at 546 nm. The value of triglyceride present in the serum was expressed mg/dl.

TGL concentration = A *sample*/A *standard x* 194.0 mg/dl

3.2.6.3 Determination of serum high density lipoprotein (HDL)

The serum level of HDL was measured by the method of Wacnic and Albers, 1978. Low-density lipoproteins (LDL and VLDL) and chylomicron fractions in the sample were precipitated quantitatively by addition of phosphaturic acid in the presence of magnesium ions. The mixture was allowed to stand for 10 minutes at room temperature and centrifuged for 10 minutes at 4000

rpm. The supernatant represented the HDL fraction. The cholesterol concentration in the HDL fraction, which remained in the supernatant, was determined. The value of HDL was expressed in the unit of mg/dl.

3.2.6.4 Determination of serum low density lipoprotein (LDL)

The serum level of (LDL) was measured according to the protocol of Friedewald *et al*., 1972, using the equation below:

LDL-C = TC - (TGL/5 + HDL)

The value was expressed in mg/dl.

3.2.7 Determination of liver enzyme assay

Blood sample were collected via cardiac puncture, which was centrifuged to get serum for liver enzymes assay. These include: Alkaline phosphatase, Alanine aminotranferase and Aspartate aminotranferase using Reitman and Frankel method (1957).

3.2.7.1 Determination of serum aspartate aminotransferase (AST)

Aspartate aminotransferase was estimated by the method described by Bergmeyer and Walefeld (1978). 100 µL of the reagent was added to 100 µL of the samples and then mixed and incubated at 37^{o}C for 1 minute. The change in absorbance of the sample was measured per minute spectrophotometrically at the wavelength of 590 nm as follows:

AST activity (U/L) = ΔAB/min×1768

3.2.7.2 Determination of serum alanine aminotransferase(ALT)

This was estimated by the method described by Bergmeyer and Walefeld (1978). 1000µL of the reagent was added to 100 µL of the samples and then mixed and incubated at 37°C for 1minute. The change in absorbance of the sample will be measured per minute spectrophotometrically at the wavelength of 590 nm as follows:

AST activity (U/L) = ΔAB/min×1768

3.2.7.3 Determination of serum alkaline phosphatase (ALP)

Alkaline phosphatase was estimated by the method described by, Bowers and Mc Comb (1966). 0.5 ml of the reagent was added to 0.05 ml (50 µL) of the sample and then mixed and incubated at 37°C for 10 minutes. The change in absorbance of the sample was measured per minute spectrophotometrically at the wavelength of 590 nm as follows:

Absorbance of sample / Absorbance standard × Value of standard (U/L).

3.3 Statistical Analysis

All data were expressed as mean ± SEM. The data obtained were statistically analyzed using analysis of variance (ANOVA) with *Tukey's* multiple comparison post hoc tests to compare the level of significance between control and experimental groups. All statistical analysis were evaluated using SPSS version 17.0 software. The values of $P < 0.05$ were considered as significant.

CHAPTER FOUR

4.0 RESULTS

4.1 Effect of Cabbage Supplement of Blood Glucose Levels of Cabbage on Alloxan induced Diabetic Wistar Rats

Figure 4.1 show the result of the effect of three percentages of cabbage supplementation (10%, 25% and 50%) on alloxan induced diabetic Wistar rats. The result showed significant ($p < 0.05$) decrease in the blood glucose level at fourth week of cabbage intake with values of 108.80 ± 9.75 mg/dl for 10%, 95.20 ± 16.65 mg/dl for 25% and 59.80 ± 5.88 mg/dl for 50% cabbage supplement compared to the diabetic positive and negative control with values of 93.20 ± 5.23 and 281.80 ± 3.65 mg/dl, respectively.

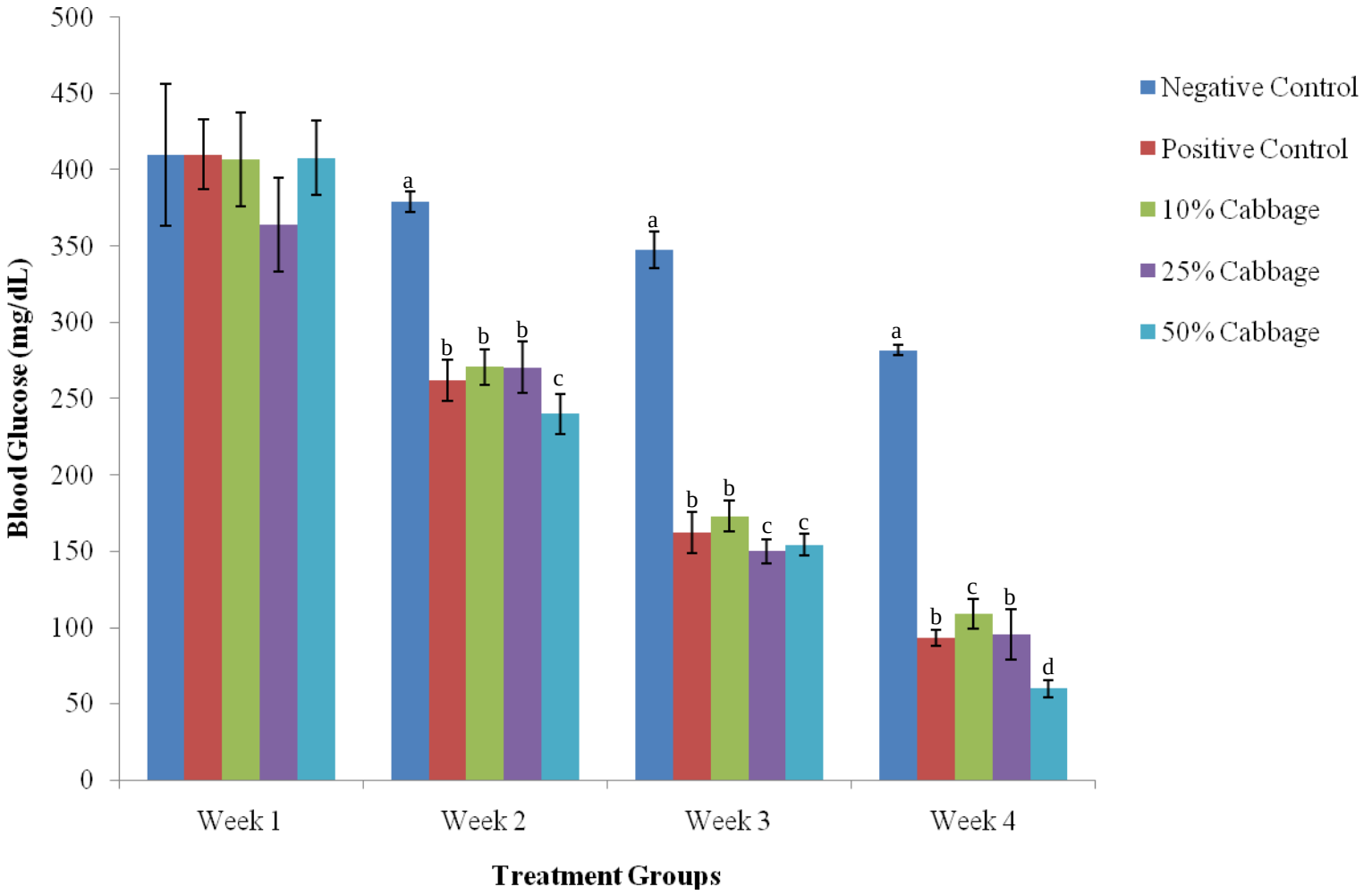


Figure 4.1: The effect of cabbage supplement on blood glucose levels on alloxan induced diabetic rats as compared with the control groups (n=5). Values with different errors bars are statistically significant ^{a,b,c} ($P < 0.001$).

4.2 The effect of cabbage Supplement on Lipid Profile on Alloxan Induced Diabetic Wistar Rats

4.2.1 Total cholesterol assessment

Figure 4.2 shows the effect of 10%, 25% and 50% cabbage supplementation on serum total cholesterol. The result showed significant ($P < 0.05$) decrease in total cholesterol with 25% (71.50 ± 9.95 g/L) and 50% (59.00 ± 13.70 g/L) cabbage supplement treated groups compared to the diabetic negative control with a value of 99.50 ± 10.15 g/L.

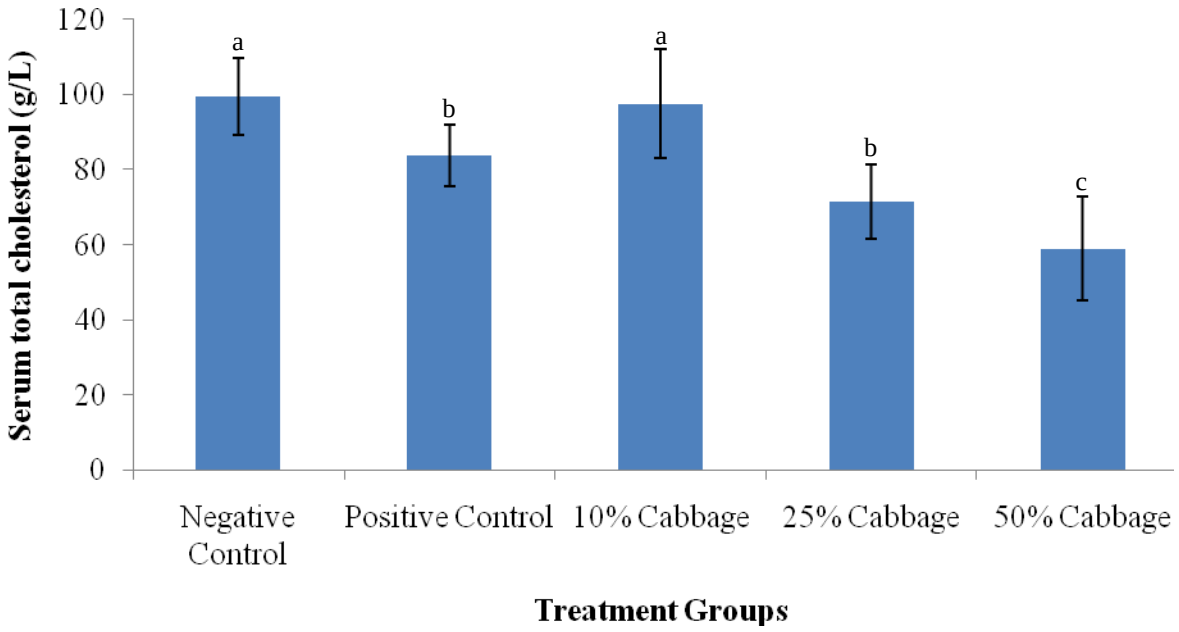


Figure 4.2: The effect of cabbage supplement on serum total cholesterol on alloxan induced diabetic Wistar rat as compared with control groups, n=5. Values with different errors bars are statistically significant ^{a,b,c} ($P < 0.05$).

4.2.2 Serum triglyceride

Figure 4.3 shows the results of the effect of cabbage supplement on serum triglyceride levels. The result showed significant ($P < 0.05$) decrease in total triglyceride with a values of 36.87 ± 4.89 g/L for 10%, 75.00 ± 15.30 g/L for 25% and 32.22 ± 2.97 g/L for 50% cabbage supplemented groups compared to the diabetic negative control group with a value of 115.42 ± 8.87 g/L and diabetes positive control group with a value of 163.40 ± 53.72 g/L, respectively.

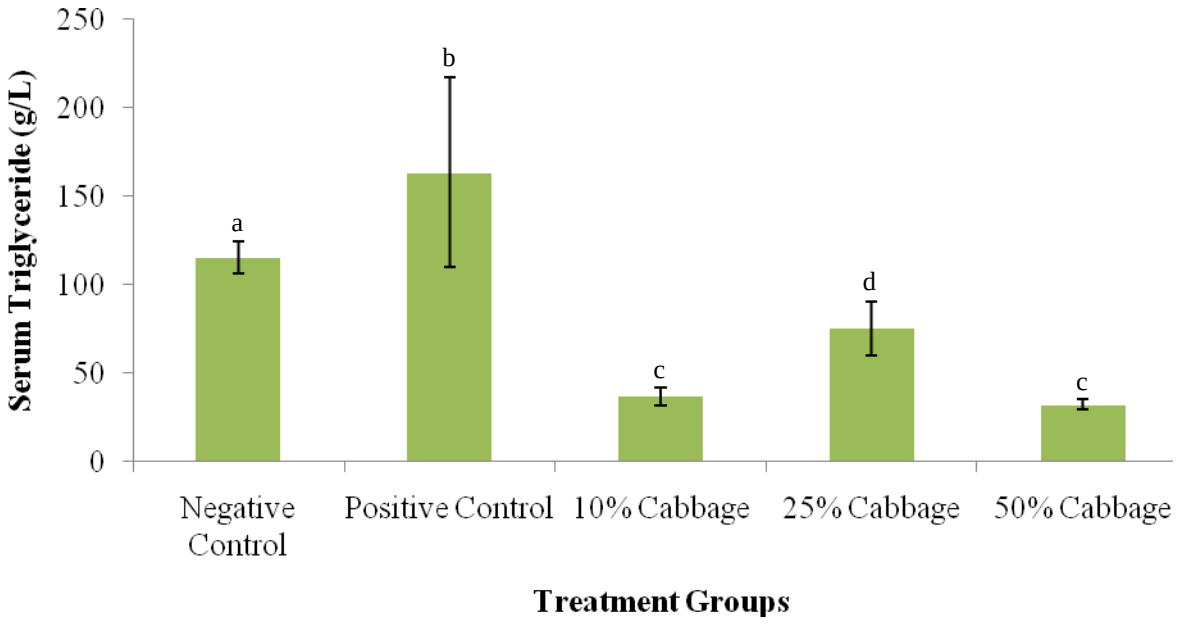


Figure 4.3: The effect of cabbage supplement on serum triglyceride on alloxan induced diabetic Wistar rats as compared with the control groups. Values with different errors bars are statistically significant ^{a,b,c} ($P < 0.05$).

4.2.3 High density lipoprotein

Figure 4.4 depicts the results of the effects of cabbage supplementation on alloxan induced diabetic Wistar rats. The results shows a significant ($P < 0.05$) increase in serum high density lipoprotein in rats fed with 10% cabbage supplement with a value of 39.59 ± 1.60 g/L and 50% cabbage supplement with a value of 65.12 ± 8.99 g/L when compared to diabetic positive control with a value of 29.43 ± 5.17 g/L.

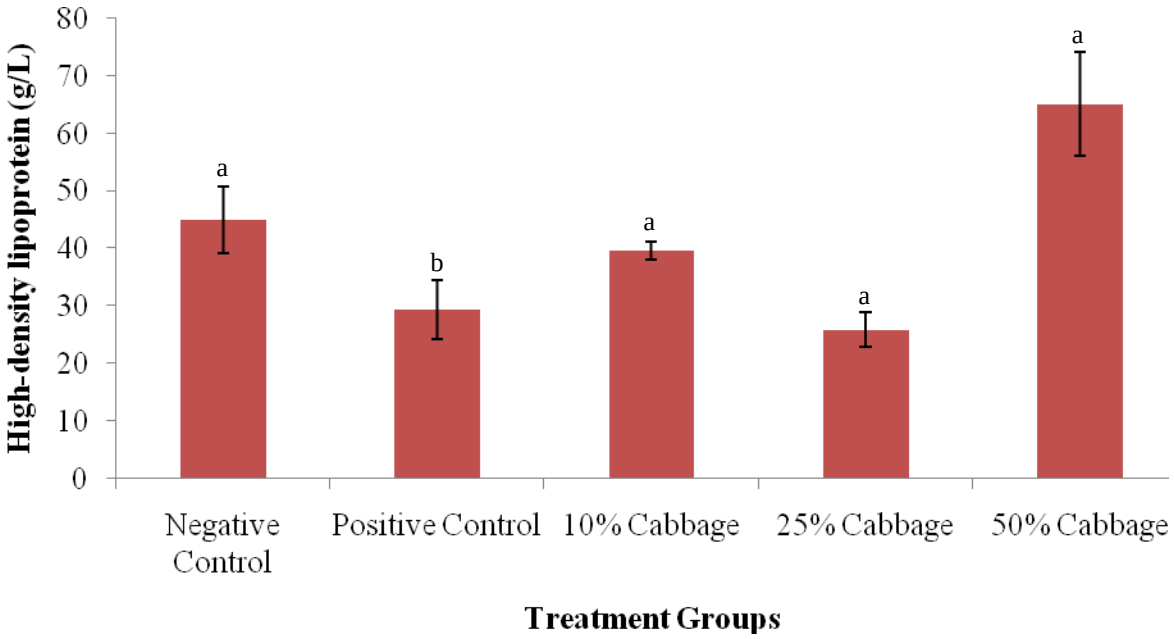


Figure 4.4: The effect of cabbage supplement on serum high density lipoprotein on alloxan induced diabetic Wistar rats as compared with the control groups. Values with different errors bars are statistically significant ^{a,b,c} ($P < 0.05$).

4.2.4 Low density lipoprotein

Figure 4.5 shows the results of cabbage supplementations of LDL. Cabbage supplement caused significant ($P < 0.05$) decrease in serum low density lipoprotein with a values of 52.64 ± 5.50 g/L, 45.68 ± 3.31 g/L and 44.93 ± 11.56 g/L, when compared to the diabetic negative control group with a value of 63.48 ± 3.88 g/L. The positive control group showed a significant ($P < 0.05$) decrease in low-density lipoproteins concentration compared with the three cabbage supplemented and negative control groups.

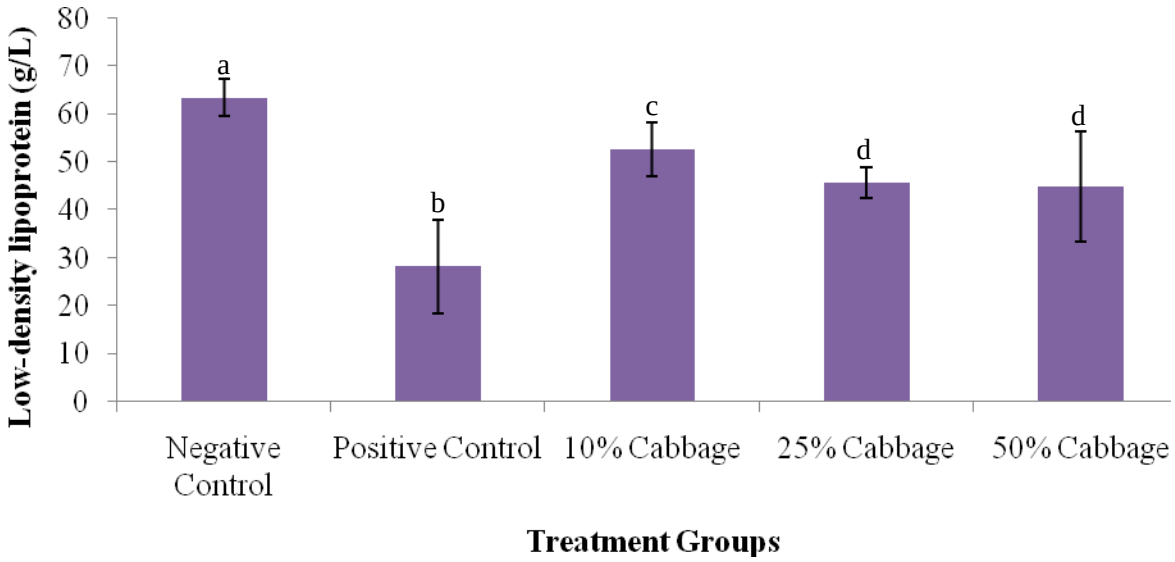


Figure 4.5: The effect of cabbage supplement on serum low density lipoprotein on alloxan induced Wistar rat as compared with the control group. Values with different errors bars are statistically significant ^{a,b,c} ($P < 0.05$).

4.3 Liver Enzymes

4.3.1 Alanine Aminotransferase

Figure 4.6 shows serum alanine aminotransferase activity in rats administered with different percentages of cabbage supplement. The result showed significant ($P < 0.05$) increase in alanine aminotransferase activity in all the treated groups when compared to the negative control group.

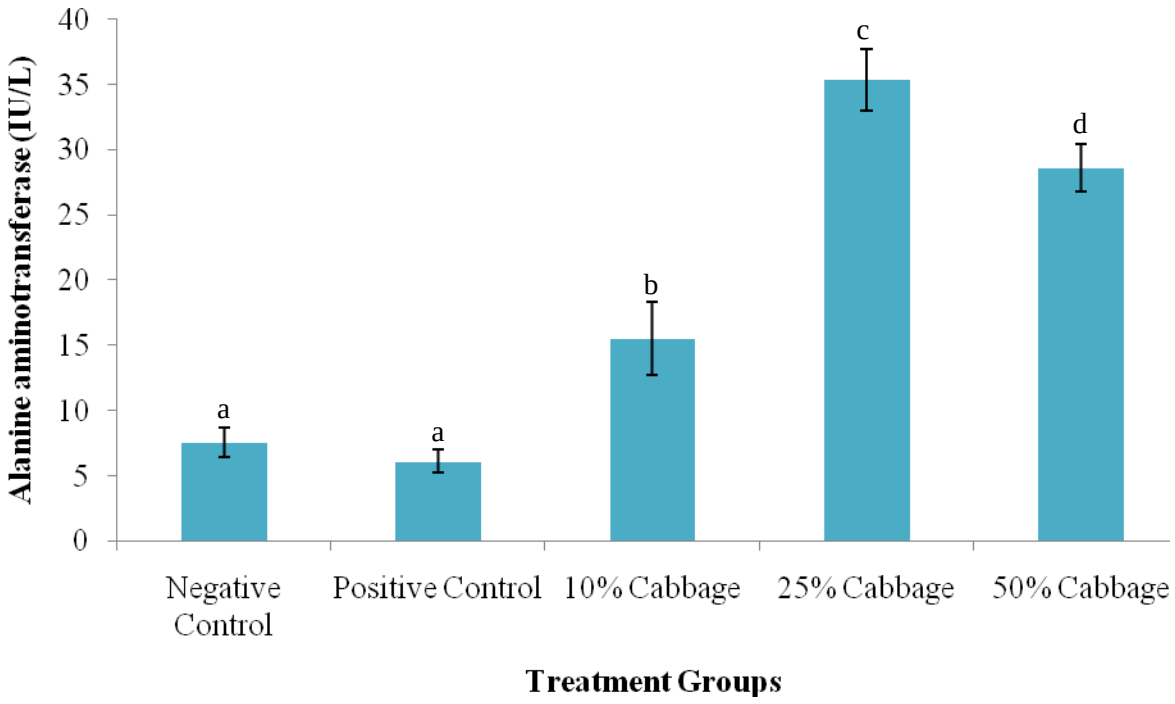


Figure 4.6: The effect of cabbage supplement on serum alanine aminotransferase on alloxan induced diabetic Wistar rats as compared with the control groups. Values with different errors bars are statistically significant ^{a,b,c} ($P < 0.05$).

4.3.2 Alkaline Phosphatase

Figure 4.7 shows the result of the serum activities of alkaline phosphatase in rats administered with cabbage supplement at different percentages. There was a significant ($P < 0.05$) decrease in the groups treated with 10% and 50% cabbage supplements compared with the control group.

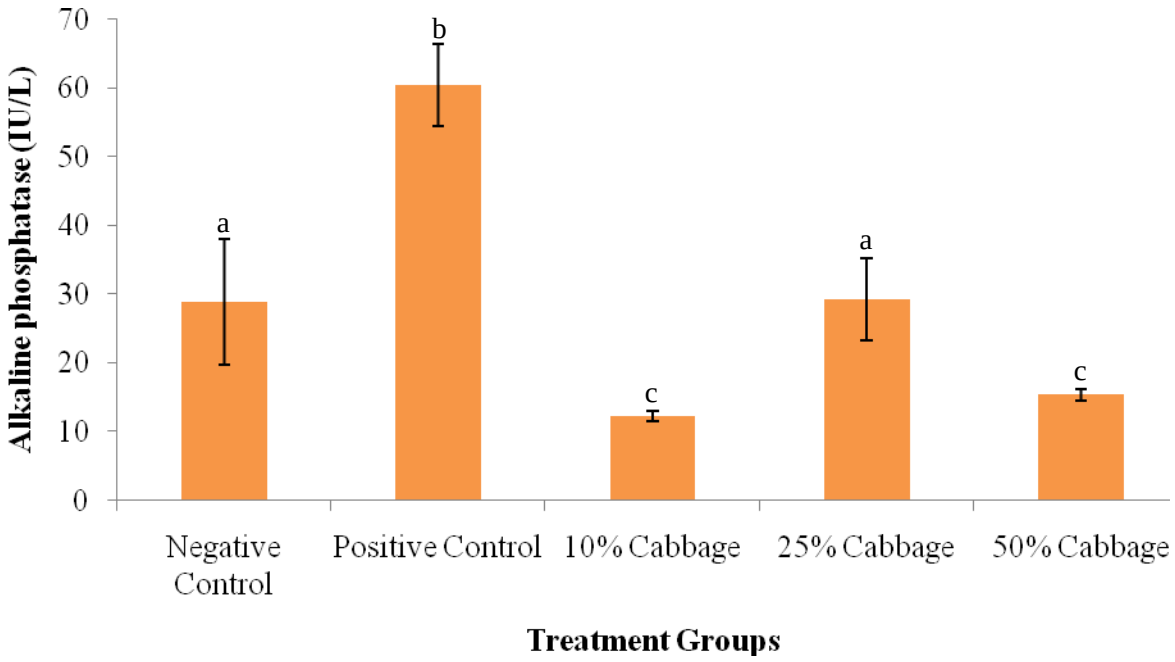


Figure 4.7: The effect of cabbage supplement on serum alkaline phosphatase on alloxan induced Wistar rats as compared to the control groups. Values with different errors bars are statistically significant ^{a,b,c} ($P < 0.05$).

4.3.3 Aspartate Aminotransferase

Figure 4.8 depicts the result of serum aspartate aminotransferase activity in rats administered with different percentages of cabbage supplement. There was a significant increase ($P < 0.05$) increase in aspartate aminotransferase activity in all the treated groups with cabbage supplement compared to the control group.

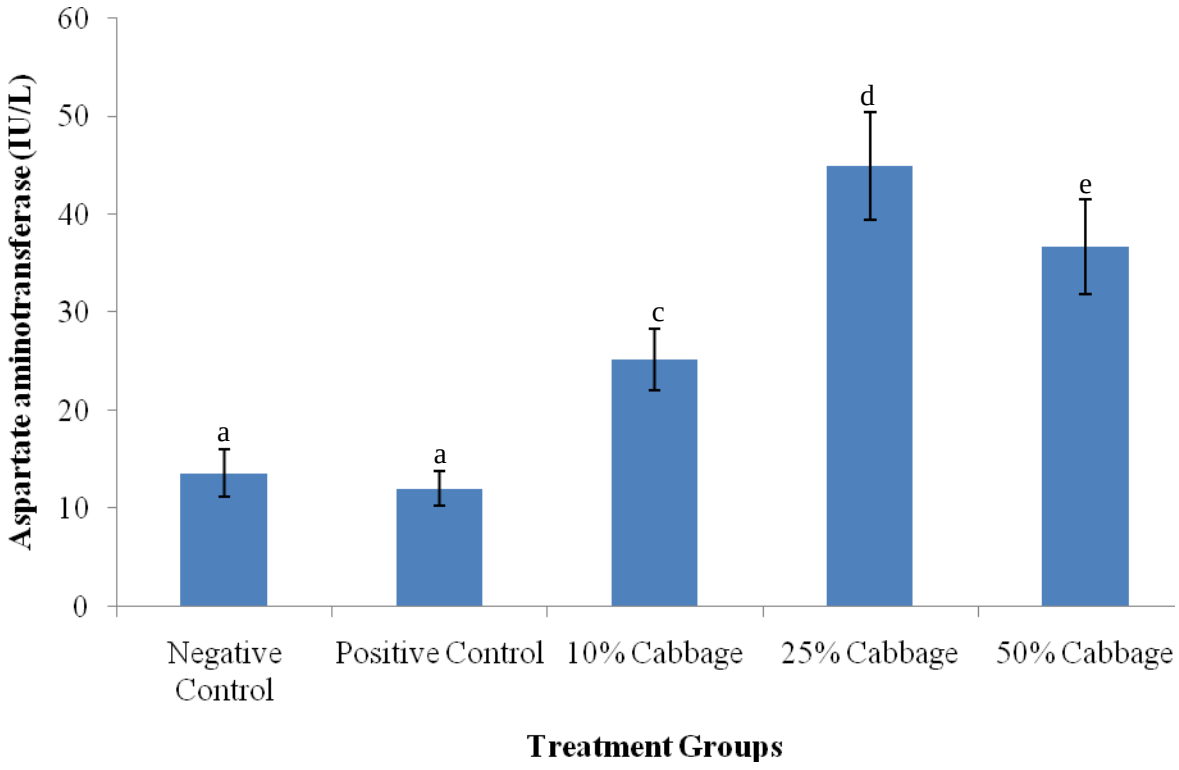


Figure 4.8: The effect of cabbage supplement on serum aspartate transferase on alloxan induced Wistar rats as compared with the control groups Values with different errors bars are statistically significant ^{a,b,c,d,e} ($P < 0.05$).

CHAPTER FIVE

5.0 DISCUSSION

Diabetes is a complex and complicated disease characterized by abnormalities in carbohydrate, protein, and lipid metabolism, causing a great challenge to physicians and other healthcare professionals who care for people with diabetes (Borokini, *et al.,* 2013; Cheng, 2013). Several studies have shown that vegetables play a protective role against the development of human diseases (Baboota *et al.,* 2013; Liu, 2013; Magrone *et al.,* 2013; Scicchitano *et al.,* 2014). Vegetables like cabbage belonging to the cruciferous family exert a protective effect against many chronic degenerative diseases, including cancer (Herr and Buchler, 2010; Manchali *et al.,* 2012). Glucosinolates are particularly abundant in cabbage and are believed to be the bioactive compound responsible for many of the biological effects attributed to them (Yeh and Yen, 2009; Herr and Buchler, 2010). However, cabbage is also an important source of other essential compounds such as polyphenols, carotenoids and phytosterols that exert an anti-inflammatory and antioxidant effect (Bacchetti *et al.,* 2014).

The observed decrease in serum levels of blood glucose in this study demonstrated the hypoglycaemic effect of cabbage supplement. These results agrees with the work of Gaafar *et al.* (2014), who demonstrated the hypoglycaemic effect of both extract of white and red cabbages. The decrease may be due to the effect of different polyphenolic compound present in cabbage. These compounds, for example flavonoids and other alkaloids have been reported to have anti-diabetic properties. Our findings also agreed with that of Alsuhaibani, (2013) who demonstrated

the hypoglycaemic and hypolipidaemic activities of red cabbage and manganese for the treatment of diabetes in rats. Nizamutdinova *et al.* (2009), also reported that the administration of anthocyanins (also found in cabbage) markedly decreased glucose levels and increase utilization of glucose by tissues in diabetic rats.

Lipid profile results showed abnormally high level of serum lipids in the diabetic subjects compared to the supplemented groups. The increased in lipid profile may be due to an increase in the mobilization of free fatty acids from the peripheral fat depots since insulin inhibits the hormone-sensitive lipase (Murali *et al.,* 2002). Insulin deficiency or insulin resistance may be responsible for dyslipidaemia because insulin has an inhibitory action on HMG-CoA reductase, a key rate-limiting enzyme responsible for the metabolism of cholesterol-rich LDL-particle. An abnormality in lipid profile is one of the most common complications in diabetes and is associated with an increased risk of coronary heart disease (Maghrani *et al.*, 2004). Since lipid abnormalities accompanied with premature atherosclerosis are the major causes of cardiovascular disease in patients with diabetes, ideal management for diabetes, should have a favourable effect on lipid profile (Kesari *et al.*, 2007).

The observed significant decreased in total triglycerol, total cholesterol, and LDL and an increase in the HDL level indicates that cabbage supplement have an antilipidaemic effect. This result is in accordance with the findings of Waqar and Mahmood, (2010), who reported that ethanol extract of cabbage caused reduction in serum LDL, with increased HDL significantly. The glucose lowering action of the cabbage might be due to improved lipid metabolism apart from direct interaction with glucose homeostasis. The TG-lowering property could indirectly

contribute to the overall antihyperglycemic activity through glucose-fatty acid cycle mechanism (Hue and Taegtmeyer, 2009). Epidemiological data as well as *in vitro* studies strongly suggest that cabbage having antioxidant phytochemical compound have strong protective effects against major degenerative diseases including cardiovascular diseases, antihyperglycemic and hypocholesterolaemic (Baynes 1991; Roman-Ramos *et al.*, 1995 and Komatsu *et al.*, 1998).

The liver is regarded as the major organ of metabolism. The liver enzymes alanine aminotransferase (ALT), aspartate aminotransferase (AST) and alkaline phosphatase (ALP) are considered as biomarkers of liver toxicity and are used in the evaluation of hepatic disorders (Badole and Bodhankar, 2010).The result showed that serum level of ALT and AST in the cabbage supplement group were significantly higher than the control. While that of ALP was lower than that of control. These results is in accordance with the work of Gaafar *et al.* (2014), who reported a significant increase in serum level of ALT and AST in diabetic rats treated with extract of both white and red cabbages in comparison with the control but in contrast with the ALP serum level result which also showed a significant increase. Our findings also disagree with the work of Maha and El-Motaleb, (2012) and Alsuhaibani, (2013), they observed a significant decrease in serum liver enzymes (ALT, AST and ALP) in diabetic rats who were treated with red cabbage powder and red cabbage extract. The increase in the activities of these serum enzymes may be due to the fact that cabbage when eaten in excess can be giotrogenic, and in goitrogenic conditions, liver enzymes activities are increase. This may explain the marked increase in the liver enzymes of AST and ALT (Oh and Hustead 2011).

CHAPTER SIX

6.0 SUMMARY, CONCLUSION AND RECOMMENDATIONS

6.1 Summary

Cabbage supplementation has been observed to possess an anti hyperglycaemic and anti hyperlipidaemic effect, indicated by the significant reduction in serum glucose and lipid levels in the groups of animals supplemented with different percentages of cabbage. Cabbage supplementation was also observed to increase liver enzymes (ALP, ALT and AST) which are biomarkers of liver toxicity although this may be as result of the diabetic state or due to the goitrogenic effect the cabbage might have.

6.2 Conclusion

The results obtained from the study demonstrated that cabbage supplementation alleviated hyperglycaemia and hyperlipidaemia associated with experimentally induced type2 diabetes in Wistar rats. However, there is the need for more investigation on the toxicological profile of cabbage supplementation on the liver.

6.3 Recommendations

Based on the results obtained in this study, the following recommendations made:

a. Further studies should be carried out to investigate the precise mechanism(s) of cabbage underlying its hypoglycaemic and hypolipidaemic effects.

b. Studies should be carried out on the effect of fermented cabbage supplement on serum glucose and some physiological profiles.

6.4 Contribution to Knowledge

1. The result of the present study shows that 10%, 25% and 50% cabbage supplement significantly reduced the blood glucose level of diabetic Wistar rats.
2. The result of this study revealed that cabbage supplement lowers total cholesterol, triglyceride and low density lipoprotein of diabetic Wistar rats after administration for four weeks.
3. The result of this study revealed that cabbage supplementation increased some of the liver enzymes which may indicate toxicity to the liver.

REFERENCES

Abougalambou S.S.I., Mohamed M., Sulaiman S.A.S., Abougalambou A.S., and Hassali M. A. (2010). Current clinical status and complications among type 2 diabetic patients in Universiti Sains Malaysia hospital. *International Journal of Diabetes Mellitus.* 2(3): 184-188.

Adeniyi, A. F., Adeleye, J. O. and Adeniyi, C. Y. (2011). Diabetes, sexual dysfunction and therapeutic exercise: a 20 year review. *Current Diabetes Reveiw,* 6: 201-206.

Aguilar-Bryan L., and Bryan, J. (2008). Neonatal diabetes mellitus. *Endocrine Reviews*, 29(3):265–291.

Akhtar, M.A., Rashid, M., Mir Imam Ibne Wahed, M.I., Islam, R., Shaheen, S.M., Islam .A., Amran, S., and Ahmed, M. (2007).Comparison of Long-term Antihyperglycemic and Hypolipidemic Effects Between Coccinia cordifolia (Linn.) And Catharanthus roseus (Linn.) InAlloxan-induced Diabetic Rats. *Research Journal of Medicine and Medical Sciences,* 2(1): 29-34.

Akhtar, M.A., Rashid, M., Wahed, M.I., Islam R., Shaheen, S.M., Islam, .A., Amran, .A., and Ahmed, .M. (2007). Comparison of Long-term Antihyperglycemic and Hypolipidemic Effects Between Coccinia cordifolia (Linn.) And Catharanthus roseus (Linn.) In Alloxan-induced Diabetic Rats. *Journal of Medicine and Medical Sciences*, 2(1): 29-34.

Alsuhaibani, A. M. A. (2013). Hypoglycemic and HypolipidemicActivities of Red Cabbage and Manganese in Diabetic Rats. *Journal of American Science,* 9(10): 13.

Amreen, F., Parul, A. and Prem, P. S. (2012). Herbal option for diabetes: an overview. *Asian Pacific Journal of Tropical Disease*, 2 (1): 536–544.

Angulo, J., Cuevas, P., Fernández, A., Allona, A., Moncada, I., Martín-Morales, A., La Fuente, J.M. and de Tejada, I.S. (2006). Enhanced thromboxane receptor-mediated responses and impaired endothelium-dependent relaxation in human corpus cavernosum from diabetic impotent men: role of protein kinase C activity. *Journal of Pharmacology and Experimental Therapeutics,* 319(2): 783-789.

Angulo, J., González-Corrochano, R., Cuevas, P., Fernández, A., La Fuente, J. M., Rolo, F., Allona, A. and Sáenz de Tejada, I. (2010). Diabetes exacerbates the functional deficiency of NO/cGMP pathway associated with erectile dysfunction in human corpus cavernosum and penile arteries. *Journal of Sex Medicine,* 7(2): 758-768.

Angulo, J., Peiró, C., Cuevas, P., Gabancho, S., Fernández, A., González-Corrochano, R., La Fuente, J. M., Baron, A. D., Chen, K. S. and de Tejada, I. S.(2009).The novel antioxidant, AC3056 (2,6-di-t-butyl-4-((dimethyl-4-methoxyphenylsilyl)methyloxy)phenol), reverses erectile dysfunction in diabetic rats and improves NO-mediated responses in penile tissue from diabetic men. *Journal of Sex Medicine,* 6(2): 373-387.

Asadujjaman, M., Hossain, M.S., Khan, M.R.I., Anisuzzaman, A.S.M., Ahmed, M., and Islam, A. (2011). Antihyperglycemic and Glycogenesis Effects of Different Fractions of Brassica Oleracea in Alloxan Induced Diabetic Rats. *International Journal of Pharmaceutical Sciences and Reaserch*. 2(6) 1436-1442.

Ayodele, O. E., Alebiosu, C. O. and Salako, B. L. (2004). Diabetic nephropathy - a review of the natural history, burden, risk factors and treatment. *Journal of the National Medical Association,* 96(11): 1445–1454.

Baboota, R. K., Bishnoi, M., Ambalam, P., Kondepudi, K. K., Sarma, S. M., Boparai, R. K. and Podili, K. (2013). Functional food ingredients for the management of obesity and associated co-morbidities – a review. *Journal of Functional Foods,* 5: 997-1012.

Bacchetti, T., Tullii, D., Masciangelo, S., Gesuita, R., Skrami, E., Brugè, F., Silvestri, S., Orlando, P., Tiano, L. and Ferretti, G.(2014). Effect of black and red cabbage on plasma carotenoid levels, lipid profile and oxidized low density lipoprotein. Journal of Functional Foods, 8: 128-137.

Badawi A., Klip A., Haddad P., Cole D.E.C., Bailo B.G., El-Sohemy A., and Karmali M. (2010).Type 2 diabetes mellitus and inflammation: Prospects for biomarkers of risk and nutritional intervention. *Journal of Diabetes, Metabolic syndrome and Obesity,* 3: 173-186.

Badole, S.L. and Bodhankar, S.L. (2010). Antidiabetic activity of cycloart-23-ene-3beta, 25-diol (B2) isolated from Pongamia pinnata (L. Pierre) in streptozotocin-nicotinamide induced diabetic mice.*European Journal of Pharmacology,* 632(1-3):103-109.

Bailey, C. J. (1992). *Biguanides* and NIDDM. *Diabetes Care,* 15: 755-772.

Bailey, C.J., and Day, C. (1989). Traditional Plant Medicines as Treatments for Diabetes. *Diabetes Care*, Vol. 12, No. 8, pp 553-564.

Baltazar, J.C., Ancheta, C.A., Aban, I.B., Fernando, R.E.,and Baquilod, M.M. (2004). Prevalence and correlates of diabetes mellitus and impaired glucose tolerance among adults in Luzon, Philippines . *Diabetes Research and Clinical Practice*. (64):107–115.

Barthel, P., Bauer, A., Muller, A., Junk, N., Huster, K. M., Ulm, K., Malik, M. and Schmidt, G. (2011). Reflex and tonic autonomic markers for risk stratification in patients with type 2 diabetes surviving acute myocardial infarction. *Diabetes Care,* 34: 1833-1837.

Bastaki, S. (2005). Diabetes mellitus and its treatment. *International Journal of Diabetes and Metabolism,* 3: 111-134.

Baynes, J.W. (1991).Role of oxidative stress in development of complications in diabetes. *Diabetes,* 40: 405-412.

Beach, E.F., and Turner, J.J. (1958). An enzymatic method for glucose determination in body fluids. *Clinical Chemistry,* 4:468.

Bergmeyer, H., and M. Walefeld, M.1978. Méthodecinétique pour la détermination du TGO et TGP sans phosphate de pyridoxal *Clinica Chimica Acta*, 24: 58.

Bhuva, F., and Addepalli, V. (2011). Cognitive Dysfunction in Diabetic Rats. Effect of Minocycline. *Pharmacologyonline,* 1: 1255-1261.

Bischoff, H. (1995). The mechanism of a-glucosidase inhibition in the management of diabetes. *Clinical and Investigative Medicine,* 18: 303-311.

Boden, G., Lebed, B., Schatz, M., Homko, C. and Lemieux, S. (2001). Effects of acute changes of plasma free fatty acids on intramyocellular fat content and insulin resistance in healthy subjects. *Diabetes,* 50(7): 1612-1617.

Borokini, T. I., Ighere, D. A., Clement, M., Ajiboye, T. O. and Alowonle, A. A. (2013). Ethnobiological Survey of Traditional Medicine Practice for The Treatment of Piles and Diabetes Mellitus in Oyo State. *Journal of Medicinal Plants Studies,* 1(5): 30-40.

Bowers., G.N.J. and Mc-Comb.,R.B. (1966). A continuous spectrophotometric method for measurement the activity of serum alkaline phosphatase. *Clinical Chemistry,* 12: 73.

Brock, G. and Harper, W. (2013). Erectile Dysfunction. *Canadian Diabetes Association,* 37: 1499-2671.

Callaghan B.C Cheng H.T., Stables C.L., Smith A.L., and Feldman E.L. (2012). Diabetic neuropathy: clinical manifestations and current treatments. *The Lancet Neurology,* 11(6): 521-534.

Cheng A.Y.Y. (2013). Clinical Practice Guidelines. *Canadian Journal of Diabetes.*37: 3-13.

Chinenye S., and Young E. (2011). State of Diabetes Care in Nigeria: A Review. *The Nigerian Health Journal,* 11(4): 101-106.

Cinti, S., Mitchell, G., Barbatelli, G., Murano, I., Ceresi, E., Faloia, E., Wang, S., Fortier, M., Greenberg, A.S., and Obin, M.S.(2005). Adipocyte death defines macrophage localization and function in adipose tissue of obese mice and humans. *Journal of Lipid Research,* 46(11):2347-55.

Cukierman, T., Gerstein, H.C., and Williamson, J.D. (2005). Cognitive decline and dementia in diabetes-systematic overview of prospective observational studies. *Diabetologia,* 48: 2460-2469.

Daneman, D. (2006). Type 1 diabetes. *The Lancet,* 367(9513): 847-858.

Dhandapani, S., Ramasamy, S.V., Rajagopal, S., and Namasivayam, N. (2002). Hypolipidemic effect of *Cuminumcyminum* L. on alloxan-induced diabetic rats. *Pharmacology Research***,** 46 (3): 251-255.

Dhaon P.,and Shah V.N. (2014). Type 1 diabetes and osteoporosis: A review of literature. *Indian Journal of Endocrinology and Metabolism,* 18(2): 159-165.

Dheer, R. and Bhatnagar, P. (2010). A study of the antidiabetic activity of Barleria prionitis Linn. *Indian Journal of Pharmacology,* 42: 70-73.

Dib, S. A. and **Gomes, M. B. (2009).** Etiopathogenesis of type 1 diabetes mellitus: prognostic factors for the evolution of residual β cell function. *Journal of Diabetology and Metabolic syndrome.* **1**:25.

Duby, J. J., Campbell, R. .K., Setter, S. M., White, J. R. and Rasmussen, K. A. (2004). Diabetic neuropathy: an intensive review. *American Journal of Health-System Pharmacy,* 61(2): 160-173.

Eardley, I., Fisher, W., Rosen, R. C., Niederberger, C., Nadel, A. and Sand, M. (2007). The multinational Men's Attitudes to Life Events and Sexuality study: the influence of diabetes on self-reported erectile function, attitudes and treatment-seeking patterns in men with erectile dysfunction. *International Journal of Clinical Practice,* 61(9):1446-1453.

Enas, A.K., and Atif, I.A. (2010). Study the Possible Protective Influence of White Cabbage and Septilin on the Cardiac Muscle of Male Rats exposed to Gamma Radiation. *Researcher*; 2 (6): 1-14.

Evans T.C., and Capell P. (2000). Diabetic Nephropathy. *C**linical Diabetes,* **18: 1.**

Forbes, J.M., and Cooper, M.E. (2013). Mechanisms of diabetic complications. *Physiological Reviews,* 93:137-188.

Friedewald, W.T., Levey, R., and Fradrickson, D.S. (1972). Estimation of concentration of Low density lipoprotein cholesterol in plasma without the use of preparative ultracentrifugation. *Clincal Chemistry*. 19; 449-452.

Fumeron, F., Aubert, R., Siddiq, A., Betoulle, D., Péan, F., Hadjadj, S., Tichet, J., Wilpart, E., Chesnier, M. C., Balkau, B., Froguel, P. and Marre, M. (2004). Adiponectin gene polymorphisms and adiponectin levels are independently associated with the development of hyperglycemia during a 3-year period: the epidemiologic data on the insulin resistance syndrome prospective study. *Diabetes*, 53(4): 1150-1157.

Gaafar, A. A., Aly, H. F., Zeinab, A. Salama, Z. A. and Mohamed, N. Z. (2014). Hypoglycemic effects of white cabbage and red cabbage (*Brassica Oleracea)* in STZ induced type-2 diabetes in rats. *World Journal of Pharmaceutical Research,* 3(4): 1583-1610.

Garg, S. K., Rosenstock, J. and Ways, K. (2005). Optimized basal-bolus insulin regimens in type1 diabetes: insulin glulisine versus regular human insulin in combination with basal insulin glargine. *Endocrine Practice* 11: 11-7.

Gatling, W., Bingley, P. J., and Patterson, C. C. (2003). Mortality from heart disease in a cohort of 23,000 patients with insulin-treated diabetes. *Diabetologia,* 46: 760-765.

Gilbert, R.E., Rabi, D., LaRochelle , P., Leiter, L.A., Jones, C., Ogilvie, R., Tobe, S., Khan, N., Poirier., L., and Woo, V. (2013). Treatment of Hypertension. *Canadian Journal of Diabetes,* 37: 117-118.

Glass, C. K. and Witztum, J. L. (2001). Atherosclerosis: the road ahead. *Cell,* 104: 503-516.

Grulich-Henn, J., Wagner ,V., Thon, A., Schober, E., Marg, W., Kapellen, T.M., Haberland, H., Raile, K., Ellard, S., Flanagan, S.E., Hattersley ,A.T., and Holl, R.W. (2010). Entities and frequency of neonatal diabetes: data from the diabetes documentation and quality management system (DPV). *Diabetic Medicine,* 27(6):709–712.

Grundy, S. M., Cleeman, I. J, Daniels, S. R., Donato, K. A., Eckel, R. H., Franklin B. A., Gordon, D. A., Krauss, R. M, Savage, P. J., Smith, J. S. C., Spertus, J. A. and Costa, F. (2005). Diagnosis and management of the metabolic syndrome: an American Heart Association/National Heart, Lung, and Blood Institute scientific statement, *Circulation*, 112(17): 2735–2752.

Haffner, S.M., Lehto, S., Ronnemaa, T., Pyorala, K. and Laakso, M. (1998). Mortalityfrom coronary heart disease in subjects with type 2 diabetes and in nondiabetic subjects with and without prior myocardial infarction. *New England Journal of Medicine,* 339: 229-234.

Halberg, N., Wernstedt-Asterholm, I. and Scherer, P. E. (2008).The adipocyte as an endocrine cell, *Endocrinology and Metabolism Clinics of North America*, 37(3): 753–768.

Harande Y. I. (2011). Exploring the literature of diabetes in Nigeria: a bibliometrics study. *African Journal of Diabetes Medicine*. 19(2): 8-11.

Herr, I. and Buchler, M. W. (2010). Dietary constituents of broccoli and other cruciferous vegetables: implications for prevention and therapy of cancer. *Cancer Treatment Reviews*, 36: 377-383.

Hotamisligil, G. S., Peraldi, P., Budavari, A., Ellis, R., White, M. F. and Spiegelman, B. M. (1996). IRS-1-mediated inhibition of insulin receptor tyrosine kinase activity inTNF-α-and obesity-induced insulin resistance. *Science*, 271(5249): 665–668.

Hu F.B., Manson J.E., Stampfer M.J., Colditz G., Simin Liu S., Solomon C.G., and Willett W.C. (2001). Diet, Lifestyle, and the Risk of Type 2 Diabetes Mellitus in Women. *New England Journal of Medicine*, 345: 790-797.

Hu, F. B., van Dam, R. M. and Liu, S. (2001). Diet and risk of type II diabetes: the role of types of fat and carbohydrate. *Diabetologia*, 44: 805-817.

Hue, L., and Taegtmeyer, H. (2009). The Randle cycle revisited: a new head for an old hat. *American Journal of Physiology, Endocrine and Metabolism*, 297(3):E578-91.

Jacobs, M ., van Greevenbroek, M. M., van der Kallen, C. J., Ferreira, I., Blaak, E. E., Feskens, E. J., Jansen, E. H., Schalkwijk, C. G., and Stehouwer, C. D. (2009). Low-grade inflammation can partly explain the association between the metabolic syndrome and either coronary artery disease or severity of peripheral arterial disease: the CODAM study. *European Journal of Clinical Investigation*, 39(6): 437-444.

Jimenez-Cohl, P., Grekin C., Leyton, C., Vargas C., and Villaseca, R. (2012). Thermal Threshold: Research Study on Small Fiber Dysfunction in Distal Diabetic Polyneuropathy *Journal of Diabetes Science and Technology*, 6: 1.

Jordan D.N., and Jordan J.L. (2012) .Paediatric type 2 diabetes mellitus complications: a systematic review of the literature. *Journal of Diabetes Research & Clinical Metabolism,*

Jovanovic, L., Giammattei, J., Acquistapace, M., Bornstein, K., Sommermann, E. and Pettitt, D. J. (2004). Efficacy comparison between preprandial and postprandial insulin aspart administration with dose adjustment for unpredictable meal size. *Clinical Therapeutics,* 26: 1492-1497**.**

Kataria, A., Gopi, R. P., Mally, P. and Shah, B. (2014). Neonatal diabetes mellitus: current perspective. *Research and Reports in Neonatology*, 4: 55- 64.

Katsumata, K.Y., Katsumata, T.O., and Katsumata, K. (1999): Potentiating effects of combined usage of three sulfonylurea drugs on the occurrence of alloxan-induced diabetes in rats. *Hormone Metabolism Research*, 25: 125-126.

Kesari, A.N., Kesari, S., Singh, S.K., Gupta, R.K., and Watal, G. (2007). Studies on the glycaemia and lipidaemic effect of Murraya koenigii in experimental animals. *Journal of Ethnopharmacol,* 112(2):305–11.

Komatsu, W., Miura, Y., and Yagasaki, K.. (1998). Suppression of hypercholesterolemia in hepatomabearing rats by cabbage extract and its component, S-methyl-L-cysteine sulfoxide. *Lipids*, 33(5):499-503.

Lago, R.M., Singh, P.P and Nesto, R.W. (2007). Diabetes and hypertension. *Nature Clinical Practice Endocrinology and Metabolism*, 3(10): 667.

Laing, S. P., Swerdlow, A. J., Slater, S. D., Burden, A. C., Morris, A., Waugh, N. R., Lau, D. C. W., Dhillon, B., Yan, H., Szmitko, P. E. and Verma, S. (2005). Adipokines: molecular links between obesity and atheroslcerosis. *The American Journal of Physiology, Heart and Circulatory Physiology*, 288(5): 2031-2041.

Lau, D. C. W., Dhillon, B., Yan, H., Szmitko,P. E. and Verma, S. (2005). Adipokines: molecular links between obesity and atheroslcerosis. *The American Journal of Physiology -Heart and Circulatory Physiology*, 288(5): 2031-2041.

Linsay, R. S. (2009). Gestational diabetes: causes and consequences. *The British Journal of Diabetes and Vascular Disease*, 9(1): 27-31.

Liu, R. H. (2013). Dietary bioactive compounds and their health implications. *Journal Food Science,* 78: 18-25.

Maahs, D. M., West, N. A., Lawrence J. M. and Mayer-Davis, E. J. (2011) .Epidemiology of Type 1 Diabetes*. Endocrinology and Metabolism Clinics of North America, 30(3): 481-497.*

Maghrani, M., Lemhadri, A., Zeggwagh, N.A., El Amraoui, M., Haloui, M., Jouad, H., and Eddouks M. (2004). Effects of an aqueous extract of Triticum repens on lipid metabolism in normal and recent-onset diabetic rats. *Journal of Ethnopharmacology,* 90(2-3):331–7.

Magrone, T., Perez de Heredia, F., Jirillo, E., Morabito, G., Marcos, A. and Serafini, M. (2013). Functional foods and nutraceuticals as therapeutic tools for the treatment of diet-related diseases. *Canadian Journal of Physiology and Pharmacology*, 91: 387-396.

Maha A. and El-Motaleb el-Mowafy.(2012). Treatment Effect of Red Cabbage and Cysteine Against Paracetamol Induced Hepatotoxicity In Experimental Rats. *Journal of Applied Sciences Research,* 8(12): 5852-5859.

Malhotra, A., Kang, B. P. S., Cheung, S., Opawumi, D. and Meggs, L. G. (2001). Angiotensin II promotes glucose-induced activation of cardiac protein kinase C isozymes and phosphorylation of troponin I. *Diabetes*, 50(8): 1918–1926.

Manchali, S., Murthy, K. C. and Patil, B. S. (2012). Crucial facts about health benefits of popular cruciferous vegetables. *Journal of Functional Foods,* 4: 94-106.

Mard, S. A., Jalalvand, K., Jafarinejad, M., Balochi, H. and Naseri, M. K. G.(2010). Evaluation of the antidiabetic and antilipaemic activities of the hydroalcoholic extract of Phoenix dactylifera palm leaves and its fractions in alloxan-induced diabetic rats. *Malaysian Journal of Medical Sciences,* 17: 4-13.

Matheus, A.S.M., Tannus, L.R.M., Cobas, R.A., Palma, C.C.S., Negrato, C.A., and Gomes, M.B. (2013). Impact of Diabetes on Cardiovascular Disease: An Update. *International Journal of Hypertension,* http://dx.doi.org/10.1155/2013/653789.

McGibbon, A., Richardson, C., Hernandez, C. R.N. and Dornan, J. (2013). Pharmacotherapy in Type 1 Diabetes. *Canadian Journal of Diabetes,* 37: 56-60.

McLaughlin, T., Abbasi, F., Lamendola, C., Liang, L., Reaven, G., Schaaf, P. and Reaven, P. (2002). Differentiation between obesity and insulin resistance in the association with C-reactive protein. *Circulation*, 106(23): 2908-2912.

Miles, J. M. and Jensen, M. D. (2005). Counterpoint: visceral adiposity is not causally related to insulin resistance. *Diabetes Care*, 28(9): 2326-2328.

Mohammed, A., Adelaiye A.B., Abubakar, M.S., and Abdurahman, E.M. (2007). Effects of aqueous extract of Ganoderma Lucidum on blood glucose levels of normoglycemic and alloxan induced diabetic wistar rats. *Journal of Medicinal Plants Research*, 1(2):34-37.

Morse, S.A., Zhang, R., Thakur, V. and Reisin, E. (2005). Hypertension and the metabolic syndrome. *The American Journal of the Medical Sciences*, 330(6): 303-310.

Murali, B., Upadhyaya, U.M., and Goyal, R.K. (2002). Effect of Chronic treatment with Littorale in non-insulin dependent diabetic (NIDDM) rats. *Journal of Ethnopharmacology*, 81(2):199-204.

Mutlu F., Bener A., Eliyan A., Delghan H., Nofal E., Shalabi1 L., and Wadil N. (2014). Projection of Diabetes Burden through 2025 and Contributing Risk Factors of Changing Disease Prevalence: An Emerging Public Health Problem. *Journal Diabetes and Metabolism*. 5(2): 1- 7.

Nathan, D. M., Cleary, P. A., Backlund, J. Y., Genuth, S. M., Lachin, J. M., Orchard, T. J., Raskin, P. and Zinman, B. (2005). Intensive diabetes treatment and cardiovascular disease in patients with type 1 diabetes. *New England Journalof Medine,* 329(14): 977-86.

Nizamutdinova, I. T., Kim, Y. M., Chung, J. I., Shin, S. C., Jeong, Y. K., Seo, H. G., Lee, J. H., Chang, K. C. and Kim, H. J.(2009). Anthocyanins from black soybean seed coats preferentially inhibit TNF-α-mediated induction of VCAM-1 over ICAM-1 through the regulation of GATAs and IRF-1. *Journal of Agricultural and Food Chemistry*, 57: 7324-7330.

Nouwen, A., Nefs, G., Caramlau, I., Connock, M., Winkley, K., Lloyd, C. E., Peyrot, M. and Pouwer, F. (2011). Prevalence of depression in individuals with impaired glucose metabolism or undiagnosed diabetes: a systematic review and meta-analysis of the European Depression in Diabetes (EDID) Research Consortium. *Diabetes Care,* 34:752-762.

O'Brien, K. D., McDonald T. O., Chait, A., Allen, M.D. and Alpers, C. E. (1996). Neovascular expression of E-selectin, intercellular adhesion molecule-1, and vascular cell adhesion molecule-1 in human atherosclerosis and their relation to intimal leukocyte content. *Circulation,* 93: 672-682.

Oh, R.C., and Hustead, T.R. (2011).Causes and evaluation of mildly elevated liver transaminase levels. *American family physician,* 84 (9): 1003–8

Okokon, J. E., Umoh, E. E., Etim, E. I., and Jackson, C. L. (2009). Antiplasmodial and antidiabetic activities of ethanolic leaf extract of Heinsia crinata. *Journal of Medicinal Food,* 12: 131-136.

Okon, E. B., Chung, A. W., Rauniyar, P., Padilla, E., Tejerina, T., McManus, B. M., Luo, H. and van-Breemen, C. (2005). Compromised arterial function in human type 2 diabetic patients. *Diabetes,* 54: 2415-2423.

Olaitan, O.L. (2012). Patients' knowledge of causes, effects and complications of diabetes mellitus in Ilorin, Kwara state, Nigeria. *Journal of Biotechnology and Pharmaceutical Research,* 3 (6) 112-117.

Ozougwu, J. C., Obimba, K. C., Belonwu, C. D. and Unakalamba, C. B. (2013). The pathogenesis and pathophysiology of type1 and type 2 diabetes mellitus. *Journal of Physiology and Pathophysiology*, 4(4): 46-57.

Park, K. S., Ciaraldi, T. P., Abrams-Carter, L., Mudaliar, S., Nikoulina, S. E. and Henry, R. R. (1998). Troglitazone regulation of glucose metabolism in human skeletal muscle cultures from obese type 2 diabetic subjects. *Journal of Clinical Endocrinology and Metabolism,* 83: 1636-1643.

Patel, D.K., Prasad, S.K., Kumar, R., and Hemalatha, S. (2012). An over view on antidiabetic medicinal plants having insulin mimetic property. *Asian Pacific Journal of Tropical Biomedicine*, 2 (6): 320-330.

Patil, R., Patil, R., Ahirwar, B., and Ahirwar, D. (2011). Current status of Indian medicinal plants with antidiabetic potential: a review. *Asian Pacific Journal of Tropical Biomedicine*, 1(2): 291-298.

Pearson, E. R., Badman, M. K., Lockwood, C. R., Clark, P. M., Ellard, S., Bingham, C. and Hattersley, A. T. (2004). Contrasting diabetes phenotypes associated with hepatocyte nuclear factor-1a and -1B mutations. *Diabetes Care*, 27: 1102-1107.

Podsędek, A. (2007). Natural antioxidants and antioxidant capacity of Brassica vegetables: A review. *LWT- Food Science and Technology*, 40: 1–11.

Polak, M. and Cavé, H. (2007). Neonatal diabetes mellitus: a disease linked to multiple mechanisms. *Orphanet Journal of Rear Diseases*, 2: 2-12.

Priya, L. S. S. (2012). Cabbage – A Wonderful and Awesome Remedy for various ailments. *Research Gate: Pharmaceutical Sciences*, 1: 28-34.

Puddephat, I.J., Riggs, T.J., and Fenning, T.M. (1996). oleracea L.: a Transformation of Brassica critical review. *Molecular Breeding*, 2(3):185-210.

Rabasa-Lhoret, R. and Chiasson, J. L. (1998). Potential of alphaglucosidase inhibitors in elderly patients with diabetes mellitus and impaired glucose tolerance. *Drug Aging*, 13: 131-143.

Ravikumar, R., Krishnamoorthy, P. and Kalidoss, A. (2010). Antidiabetic and antioxidant efficacy of Andrographis paniculata in alloxanized albino rats. *International Journal of Pharmacy and Technology*, 2: 1016-1027.

Reitman, S., and Frankel, S. (1957). A colorimetric method for the determination of serum glutamic oxalacetic and glutamic pyruvic transaminases. *American Journal of Clinical Pathology*, 28(1):56-63.

Rheney, C.C., and Kirk, K. K. (2000). Performance of three blood glucose meters. *Annals of Pharmacotherapy*, 34 (3): 317-321.

Ridker, P. M., Buring, J. E., Cook, N. R. and Rifai, N. (2003). C-reactive protein, the metabolic syndrome, and risk of incident cardiovascular events: an 8-year follow-up of 14 719 initially healthy American women. *Circulation*, 107(3): 391–397.

Roman-Ramos, R., Flores-Saenz, J.L., and Alarcon-Aguilar, F.J. (1995). Antihyperglycemic effect of some edible plants. *Journal of Ethnopharmacology*, 48(1):25-32.

Ross, R. (1995). Cell biology of atherosclerosis. *Annual Review Physiology*, 57: 791-804.

Saleem, U., Khaleghi, M., Morgenthaler, N. G., Bergmann, A., Struck, J., Mosley, T. H. Jr. and Kullo, I. J. (2009). Plasma carboxy-terminal provasopressin (copeptin): a novel marker of insulin resistance and metabolic syndrome. *Journal of Clinical Endocrinology and Metabolism*, 94 (7): 2558–2564.

Samec, D., Bogovic, M., Vincek, D., Martincic, J., and Salopek-Sondi, B. (2013). Assessing the authenticity of the white cabbage (Brassica oleracea var. Capitata f. Alba) cv. 'Varaždinski' by molecular and photochemical markers. *Food Research International*. Available at:http://dx.doi.org/10.1016/jfoodres.2013.07.015. Accessed : 31 August 2013.

Sarikamiş, G., Balkaya, A. and Yanmaz, R. (2009). Glucosinolates within a collection of white head Cabbages (*Brassica Oleracea*) from Turkey. *African Journal of Biotechnology*, 8(19): 5046-5052.

Schernthaner, G., Wein, W., Shnawa, N., Bates, P. C. and Birkett, M. A. (2004). Preprandial vs. post-prandial insulin lispro: a comparative crossover trial in patients with type 1 diabetes. *Diabetic Medicine,* 21(3): 279-284.

Scicchitano, P., Cameli, M., Maiello, M., Modesti, A. P., Muiesan, M. L., Novo, S., Palmiero, P., Saba, P. S., Pedrinelli, R., Ciccone, M. M. and Gruppo di Studio Ipertensione, Prevenzione e Riabilitazione and Società Italiana di Cardiologia (2014). Nutraceuticals and dyslipidaemia: beyond the common therapeutics. *Journal of Functional Foods,* 6: 11-32.

Shirasaka, T., Takasaki, M. and Kannan, H. (2003). Cardiovascular effects of leptin and orexins. *The American Journal of Physiology-Regulatory Integrative and Comparative Physiology*, 284(3): 639-651.

Singh, A. K. J., Upadhyay, A., Bahadur, B., Singh, B., Singh, K. P. and Rai A. K. M. (2006). Anti – oxidant phytochemicals in Cabbage (Brassica oleracea L. Var. Capitata), *Scientia Horticulturae,* 108: 233-237.

Stein, E.A. (1987). Lipids, Lipoproteins and Apolipoproteins In: Treitz, N.W (Ed). Fundamentals of Clinical Chemistry, 3rd Edition. W.B. Saunders Philadelphia, pp 470 – 479.

Subramanian, V. (2011). Hepatoprotective activity of Brassica oleracea Italica against carbon tetrachloride induced in albino rats**.** *Journal of Pharmacy Research*, 4(4):1143.

Tabish, S. A. (2007). Is Diabetes Becoming the Biggest Epidemic of the Twenty-first Century? *International Journal of Health Sciences,* 1(2):5-8.

Takai, M., Suido, H., Tanaka, T., Kotani, M., Fujita, A., Takeuchi, A., Makino, T., Sumikawa, K., Origasa, H., Tsuji, K. and Nakashima, M. (2003). LDL – Cholesterol – Lowering Effect of Mixed Green Vegetable and Fruit Beverage containing Broccoli and Cabbage in hyper cholesterolemic subjects. *Rinsho Byori,* 51: 1073-1083.

Teimoori, M., Kouhsari, M. S., Ghafarzadegan, R. and Hajiaghaee, R. (2010). Antidiabetic effects of Juglans regia leave's methanolic extract on alloxan-induced male Wistar rats. *Journal of Medicinal Plants,* 9: 143-149.

Thirumalai, T., Therasa, S. V., Elumalai, E. K. and David, E. (2011). Hypoglycemic effect of Brassica juncea (seeds) on streptozotocin induced diabetic male albino rat. *Asian Pacific Journal of Tropical Biomedicine,* 1(6): 323-325.

Thorve, V.S., Kshirsagar, A.D., Vyawahare, N.S., Joshi, V.S., Ingale, K.G., and Mohite, R.J. (2011). Diabetes-induced erectile dysfunction: epidemiology, pathophysiology and management. *Journal Diabetes Complications,* 25(2):129-36.

Tietz, N.W. (1990). Clinical Guide to Laboratory Test, Second Edition W.B. Saunders Company, Philadelphia, U.S.A. 554-556.

Trayhurn, P. and Wood, I. S. (2004). Adipokines: inflammation and the pleiotropic role of white adipose tissue, *British Journal of Nutrition*, 92(3): 347-355.

Tsimikas, S., Willeit, J., Knoflach, M., Mayr, M., Egger, G., Notdurfter, M., Witztum, J. L., Wiedermann, C. J., Xu, Q. and Kiechl, S. (2009). Lipoproteinassociated phospholipase A2 activity, ferritin levels, metabolic syndrome, and 10-year cardiovascular and non-cardiovascular mortality: results from the Bruneck study. *European Heart Journal*, 30(1): 107-115.

Wacnic, R.G., and Alber, J.J.(1978). A comprehensive evaluation of the heparin manganese precipitation procedure for estimating high density lipoprotein cholesterol. *Journal of Lipid Research,* 19: 65-76.

Waqar, M. A., and Mohmood, Y. (2010). Antiplatelet, antihypercholesterolemic and, antioxidant effects of ethanolic extracts of Brassica oleracea in high fat diet provided rats.*World Applied Science Journal,* 8(1):107-112.

Wendt, T. M., Nozomu, T. N., Guo, J., Kislinger, T. R., Qu, W., Lu, Y., Bucciarelli, L. G., Rong, L. L., Moser B., Markowitz G.S., Stein G., Bierhaus A., Liliensiek B., Arnold B., Nawroth P.P., Stern D.M., D'Agati V.D., and Schmidt, A. N. (2013). RAGE Drives the Development of Glomerulosclerosis and Implicates Podocyte Activation in the Pathogenesis of Diabetic Nephropathy. *American Journal of Pathology,* 162(4): 1123-1137.

Wild, S., Roglic, G., Green, A., Sicree, R., and King, H. (2004). Global Prevalence of Diabetes Estimates for the year 2000 and projections for 2030. *Diabetic Care*. 27(5):1047-53.

Williams, D. J., Edwards, D., Hamernigb, I., Jian, L., James, A. P., Johnson, S. K. and Tapsell, L. C. (2013). Vegetables containing phytochemicals with potential anti-obesity properties: a review. *Food Research International*, 52(1): 323-333.

Winter, W. E. (2003). Newly defined genetic diabetes syndromes: maturity onset diabetes of the young. *Reviews in Endocrine and Metabolic Disorders,* 4: 43-51.

Xydakis, A. M., Case, C. C., Jones, P. H., Hoogeveen, R. C., Liu, M. Y., Smith, E. O., Nelson, K.W. and Ballantyne, C. M. (2004). Adiponectin, inflammation, and the expression of the

metabolic syndrome in obese individuals: the impact of rapid weight lose through caloric restriction. *Journal of Clinical Endocrinology and Metabolism*, 89(6): 2697-2703.

Yeh, C. T., and Yen, G. C. (2009). Chemopreventive functions of sulforaphane: a potent inducer of antioxidant enzymes and apoptosis. *Journal of Functional Foods,* 1: 23-32.

Zhang S., Liu C., Chuang C.L., Li X., Au M., Zhang M., Phillips A.R.J., Scott D.W., and Cooper G.J.S. (2014). The pathogenic mechanism of diabetes varies with the degree of over expression and oligomerization of human amylin in the pancreatic islet β cells.*The Federation of American Societies for Experimental Biology, Journal.* doi:10.1096/fj.14-251744.

APPENDIX 1

BLOOD GLUCOSE LEVEL

	Week 1	Week 2	Week 3	Week 4
Negative Control	410.00 ± 46.67	379.20 ± 6.67[a]	347.40 ± 11.96[a]	281.80 ± 3.65[a]
Positive Control	410.00 ± 22.98	262.00 ± 13.50[b]	162.20 ± 13.77[b]	93.20 ± 5.23[b]
10% Cabbage	406.80 ± 30.65	270.80 ± 11.48[b]	173.00 ± 10.38[b]	108.80 ± 9.75[c]
25% Cabbage	394.20 ± 30.59	270.60 ± 16.85[b]	149.80 ± 7.66[c]	95.20 ± 16.65[b]
50% Cabbage	407.80 ± 24.60	240.00 ± 13.11[c]	154.20 ± 6.90[c]	59.80 ± 5.88[d]

APPENDIX 11

LIPID PROFILE

	Total Cholesterol	Triglyceride	HDL	LDL
Negative Control	99.50 ± 10.15^{a}	115.42 ± 8.87^{a}	44.98 ± 5.85^{a}	63.48 ± 3.88^{a}
Positive Control	83.70 ± 8.25^{b}	163.40 ± 53.72^{b}	29.43 ± 5.17^{b}	28.23 ± 9.71^{b}
10% Cabbage	97.60 ± 14.47^{a}	36.87 ± 4.89^{c}	39.59 ± 1.60^{c}	52.64 ± 5.50^{c}
25% Cabbage	71.50 ± 9.95^{c}	75.00 ± 15.30^{d}	25.82 ± 3.00^{d}	45.68 ± 3.31^{c}
50% Cabbage	59.00 ± 13.70^{d}	32.22 ± 2.97^{c}	65.12 ± 8.99^{d}	44.93 ± 11.56^{c}

APPENDIX III

Liver Enzymes

	AST	ALT	ALP
Negative Control	13.62 ± 2.40^{a}	7.60 ± 1.12^{a}	28.88 ± 9.14^{a}
Positive Control	12.07 ± 1.72^{a}	6.13 ± 0.88^{a}	60.47 ± 6.02^{b}
10% Cabbage	25.22 ± 3.08^{b}	15.56 ± 2.82^{b}	12.24 ± 0.71^{c}
25% Cabbage	44.95 ± 5.46^{c}	35.35 ± 2.38^{c}	29.26 ± 5.95^{a}
50% Cabbage	36.68 ± 4.82^{d}	28.60 ± 1.83^{c}	15.41 ± 0.83^{c}