COMPARATIVE EFFECTS OF CARBONATED DRINK (BIGI TROPICAL) AND HEALTH DRINK (EVIRON) ON SOME BIOCHEMICAL INDICES IN HUMANS

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CHAPTER ONE

INTRODUCTION

1.1 Background of the study

Throughout the long-term soft drinks have been taken to extinguish thirst, top off human's sugar hunger, etc., they incorporate carbonated beverages, juices and caffeinated drinks (British Soft Drinks Association Annual Report, 2016). Soda pops are any class of non-mixed refreshment typically yet not really carbonated, regularly containing a characteristic or counterfeit improving specialist, palatable acids, normal or fake flavors and some of the time juice. Common flavors are gotten from natural products, nuts, berries, roots, spices and other plant sources (Pietka, 2019).

Business soda drinks initially showed up in 1884 when an item called "Moxie" was made by a drugstore proprietor in Lisbon (Tahmassebi et al, 2006). Before long a short time later, comparative items showed up including Coca-Cola and Pepsi. Over the previous century, soda pops have changed drastically from being a neighborhood drug store item to the overall business that procures $60 billion and produces 1 billion liters for every year. These progressions have been because of advances in assembling innovation and showcasing developments (Shenkin et al. 2003).

Healthy living is prime concern of today’s society, many studies show that people are adopting health related changes in their eating habits and these changes create new opportunities and better varieties in the society, and slowly health drink industry is emerging as the most popular and developing portion of the general soda pops industry on the grounds that the impacts of these beverages on the human wellbeing is of a higher rate (Dave et al, 2016).

Some carbonated drinks have been proposed to harmfully affect the dental and general wellbeing of individuals including kids and young people (Scott et al, 2019). The high content of sugar and acids, which have cancer-causing and acidogenic potential, can add to dental caries, tooth disintegration, as well as adding to health effects such as overweight and obesity and may be associated with an increased risk of type 2 diabetes.

Health drinks then again, are drinks which claim to be beneficial to the health having medicinal properties. The acclaimed benefits of health drinks include anti-oxidants properties, blood pressure control, anti-cancer properties, anti-inflammatory properties, digestion, hydration, bone health, weight loss among others (Dave et al, 2016).

Carbonated drinks cause weight increase in several ways, one of the ways put forward by nutritionists is the consumption of high fructose syrup extracted from corn because it deprives the body of its ability to control appetite. At the moment, in Nigeria 64.5% of adults above 20 are overweight, 30.5% are obese and 4.7% are severely obese (Scott et al, 2019). This could be the main reason behind the rise of epidemiological research on children and adults which have identified that carbonated drinks have strong link with over-weight and type 2 diabetes (Scott et al, 2019).

Late investigations have brought up that fructose utilized in most sodas are hazardous. Fructose is found in numerous plants; it is a segment in sucrose (table sugar) and high fructose corn syrup (HFCS) produced using corn. Fructose is processed in the liver and stances health risk to humans since sugar forms the base for calories. Due to lack of physical activities this sugar accumulates in the human system as fats and this accumulation of fats leads to development of chronic diseases like type 2 diabetes or obesity (Scott et al, 2019).

Carbonated drinks have gotten one of the best social issues in this day and age. Their ready accessibility makes them a quicker answer for thirst and urge of refreshment. For instance, in our country we have over 100 different types of carbonated drinks and the distribution of these drinks is made easy due to hawkers found on the streets, increase of shop owners etc.

Endeavors have been made by manufacturers and government organizations to lessen the possible hurtful impacts of sugar-containing soda drinks on teeth and general health. These include prohibiting the offer of soda drinks for schools, confining sodas publicizing, adjusting the synthesis of sodas and presenting charge on sugar-containing soda drinks.

A key inquiry is whether activities taken to diminish soda utilization are justified given the accessible science and whether diminishing populace utilization of sodas would profit general wellbeing. The present study therefore compares the effects of a Nigerian brand of carbonated drink (Bigi tropical) and health drink (Eviron) on blood glucose and lipid profile of human volunteers to further elucidate the previous claims.

1.2 Statement of the problem

The relationship between carbonated drink consumption, nutrition and health results was examined over time and a strong correlation of carbonated drink intake with increased energy intake and body weight, with lower milk, calcium and other nutrients intake and increased risk of multiple medical problems such as diabetes (Vartanian, 2007).

Carbonated drinks are beverages containing caloric sweeteners, and are also classified as drinks loaded with sugar. Such beverages are the largest single source of added sugar in a human diet and play a major role in driving current trends in obesity, tooth ache etc. People of different ages consume one or more sodas (carbonated drinks) every day. Studies show

that the risk of childhood obesity rises with every extra daily supply of carbonated drinks, and this is a serious concern for the society in general. (childhood obesity initiative, 2018).

The utilization of carbonated drinks has become an exceptionally obvious and hostile general wellbeing and public strategy issue. Such beverages were viewed as a significant supporter of obesity and related health problems and were subsequently focused as methods for assisting with controlling the developing pervasiveness of corpulence, particularly among youngsters.

The issue is not new. In 1942 the American Medical Association referenced sodas explicitly in a solid suggestion to confine the utilization of added sugar. At that time, annual US production of carbonated soft drinks was 90 8-oz (240-mL) servings per person; by 2000 this number had risen to more than 600 servings. Over the years, debate has emerged over several fundamental concerns: whether these drinks lead to over-consumption of energy; whether they displace certain foods and beverages and therefore nutrients; whether they contribute to diseases such as obesity and diabetes (Vartanian, 2007). Therefore, it is expedient to find answers to these questions through scientific investigations.

1.3 Aim of the study

The aim of this research is to investigate the effect of carbonated drink and health drink on blood glucose levels and lipid profile of humans, as indicators of risks of diabetes mellitus and cardiovascular diseases.

1.4 Objectives of the study

This study aims at investigating the effects of carbonated (Bigi Tropical) and health (Eviron) drinks on the health of human volunteers by achieving the specific objectives which are to:

I. determine the effect of carbonated and health drinks on the blood glucose levels of human volunteers.

II. determine the effects of carbonated and health drinks on the blood lipid profile (total cholesterol, triglycerides, HDL-cholesterol, VLDL and LDL- cholesterol) of the human volunteers.

III. estimate the effect of carbonated and health drinks on the atherogenic indices (atherogenic index of plasma (AIP) and atherogenic coefficient (AC) of the human volunteers.

1.5 Justification of the study

There are so many carbonated drinks in the country today; almost yearly new drinks are being introduced into the Nigeria market. Most of the citizens who take these drinks do not take note of the contents of the drinks. The variety of drinks available in the Nigerian market range from carbonated, hot/alcoholic, and health drinks to modified/fortified water.

The carbonated drinks have been reported to increase blood glucose level, lipid level, and the risk of cardiovascular diseases among others. While the health drinks are reported to improve human health and decreases the risk of the above listed diseases (Sethi, 2004**).** However, if not informed properly, the health drink could be taken wrongly and excessively leading to its toxicity. Therefore, it is essential to investigate the effect of carbonated and health drink (Eviron) on human health.

It is important consumers enlighten themselves on the right choice of drink. This study will enlighten the consumers on the effects of each tested drink on blood glucose level and lipid profile of human volunteers, as markers of diabetes mellitus and cardiovascular diseases. This will inform consumers' choices of advisable or better drinks. Also, some of the previously reported health effects of carbonated drinks may be confirmed.

CHAPTER TWO
LITERATURE REVIEW

2.1 Overview

The body is the product of food; diseases occur as the result of a faulty diet. The distinction between health and disease arise as a result of difference between the wholesome and unwholesome diet. Thus, the body is the outcome of the nutrition ingested in four-fold manner, eating, drinking, licking, and mastication (Sethi, 2004**).** The World Health Organization (WHO) states that unhealthy diets high in sugar, salt and fat contribute to the global epidemic of obesity and place people at higher risk of diabetes, cardiovascular disease and some cancers. These diseases, collectively called non-communicable diseases (NCDs), are the leading cause of death worldwide according to a major worldwide study on the Global Burden of Diseases (Lancet, 2012).

There are so many carbonated drinks out in the country today; almost yearly new drinks are being introduced into our society. Citizens who take these drinks do not take note of the contents of the drinks. There are so many death and even damaged health cases caused by these carbonated drinks (Gibson, 2008). Researchers have shown that high intake of sugar-sweetened soft drinks like Pepsi, Fanta can lead to increased risk of several diseases, including cardiovascular disease, obesity and type-2 diabetes (Gibson, 2008). Diabetes mellitus is a chronic disease characterized by persistent hyperglycemia and glycosuria and is the most prominent disease related to failure of blood sugar regulation. The disease was found to be uncommon in the African continent in the 1960, but in 1992 the Nigerian National Expert

Committee on non-communicable diseases discovered a high prevalence of the diabetes in the urban areas of Nigeria (Santaguida, 2005).

An article from The Guardian Newspaper published in 2019 on drinks titled “SOFT DRINKS ARE POISON” reported that several labeled soft drinks are harmful to the human health and the dangers of these drinks are known to most people, but this awareness has not stopped people’s addiction or the production of these drinks by their manufacturers. People understand the dangers of these drinks yet fail to apply this understanding to reducing their consumption. Taking soft drinks is a simple propensity to fall into and it is anything but difficult to consider diet adaptations as substitutions. A taste for the wellbeing of nostalgia or a speedy caffeine hit can prompt a three-a-day propensity. Sodas are not only terrible for the human wellbeing; it ought not be taken by any stretch of the imagination.

An assortment of beverages is accessible in the Nigerian market, and they shift from carbonated, hot, alcoholic, wellbeing, water and so forth. Subsequently, the need to survey the impacts of these beverages on blood glucose level and lipid profile which are a portion of the few biochemical markers that can give significant data on diabetes mellitus, cardiovascular illness and liver function.

A person’s beverage choice contributes significantly to dietary and calorie intake, so just the mere intake of carbonated drinks cause harm to the health, so does health drinks which improve the human system, making it of a higher advantage. Humans maintain their water balance by consuming an equal amount of water to that which is excreted and this is gotten from health drinks. “Healthcare Can Lead the Way”, gives extra data about the significant function of medical clinics and other medical services settings in advancing sound food and drink programs

as a component of accepted practice change important to battle obesity epidemic (Public health law center, 2013).

A survey brief by the Society for Human Resource Management in 2008 reported that intake of healthy drinks improves the entire capability of a person. Therefore, a healthy beverage program that includes environmentally responsible practices not only can promote better physical health for humans, but also can help boost morale (Public health law center, 2013).

Consumption of sugary beverages is a key contributor to many obesity-related health issues. This study aims at providing information on the effect of selected brand of carbonated and health drink in view of enlightenment towards the choice of beverages that support efforts to achieve and maintain a healthy living. The reduction or elimination of sugar-sweetened beverage consumption has great potential to help humans reduce caloric intake, improve diet quality, and reduce their risk for obesity, hyperglycemia and hyperlipidemia. Implementation of the recommendation for healthier beverages across a variety of places and environments, such as schools, workplaces, the gym and hospitals will support these efforts and help improve the human health (Robert, 2013).

2.2 Carbonated drinks

Carbonated drinks are beverages with added carbon dioxide (packed in hermetically sealed containers to ensure no freedom of spoilage), that gives an effervescent taste to the beverage. They are divided into cola, diet and regular soft drinks (Abdellatif, 2018). The cola flavored drinks usually contain added phosphoric acid as acidulant. Acidulants are substance exacerbates that present a tart, harsh or acidic flavor to food, non-cola drinks utilize citrus extract as acidulant on the grounds that it can reinforce the causticity (Abdellatif, 2018)). These drinks contain high

levels of sugar which is used as their sweetener and they offer little nutritional value and more detrimental effects on the health such as weight gain, obesity and dental erosion. They are water based flavored drinks prepared with water and one or more of the following ingredients: fruit juice, fruit pulp, artificial flavoring materials, permitted colorings, sweetening agents, acidulants, clouding matter and preservatives, carbon dioxide and caffeine (Abdellatif, 2018). Examples include; pepsi, fanta, bigi, sprite and coke etc.

2.3 Health drinks

Health drinks are drinks which claim to be beneficial to the health having medicinal properties along with quenching of thirst. They supply fluid and nutrients to the body (Sethi, 2004**)**. It contains little or no sugar and fat, lower in calories and contribute to weight loss and good health. There are several classes of health drinks which contain different bioactive compounds that are of benefit to the health. Examples include yogurt, trevor, smoothies, zobo and aloe vera drinks etc.

2.4 Health implication of carbonated drinks

Carbonated drinks are not the preferred or best option when it comes to taking of drinks because they have few health benefits and a lot of health disadvantages.

2.4.1 Health benefits of carbonated drinks

1. Caffeine benefits: caffeine is a stimulant that is found in some soft drinks. Its large amount can have detrimental effects on your health but it also has numerous benefits when taken in little quantities. Caffeine stimulates the central nervous system, helps breakdown fatty acids in the liver, boosts mood and alleviates headaches. According to

scientific reports, people who regularly ingest caffeine are less likely to develop Parkinson's disease (disorder of the central nervous system), colon cancer, gallstones and cirrhosis of the liver (Satish, 2018).

2. Carbonated water: this is a primary ingredient of soft drinks; created by Joseph Priestly in the year 1767 and has since proven to have many benefits for the gastrointestinal tract. Carbonated water eases stomach ache, quells nausea and has been proven to alleviate constipation (Satish, 2018).

2.4.2 Health risks of carbonated drinks

1. High sugar levels: high amounts of fructose corn syrup are added to sweeten soda (equivalent of over 10 teaspoons of sugar or about 140 calories in a 12 ounce can of soda). These drinks contain hidden calories because people do not reduce their food intake to compensate for the extra calories consumed in sugary drinks, thereby increasing their overall calorie intake (Watson, 2013).
2. High caffeine levels: energy drinks contain high amounts of caffeine that is dangerous when consumed in excess. Caffeine is mildly addictive and taking a can or two can affect performance and mood, raise anxiety and cause insomnia. Drinking excessive amounts of caffeine leads to caffeine intoxication with symptoms of nervousness, anxiety, restlessness, insomnia, gastrointestinal problems, tremors, rapid heartbeats and death (Mohinder, 2013).
3. Dental problems: different acids are added to many sodas. Phosphoric acid gives beverages a characteristic tangy or sour flavor to balance the sweetness and to prevent the growth of micro-organisms. Other acids often added include citric acid from oranges, tartaric acid from grapes, malic acid from apples as well as ascorbic and carbonic acids.

Too much intake of all these acids results in the softening of tooth enamel resulting in dental caries, which mostly occurs in children and adolescents. The sugar in these drinks also cause tooth decay (Mohinder, 2013).

4. Extra fat and obesity: fat such as cream and whipping cream, can often be found in coffee, and cream-based drinks. These are not heart friendly and they add calories to the body. Each additional daily serving of soda has been reported to increase human's risk for obesity (Satish, 2018).
5. Heart diseases and asthma: 70% of cardiovascular disease is related to obesity which is mostly caused by the intake of excess sugar into the human system. Heart disease is top killer and should be avoided as much as possible, several cases of stoke occur regularly. Sodium benzoate is a preservative used in the preservation of most soft drinks and it triggers asthma (Satish, 2018).
6. Kidney problems and osteoporosis: most soft drinks contain high levels of phosphoric acid, which has been linked greatly to kidney stones and other kidney problems, also high phosphate diet causes bone breakdown and an increased risk in osteoporosis (Satish, 2018).

2.5 Erosive potential on teeth

Soft drink intake, even of relatively short duration, has been found to reduce enamel microhardness. Carbonated drinks are associated with erosion due to its low pH and titratable acid. The total acid level, acid type, concentration of phosphate, calcium and fluoride in food drinks have a modifying effect on the development of dental erosion. Temperature and exposure time had also been discussed as important to the erosivity of beverages. (Cornelius, 2007)

It has been suggested that the total acid level (titratable acid) be considered more important than pH level in evaluating the erosive potential of acidic drinks, because it will determine the actual H^+ available to interact with the tooth surface (Cornelius, 2007). pH and titratable acidity of the erosive challenge were said to determine the degree of saturation with respect to the tooth mineral and thus the driving force for its dissolution. Most soft drinks contain one to two common acidulants (phosphoric, citric, malic, tartaric acid). Acid is used in soft drink products to accomplish two main functions. Firstly, acidity is a key factor in the taste of a drink as it balances the sweetness, People generally prefer more acidic foods and drinks. Secondly, it inhibits the growth of micro-organisms such as yeasts, molds and bacteria (Eyitope, 2007).

Phosphoric acid is very erosive at pH 2.5 but much less so at pH 3.3. Citric, malic and tartaric acids are considered to be especially erosive because of their acidic nature and the ability to chelate calcium at higher ph. Citric acid is more erosive than malic acid when formulated to experimental drinks at high pH (Bamise et al., 2007)

2.6 Drink intake and incidence of Obesity and Diabetes mellitus

All age groups in the Nigerian population are gaining weight as they age in the current obesogenic environment; however, younger generations are gaining weight faster than before. Although there are a variety of factors affecting weight gain, the WHO has recognized the particular contribution of a subset of factors, including soft drinks, to the increased risk of obesity. Soft drink consumption is associated with a higher total intake of calories, weight gain and a lower overall diet (Hattersley, 2009).

Among other disorders, the consumption of sugar sweetened beverages has been consistently linked to increased risk of obesity, type 2 diabetes, osteoporosis and cardiovascular disease.

Carbon containing beverages have risen as a percentage of the Westernized diet over the past 20 years and have substantially led to a rise in liquid energy consumption, these drinks are also known as sugar sweetened drinks (SSB) and they are consumed by both adults and children (Martin, 2017).

Hyperglycemia is referred to as high blood glucose level in the body. The blood sugar level or blood glucose level is the concentration of glucose present in the blood of humans, they are less than 100 mg/dL after fasting for eight hours and less than 140 mg/dL two hours after eating. The normal blood glucose level (tested while fasting) for a non-diabetic person is between 70 to 80 mg/dL. Some 60 mg/dL is the normal level while some it is 90 mg/dL (Michigan Diabetes Research and Training Center, NIH, 2012)

Due to excess sugar found in carbonated drinks it can lead to hyperglycemia which is known as high blood glucose. The sugar is broken down in the body to give glucose. Glucose is a simple sugar and the key metabolic substrate for tissue energy production Hyperglycemia is an indication of diabetes mellitus, meaning diabetes is characterized by hyperglycemia. Hyperglycemia can lead to long term complications such as damage to eyes, kidneys, or nerves (Shouip, 2014).

The overconsumption of soft drinks is often seen as a significant public health issue with consequences for cardiovascular diseases (Kregiel, 2014). This follows a number of studies carried out in both animals and humans suggesting that chronic consumption of refined sugars, especially fructose, may contribute to cardiovascular dysregulation and the onset of hypertension (Martin, 2017).

In recent survey it has been indicated that young adults are the nation's highest consumers of calorie carbonated drinks with a daily intake of about two to three bottles, which has been proven to be unhealthy (Martin, 2017).

2.7 Increased consumption of carbonated drinks among youths

Consumption of carbonated drinks has become a highly visible and controversial public health and policy issue. Many view carbonated drinks as a major contributor to obesity and related health problems, and have therefore been targeted as a means of helping to curtail the increasing prevalence of obesity, especially among youths (Lenny et al, 2007).

The leading source of added sugars in western diets is Sugar-sweetened beverages (SSBs). The major contributors of sugar from all SSBs are carbonated soft drinks, such as Coke, Pepsi, Sprite and Fanta. Since world war II, there has been a massive change in dietary habits from conventional to western diets in low- and middle-income countries (LMICs) (Yang et al, 2017).

There is an obvious evidence that intake of carbonated drinks, which are energy-dense and nutrient-poor, is associated with an increased risk of obesity, dental caries, early puberty and violent activity among youths, as well as obesity, diabetes and other chronic diseases (Verzeletti et al, 2010).

The level of carbonated drinks consumption was evaluated with the question "Over the past 30 days, how many times a day did you take carbonated drinks, such as Coca Cola and Fanta etc.? response choices were, I did not drink carbonated drinks in the past 30 days, "less than one time daily." "1 time daily," "2 times daily," "5 and more times daily". The mean carbonated drink intake was measured in different countries and none was coded as 0 (Yang et al, 2017).

2.8 Hyperglycemia and increased lipid profile

Diabetes is characterized by chronic hyperglycemia and disturbances of carbohydrate, lipid and protein metabolism. Hyperlipidemia is a disorder characterized by the elevation of any or more lipid or lipoprotein levels in the blood. The lipid profile is elevated due to overproduction of acetyl coA, which is caused by excess glucose in the body due to over consumption of carbonated beverages. High low-density lipoprotein cholesterol (LDL) is an indicator of atherosclerosis risk and it is caused as a result of poor diet, obesity, diabetes etc. HDL is considered good because it carries cholesterol from the tissues to the liver (Onwe, 2015).

The lipid metabolism is closely related to the carbohydrate metabolism that can be transformed to fats, and the metabolism of both is disrupted by diabetes mellitus. Accumulation of fatty liver and hepatic triglycerides is commonly associated with obesity, insulin resistance and type 2 diabetes, and is subject to dietary factors such as carbonated drink consumption. Increasing the plasma levels of glucose, insulin, and lipids is associated with impairing the endothelial function during the early stage of type 2 diabetes development (Mohammadshahi, 2014). Insufficient insulin production in humans poses the risk of diabetes mellitus, and the insulin role is appropriately regulated by a short dose. Lack of insulin contributes to elevated glucose levels in the bloodstream (Sabahelkhier, 2016).

Excess glucose in the body resulting from excess sugar consumption from carbonated beverages, contributes to the overproduction of acetyl-coA, a precursor of most lipid molecules, such as cholesterol (Mohammadshahi, 2014). When there is excess glucose in the body which is as a result of excess intake of sugar gotten from carbonated drinks, it leads to the over production of acetyl-coA which is a precursor of most lipid molecules such as cholesterol (Mohammadshahi, 2014).

A study titled lipid profile abnormalities seen in type 11 diabetes patients in primary healthcare in turkey: a cross sectional study, concluded that the study showed widespread lipid abnormalities that caused dyslipidemia in the course of diabetes such as hypercholesterolemia, hypertriglyceridemia, elevated LDL and decreased HDL. This research proposes that hyperlipidemia predominate over increased diabetic dyslipidemia prevalence (Ozder, 2014).

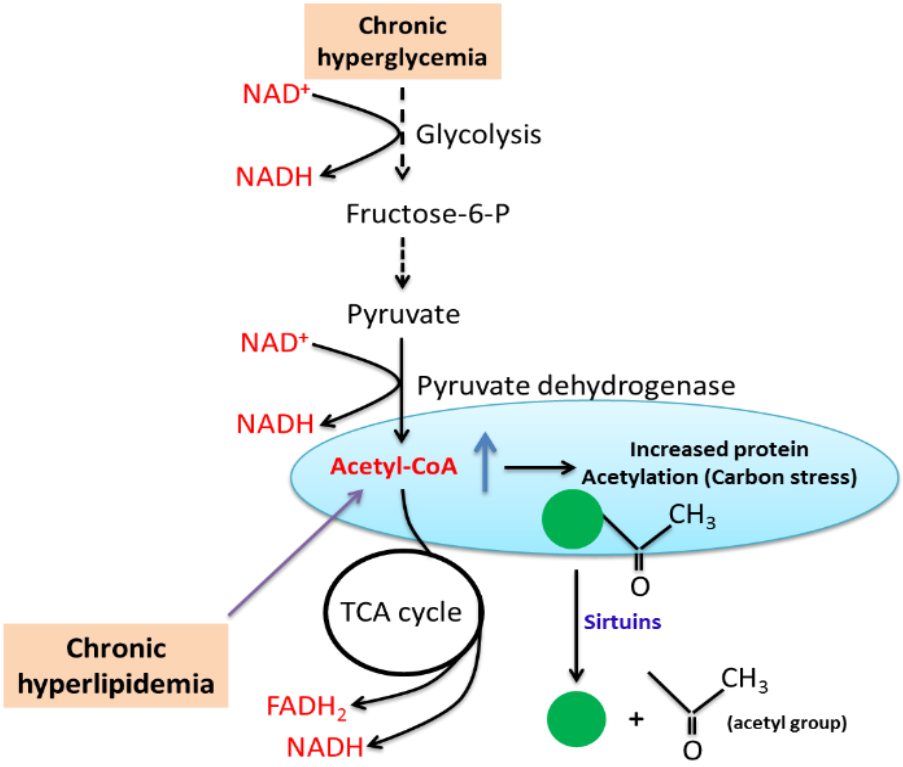

Figure 2.1: Link between hyperglycemia and hyperlipidemia (Xiaoting, (2016). Aging and disease)

2.8.1 Types of hyperlipidemia

This medical condition is sub-divided into two:

a) Primary hyperlipidemia

This usually take place as a result of genetic problems i.e., mutation within receptor protein, which may be due to single (monogenic) gene defect or multiple (polygenic) gene defect. This type may occur as a result of change in dietary and lack of proper physical activities. The table below summarizes the various classes of primary hyperlipidemia.

TYPE	DISORDER	CAUSE	OCCCURENCE	ELEVATED PLASMA LIPOPROTEIN
1	Familial lipoprotein lipase deficiency	Genetic	Very rare	Chylomicrons
2	Familial hypercholesterolemia	Genetic	Less common	LDL
3	Polygenic hypercholesterolemia	Multifactorial	Commonest	LDL
4	Familial dysbetalipoproteinemia	Genetic	Rare	IDL, Chylomicrons Remnants

5	hypertriglyceridemia	Multifactorial genetic	Common	VLDL
6	Familial combined hyperlipidemia	Genetic	Less common	VLDL, LDL

Table 2.1: Primary hyperlipidemia (Folawiyo, 2015)

b) Secondary hyperlipidemia

This arises as a result of other underlining diseases like diabetes, myxedema, nephritic syndrome, chronic alcoholism, with use of drugs like corticosteroids, oral contraceptives (Folawiyo, 2015).

Table 2.2: Secondary hyperlipidemia (Folawiyo, 2015)

Total level of cholesterol	Category
Less than 190 mg/dL	Desirable
200-240 mg/dL	Borderline
Above 240 mg/dL	High
LDL (bad) level of cholesterol	LDL category
Less than 98 mg/dL	Optimal
98-130 mg/dL	Above optimal
131-159 mg/dL	Borderline
160/190 mg/dL	High
Above 190 mg/dL	Very high

HDL (good) level of cholesterol	HDL category
Less than 39 mg/dL	Major risk for heart
39-59 mg/dL	Better
Above 60 mg/dL	Protective against heart disease

2.8.2 Causes of hyperlipidemia

The main cause of hyperlipidemia includes changes in lifestyle habits in which risk factor is mainly poor diet i.e. with a fat intake greater than 40 percent of total calories, saturated fat intake greater than 10 percent of total calories; and cholesterol intake greater than 300 milligrams per day or treatable medical conditions (Onwe, 2015). The abnormal cholesterol levels are the result of an unhealthy lifestyle including taking high-fat diet (such as dairy products, ice cream pastries, fried and junk foods, meat, soft drinks). and other lifestyle factors like obesity, genetic or inheritance, smoking heavy, diabetes, kidney disease, pregnancy, underactive thyroid gland (Kelly, 2010), several drugs such as corticosteroids, diuretics, beta- blockers and medicines used to treat depression may raise cholesterol levels, alcohol, steroids, hypothyroidism, and Lack of exercise (Onwe, 2015).

The higher levels of female hormones like estrogen, have been noted to increase or change cholesterol levels. Another modifying factors in the development and progression of hyperlipidemia are age and gender, it has been shown that cholesterol levels increase as a person gets older (Onwe, 2015). Heredity has also been a modifying factor for the progression of hyperlipidemia as it has been noted that the genes partly determine the amount of cholesterol body makes (Onwe, 2015).

2.8.3 Treatment of hyperlipidemia

The National Cholesterol Education Program Adult Treatment Panel III (NCEP ATP III) recommends that a fasting lipoprotein profile and risk factor assessment be used in the initial classification of adults. There are three categories of risk that modify the goals and modalities of LDL-lowering therapy. The highest risk category is having known CHD (coronary heart disease) or CHD risk equivalents; the risk for major coronary events is equal to or greater than that for established CHD (i.e., more than 20% per 10 years, or 2% per year). The intermediate category includes two or more risk factors, in which the 10-year risk for CHD is 20% or less. The lowest risk category is persons with zero to one risk factor, which is usually associated with a 10-year risk of CHD of less than 10% (Okorocha, 2015). Basic treatments include:

- Proper diet.
- Less weight of the body.
- Regular exercise.
- Eating non-oily food.
- Eating fruits such as pears, apples, bananas etc.
- Eating proteins such as fish, milk etc.
- Fibrates (fenofibrate) may lower triglyceride levels.
- Taking medications.

2.9 Cardiovascular diseases (CVD)

Cardiovascular diseases are conditions that affect the structure or function of the heart. Hyperlipidemia increases synthesis of cholesterol which can cause atherosclerosis, and it is the starting point of coronary heart disease and other cardiovascular diseases (NHS, 2018).

Cardiovascular diseases are caused by blockage in the blood vessels preventing blood from flowing to the heart and brain, this blockage is caused by fatty deposits (caused by excess lipid profile in the human system) in the inner walls of the blood vessels (NHS, 2018).

It is a major cause of several diseases and premature death throughout the world, and it contributes greatly to the escalating costs of health care. The underlying pathology is atherosclerosis (a disease in which plaque builds up inside the arteries) which develops over many years and is usually advanced by the time symptoms occur. Acute coronary and cerebrovascular diseases frequently occur suddenly and are often fatal before medical care can be given (NHS, 2018).

Cardiovascular diseases include:

- coronary heart disease – disease of the blood vessels supplying the heart muscle;
- cerebrovascular disease – disease of the blood vessels supplying the brain;
- peripheral arterial disease – disease of blood vessels supplying the arms and legs;
- rheumatic heart disease – damage to the heart muscle and heart valves from rheumatic fever, caused by streptococcal bacteria;
- congenital heart disease – malformations of heart structure existing at birth;
- deep vein thrombosis and pulmonary embolism – blood clots in the leg veins, which can dislodge and move to the heart and lungs. (American Heart Association, 2019)

Heart attacks and strokes are usually acute events and are mainly caused by a blockage that prevents blood from flowing to the heart or brain. The most common reason for this is a build-up of fatty deposits on the inner walls of the blood vessels that supply the heart or brain. Strokes can also be caused by bleeding from a blood vessel in the brain or from blood clots (WHO, 2007).

2.10 Association of carbonated drink preservatives (sodium benzoate) and asthma

Studies have shown that carbonated drink consumption is linked with increased asthma risk. There are two possible mechanisms for the association between soft drinks and asthma: carbonated drinks contain high amounts of sugar which could promote inflammation, also sodium benzoate is a preservative used in carbonated drinks and it has been known to worsen asthma symptoms (Berentzen et al.,2014). Sodium benzoate is bacteriostatic and fungistatic under acidic conditions. Preservatives, dyes and chemicals have been known to cause asthma in sensitive persons. (Berentzen et al.,2014).

Sodium benzoate and potassium sorbate have long been used for large scale beverage preservation. Studies have shown that benzoate can cause drinks to contain traces of carcinogen benzene, this benzene is thought to have its origins in a free radical catalyzed reaction of the benzoate with ascorbic acid (Berentzen et al., 2014). In children, high dietary intake of sodium benzoate is associated with asthma, allergy, or attention deficit-hyperactivity disorder (Berentzen et al., 2014).

In the body, benzoate readily undergoes conjugation with glycine in the liver and kidney, this conversion to Hippurate increases its water solubility in order for it to be efficiently removed from the body by the kidneys. Dietary ascorbic acid can be metabolized by the same oxidation pathway as short chain fatty acids (Piper, 2018).

Reactions to these preservatives in the body starts about 10 - 20 minutes after ingestion. Symptoms can range from coughing, urticarial rash, gastro- intestinal symptoms, and wheezing to life-threatening anaphylactic reactions. Higher concentrations of sodium benzoate resulted in more reactions. Mostly patients who react to these preservatives have not been shown to have IgE (immunoglobin E) antibodies against sulphites. IgE are antibodies produced by the immune

system. The immune system of a person with allergies overreacts to the allergy by producing antibodies called IgE (Stokkons et al.,2014).

2.11 Physiological effects of drink intake on the liver function enzymes; Aspartate aminotransferase (AST) and Alanine aminotransferase (ALT)

The liver is a large, complex organ, located in the upper right-hand side of the abdominal area just beneath the rib cage, well designed for its central role in the metabolism of carbohydrates, proteins, and fats. It is the site where metabolism waste products are detoxified by processes such as amino acid deamination which produces urea (Giannini, 2005).

The liver helps absorb the nutrients of the body, produces bile to help digest fats, produces many essential proteins, such as blood clotting factors and breaks down potentially toxic substances into harmless ones that can be used or excreted by the body (AACC, 2020).

The liver is the largest solid organ, the largest gland and one of the most vital organs which functions as a center for nutrient metabolism and waste metabolite excretion. Its main purpose is to monitor the flow and protection of substances ingested from the digestive system before they are spread to the systemic circulatory system (Ozougwu, 2017).

Aspartate aminotransferase (AST) and Alanine aminotransferase (ALT) are enzymes present in the cells of the body. They are mainly found in the heart, and liver, but in a lesser quantity in the kidneys and muscles. The levels of AST and ALT are low in healthy individuals. When liver or muscle cells are damaged, AST and ALT are released into the blood, making them a useful test for liver damage diagnosis or monitoring (AACC, 2020). ALT is predominantly present in the liver which helps the liver cells to convert proteins into energy. ALT is released into the bloodstream when the liver is damaged and the levels increase (Muneeza, 2014).

Aspartate aminotransferase (AST) and alanine aminotransferase (ALT) catalyzes the transfer of an amino group from an amino acid (L-alanine and L-aspartate) to α-ketoglutarate. The reaction products are L-glutamate, pyruvate or oxaloacetate (Mitchell, 2016).

An article from The Daily Telegraph published in 2009 on drinks titled "TWO SOFT DRINKS A DAY MAY LEAD TO LONG TERM LIVER DAMAGE" reported that alcohol abuse is normally associated with liver damage but the new study found that non-alcoholic drinks with a high sugar content can cause a condition called liver fatty disease, Israel scientists found that people who consume one liter of carbonated drinks and fresh fruit juices on a regular basis were five times more likely to develop liver disease (Ali et al, 2009).

This clinical study examined 60 non-alcoholic fatty liver disease (NAFLD) patients, comparing their soft-drinking patterns, food consumption and blood markers of inflammation and insulin resistance to 18 controls without liver disease. The study found that soft drink intake rates in those with NAFLD were significantly higher compared with those without. Obesity, high blood sugar, high blood pressure and elevated cholesterol are all symptoms of metabolic syndrome, a NAFLD-associated disorder. It seems likely, therefore, that anyone who consumes more sugar drinks can have other health habits and risk factors that lead to the risk of NAFLD (Ali et al, 2009).

2.12 Beneficial effects of health drinks

Health drinks have several benefits which are listed below.

1. **fruit based drinks – Watermelon smoothie** (Reetu, 2018)

   Benefits of watermelon smoothie include:

   i. Healthy heart: watermelon contains lycopene which is effective at protecting the cells from damage and lowers the risk of heart disease.

ii. Anti-inflammatory properties: the lycopene is an inhibitor for various inflammatory processes, and it works as an antioxidant to neutralize free radicals. It also contains choline which helps chronic inflammation (Reetu, 2018).

iii. Cancer prevention: it reduces risk of cancer through their antioxidant properties.

iv. Skin and hair: watermelon contains vitamin A which gives moisture to the hair and encourages growth of new collagen and elastin cells.

v. Hydration: the juice contains good electrolytes, which helps to prevent heat stroke increasing hydration.

vi. Digestion: the fiber in watermelon help the digestive tract to perform properly

2. **Plant based drinks – Eviron** (Penning, 2018)

This drink contains the following natural ingredients which are beneficial to the health

i. Korean ginseng: Ginseng increases stamina, exerts hematopoietic action, improves mental condition and nervous functions, increases the secretion of body fluids and quenches thirst (Penning, 2018)

ii. Hovenia dulcis: it protects the liver and has effects on alcoholism, fatty liver, hepatitis, diuretic symptoms, quenching thirst. It is also good for detoxification (Tea Kyung Hyun et al., 2010). The Korea food and drug administration approved in December 2008 that extracts of this fruit can protect and help recover the liver from alcohol.

iii. Rubus coreanus: contains high concentrations of phenolic compounds which prevents stress (Ji Eun Lee et al., 2011).

iv. Aspartic acid: it plays a role in hormone production and release, normal nervous system function and in the conversion of carbohydrates into energy. It stimulates synapses in the

central nervous system and spread messages and instructions through the brain (Penning, 2018).

v. Taurine: it helps in weight loss and obesity, cardiovascular disease, cholesterol reduction, eye protection, liver protection (Robin et al., 2010).

vi. Sucralose: it is a no-calorie sweetener used in foods and drinks (Ademir, 2009)

3. **Herbal drinks – Yoyo bitters**

   Yoyo bitters enhances weight loss, fights bacterial infections, possesses anti-aging effects, helps in relieving pain (McMullen, 2017).

4. **Beverages – Green tea**

   Green tea is rich in catechin, polyphenols (EGCG- it is a powerful anti-oxidant). It has an effect in lowering LDL cholesterol levels (Mishra, 2008), E.g. a report showed that cancer rates tend to be low in countries such as Japan because it is consumed regularly.

5. **Milk based drinks – Yogurt**

   Yoghurt provides many of the nutrients needed for optimal bone health such as calcium, protein, magnesium, zinc and phosphorus. It reduces cardiovascular disease risk and type 2 diabetes (Carrie et al., 2015).

6. **Supplements – Trevor**

   Provides the body with essential vitamins and minerals. It increases energy, enhances mental focus, anti-aging benefits, and helps in weight loss (Dannitrev, 2016).

7. **Red wine**

   It contains significant antioxidants, vitamins, and polyphenols.

The key ingredient is polyphenol resveratrol which have anti-cancer properties and can help reduce instances of coronary artery disease (Anfindsen, 2015).

8. **Water**

Water hydrates, refreshes the body, it clears acne, it has no calorie (Satish, 2018).

CHAPTER THREE

MATERIALS AND METHODS

3.1 Materials

All chemicals and drugs used were of analytical grade. Assay kits for lipid profile assays were purchased from Randox laboratories, UK. Accu-Check glucometer and test strips were used for fasting blood glucose determination.

3.2 Carbonated and Health Drinks

The drinks (Eviron, Bigi-tropical and Water) used in this study were obtained from Mallmart store in Yaba Lagos state, Nigeria. For the bigi drink the NAFDAC Reg. No.: 08-3833, for the eviron, NAFDAC Reg. No.: A1-9281, manufacturing date: 04.11.2019, expiry date: 03.05.2022.

3.3 Study Volunteers

For this research, a total of twenty-four (24) human volunteers were recruited from Mountain top University, Ogun state. The study procedure was carefully explained to all the volunteers, who willingly gave their consent for this research and signed a consent form. All the volunteers blood sample were taken by a trained nurse from the Mountain top University health Centre.

3.4 Study Design

The volunteers were divided into three (3) groups of eight (8) persons each as follows:

Group 1 (water): the volunteers in this group were given 25 mL of Viju bottle water 7am every morning on an empty stomach for 7 days.

Group 2 (health drinks): the volunteers in this group were given 25 mL of Eviron 7am every morning on an empty stomach for 7 days.

Group 3 (carbonated drinks): the volunteers in this group were given 25 mL of Bigi tropical 7am every morning on an empty stomach for 7 days.

The drinks were taken once a day, for a period of seven (7) days and the volunteers in all the groups were asked not to eat until two (2) hours after taking the drinks. Test tubes were labeled according to group and identity given to the volunteers, alongside one test tube for blank and one

test tube for standard. Blood sample collection was done by a trained nurse before they took the drinks on the first day and 2 hours after administration on the 7th day.

3.5 Preparation of Blood plasma

Blood samples were collected by venipuncture using 5 mL disposable polypropylene syringes with 21G needles, and immediately transferred into lithium heparin bottles. These were centrifuged at 2500 rpm for 10 mins. The plasma was separated and stored in the refrigerator at -20°C until use.

3.6 Determination of Fasting Blood Glucose

The fasting blood glucose was determined on day 1 before commencement of administration and on day 7, two hours after administration using AccuCheck glucometer and test-strips.

3.7 Assays for Lipid Profile

Lipid profile which includes; Total Cholesterol (TC), Triglycerides, High-density Lipoprotein-Cholesterol (HDL-C) was determined using standard laboratory kit from Randox laboratories, UK.

3.7.1 Determination of Triglycerides

$$\text{Triglycerides} + H_2O \xrightarrow{\text{lipases}} \text{glycerol} + \text{fatty acids}$$

$$\text{Glycerol} + \text{ATP} \xrightarrow{\text{GK}} \text{glycerol-3-phosphate} + \text{ADP}$$

$$\text{GPO Glycerol-3-phosphate} + 0_2 \longrightarrow \text{dihydroxyacetone phosphate} + H_20_2$$

$$2H_20_2 + \text{4-aminophenazone} + \text{4 chlorophenol} \longrightarrow \text{quinoneimine} + HCl + 4H_2O$$

Procedure: 5µl of the standard and 10µl of the samples were pipetted into different test tubes and 1000µl of the reagent was added to each tube, it was gently mixed and incubated for 10mins

at 25°C and was read at a wavelength of 500nm against the blank that contained 500µl of the reagent. The absorbance of the sample (A_{sample}) and standard ($A_{standard}$) against the reagent blank were measured within 60minutes.

Calculation:

$$\text{Triglycerides concentration (mg/dL)} = \frac{A_{sample}}{A_{standard}} \times \text{Standard concentration (mg/dL)}$$

3.7.2 Determination of Total Cholesterol

The cholesterol is determined after enzymatic hydrolysis and oxidation. The indicator quinoneimine is formed from hydrogen peroxide and 4-aminoantipyrine in the presence of phenol and peroxidase.

$$\text{Cholesterol-ester} + H_20 \xrightarrow{\text{Cholesterol esterase}} \text{Cholesterol} + \text{Fatty acids}$$

$$\text{Cholesterol} + 0_2 \xrightarrow{\text{Cholesterol oxidase}} \text{Cholestene-3-one} + H_20_2$$

$$2H_20_2 + \text{phenol} + \text{4-Aminoantipyrine} \xrightarrow{\text{peroxidase}} \text{quinoneimine} + H_20$$

Procedure: 10µl of the standard and samples were pipetted into different test tubes and 1000µl of the reagent was added to each tube, it was gently mixed and incubated for 10mins at 25°C and was read at a wavelength of 500nm against the blank that contained 10µl distilled water and 1000µl of the reagent. The absorbance of the sample (A_{sample}) and standard ($A_{standard}$) against the reagent blank were measured within 60minutes.

Calculation:

$$\text{Cholesterol concentration (mg/dL)} = \frac{A_{sample}}{A_{standard}} \times \text{Standard concentration (mg/dL)}$$

3.7.3 Determination of HDL-Cholesterol (HDL)

Low density lipoproteins (LDL and VLDL) and chylomicron fractions are precipitated quantitatively by the addition of phosphotungstic acid in the presence of magnesium ions. After centrifugation, the cholesterol concentration in the HDL (high density lipoprotein) fraction, which remains in the supernatant, is determined.

Procedure: For precipitation, 200µl of the samples were pipetted into a centrifuge tube, 500µl of the precipitant was added. It was mixed gently and allowed to sit for 10 minutes at room temperature. Then it was centrifuged for 10 minutes at 4,000 rpm. The clear supernatant was separated and the cholesterol content was used to determine the CHOD-PAP method. For cholesterol CHOD-PAP assay, 100µl of the standard and samples were pipetted into different test tubes and 1000µl of the reagent was added to each tube, it was gently mixed and incubated for 10mins at 25°C and was read at a wavelength of 500nm against the blank that contained 100µl distilled water and 1000µl of the reagent. The absorbance of the sample (A_{sample}) and standard ($A_{standard}$) against the reagent blank were measured within 60minutes.

Calculation:

$$\text{Concentration of HDL-chol in supernatant} = \frac{\Delta A_{sample}}{\Delta A_{standard}} \times \text{Conc. of standard (mg/dL)}$$

3.7.4 Estimation of VLDL-cholesterol and LDL-cholesterol concentrations

VLDL-chol and LDL-chol concentrations were estimated according to the formula of Friedwald et al., 1972 as thus stated in the commercial kit's instructions:

$$\text{Concentration of VLDL-chol (mg/dL)} = \frac{\text{Triglycerides conc.}}{5}$$

Concentration of LDL-chol (mg/ml) = Total cholesterol - VLDL-chol - HDL-chol

3.7.5 Estimation of Atherogenic indices

Atherogenic index of plasma (AIP) and atherogenic coefficient (AC) were estimated according to the following formula's

$$\text{Atherogenic index of plasma (AIP)} = \log_{10}\left(\frac{\text{Triglycerides}}{\text{HDL-chol}}\right)$$ (Sa'adah, 2016)

$$\text{Atherogenic coefficient (AC)} = \frac{\text{Total cholesterol} - \text{HDL}}{\text{HDL}}$$ (Sa'adah, 2016)

3.8 Waste Disposal

Used syringes, needles, cotton wool and sample bottles containing unused blood samples and tissue homogenates were properly disposed.

3.9 Statistical Analysis

The mean values, standard deviation and standard error of means (SEM) of the results were calculated and the data were statistically analyzed using Graph pad prism 7.0. Results were

expressed as a mean ± standard error of mean, n=8. P values less than 0.05 ($p<0.05$) were considered statistically significant.

CHAPTER FOUR

RESULTS

4.1 Effects of carbonated and health drink on fasting blood glucose concentration of volunteers.

The fasting blood glucose of the volunteers are shown in Figure 4.1. There was slight increase in the values recorded for all groups at day 7; water gave an increase of 5.2 mg/dL compared to day

one, making a percentage increase of 0.052, for eviron group, the increase was 0.14% , while bigi-tropical group had 0.082% increase.

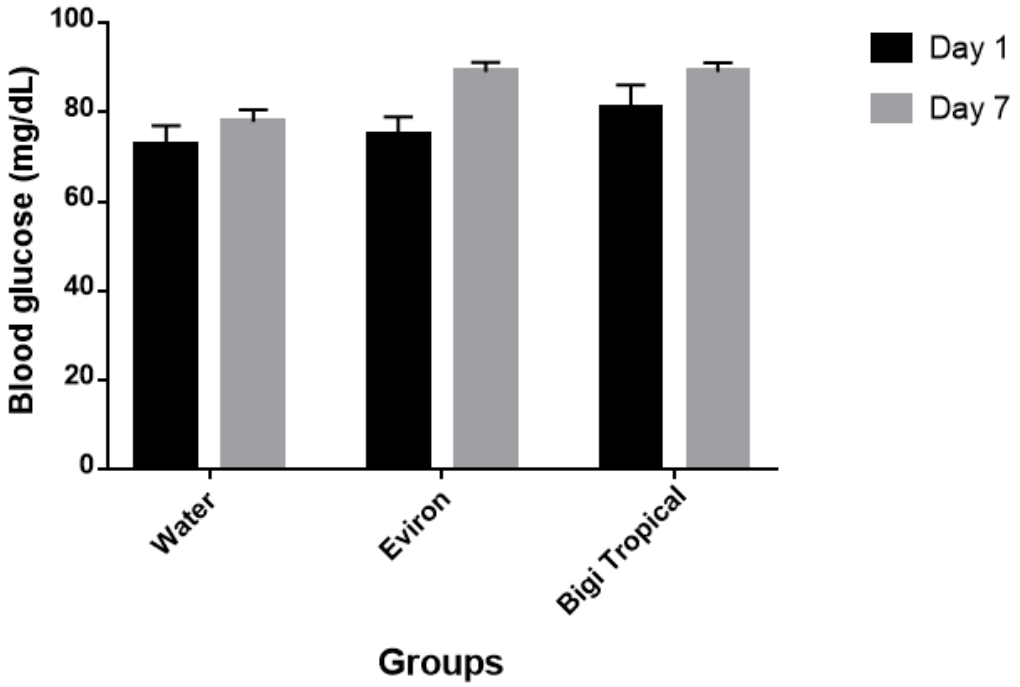

Figure 4.1: Fasting blood glucose levels of volunteers at day 1 & 7

4.2 Effects of carbonated and health drink on plasma triglycerides concentration of volunteers.

Triglycerides concentration of the volunteers are shown in Figure 4.2. The Eviron, group recorded an increase of 3.87% of plasma triglycerides concentration on day 7 compared to day 1, while a significant ($p<0.05$) increase of 12.44% was recorded for the Bigi-tropical group.

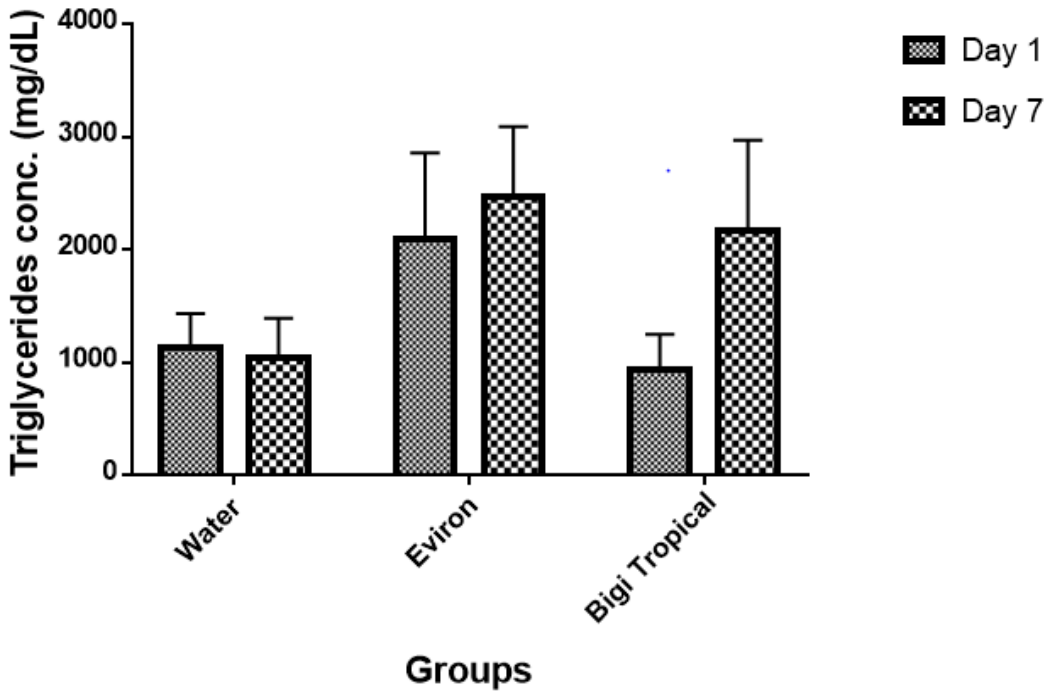

Figure 4.2: Plasma Triglycerides concentration of volunteers at day 1 & 7

4.3 Effects of carbonated and health drink on Plasma Cholesterol concentrations of volunteers.

Plasma cholesterol concentrations of the volunteers are shown in the figure below. There was 1.16% decrease in the value recorded for water group on day seven compared to day one. For eviron group, there was 2.83% decrease, while the bigi-tropical group had 6.61% increase on day 7 compared to day 1 (Figure 4.3).

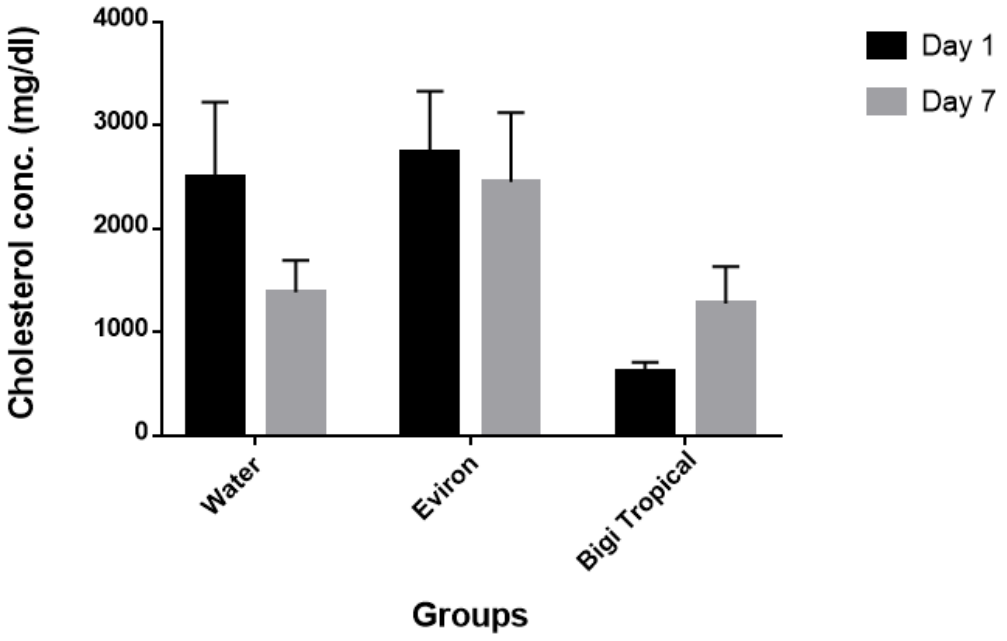

Figure 4.3: Plasma Cholesterol concentrations of volunteers at day 1 & 7

4.4 Effects of carbonated and health drink on Plasma HDL-Cholesterol concentrations of volunteers.

Plasma HDL-Cholesterol concentrations of the volunteers are shown in Figure 4.4**.** Slight increase of 0.49% was recorded in the eviron group on day 7 compared to day 1; bigi tropical group similarly recorded an increase of 2.83%, while unusual decrease was recorded in the group given water.

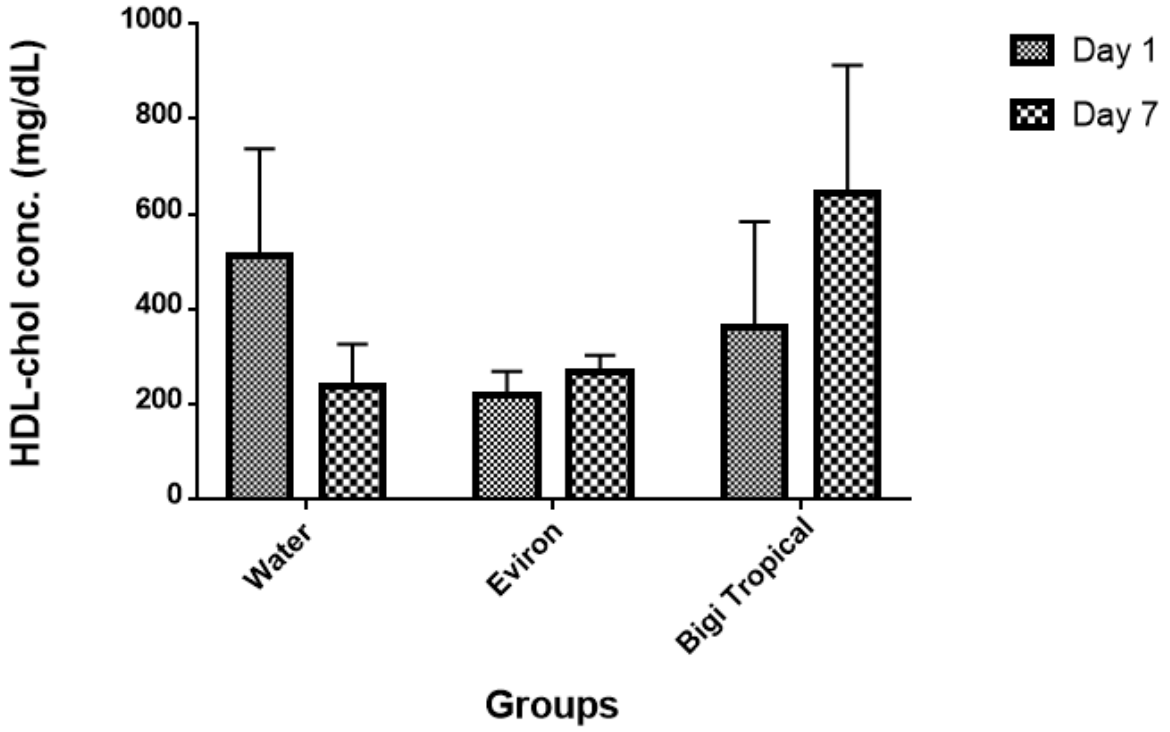

Figure 4.4: Plasma HDL-Cholesterol concentrations of volunteers at day 1 & 7

4.5 Effects of carbonated and health drink on Plasma VLDL-Cholesterol concentrations of volunteers.

Plasma VLDL-Cholesterol concentrations of the volunteers are shown in the figure below. The 25 mL of drinks the volunteers took for seven days resulted in a decrease of 26.22 mg/dL compared to day one in the group given water and thus a decrease of 0.26%. For eviron group, there was no significant ($p>0.05$) difference, while for bigi-tropical group, 2.49% increase in plasma VLDL-cholesterol concentration was recorded (Figure 4.5).

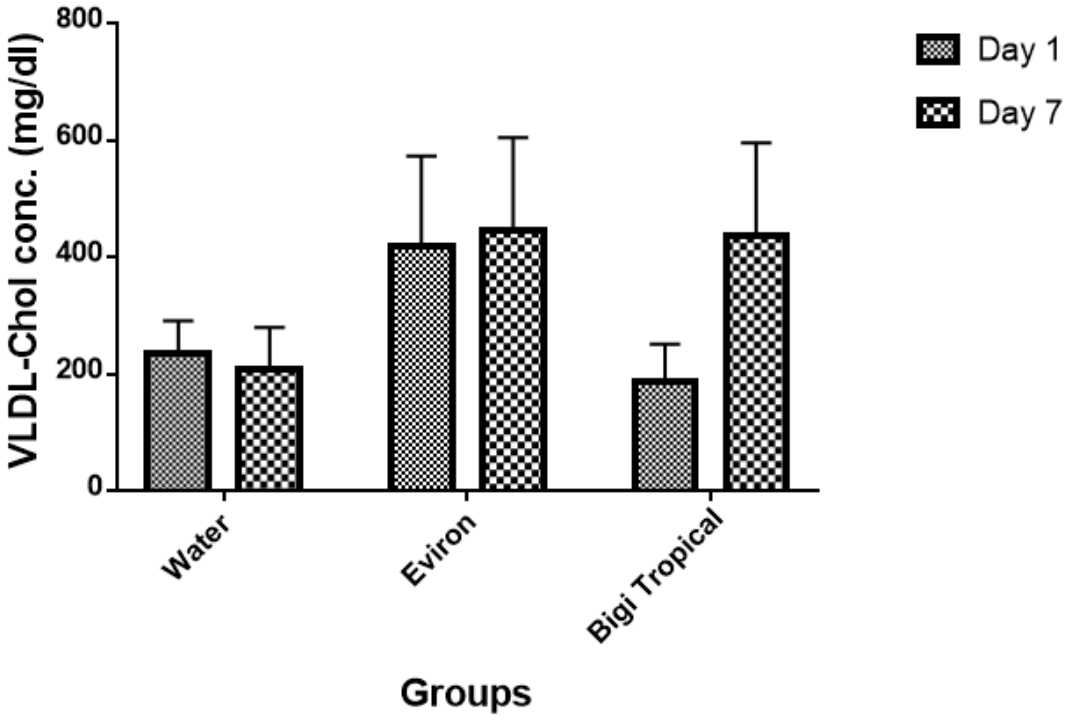

Figure 4.5: Plasma VLDL-Cholesterol concentrations of volunteers at day 1 & 7

4.6 Effects of carbonated and health drink on Plasma LDL-Cholesterol of volunteers.

Plasma LDL-Cholesterol of the volunteers are shown in Figure 4.5. The volunteers who took water for seven days had a decrease of 813.7 2mg/dL in plasma LDL-cholesterol compared to day, giving 8.14% decrease. A decrease of 3.69% was recorded in eviron group, while the bigi-tropical group had 2.01% increase.

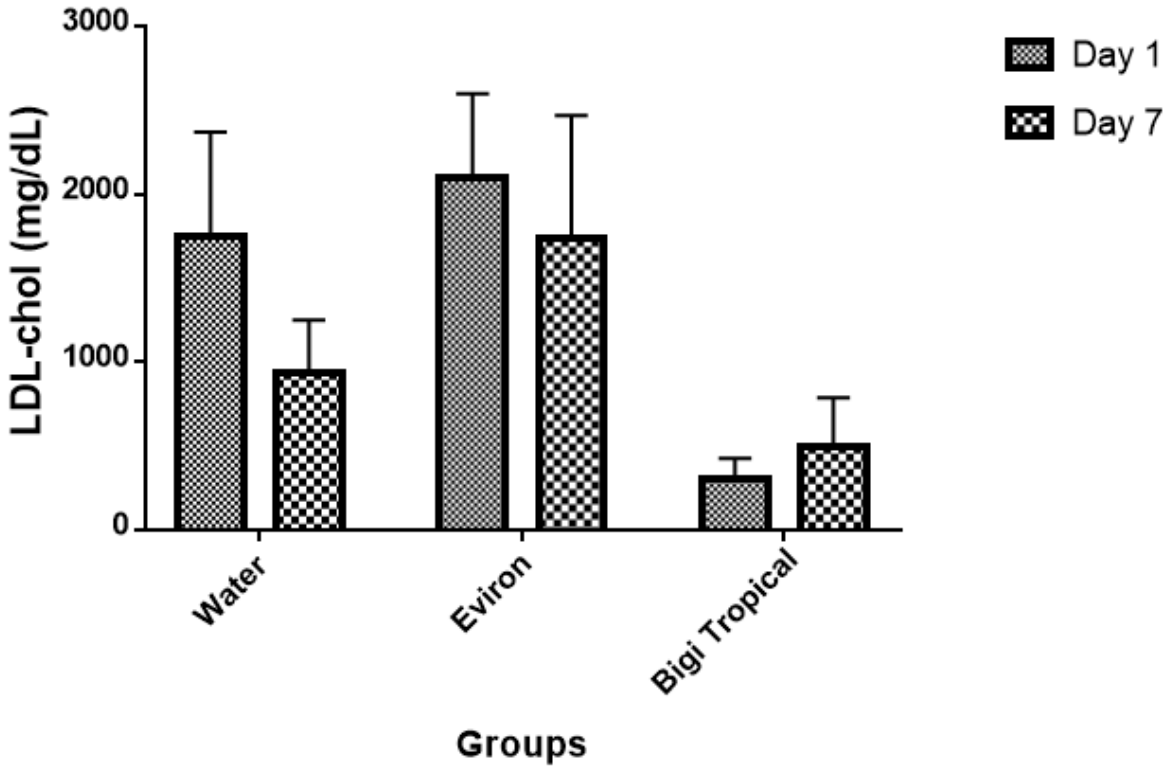

Figure 4.6: Plasma LDL-Cholesterol of volunteers at day 1 & 7

4.6: Effects of carbonated and health drink on atherogenic indices of the volunteers

The atherogenic index of plasma (AIP) and Atherogenic coefficient (AC) of volunteers are shown in the table below. Both water and eviron groups recorded increased AIP on day 7 compared to day 1 and a decrease was recorded for AC in the eviron group. However, the AIP recorded with water at day 7 was normal despite the slight increase. For bigi tropical group, the AIP increased while the AC decreased on day 7 as against day 1.

Table 4.1: Atherogenic index of plasma (AIP) and Atherogenic coefficient (AC) of volunteers

Group	AIP		AC	
	Day 1	**Day 7**	**Day 1**	**Day 7**
Group 1 (Water)	0.48 ± 0.28	0.64 ± 0.17	3.15 ± 0.65	4.1 ± 1.10
Group 2 (Eviron)	0.84 ± 0.31	3.9 ± 1.57	13.9 ± 4.79	9.1 ± 3.21
Group 3 (Bigi-tropical)	0.52 ± 0.13	0.48 ± 0.22	10.2 ± 4.74	0.7 ± 0.52

Data are Mean ± SEM; n=8. AIP: Atherogenic index of plasma, AC: Atherogenic coefficient

CHAPTER FIVE

DISCUSSION AND CONCLUSION

5.1 Discussion

Studies have shown that high sugar-sweetened soft drink consumption has the ability to increase the concentration of fasting blood glucose and can predispose one to many disease states, including obesity, type 2 diabetes and cardiovascular disease (Laughlin et al., 2014). The effects of certain drinks (Bigi-tropical, Eviron, and water) on blood glucose levels and lipid profile parameters were investigated in this research.

Evidence from this research showed that the volunteers who took bigi-tropical appeared to show an increased level of fasting blood glucose compared to the group that took water. It also showed increase in triglyceride, total cholesterol, HDL, VLDL and normal level of LDL. Bigi-tropical is a carbonated drink with added carbon dioxide and sugar. Epidemiological studies have shown that sugar or sugar-sweetened beverage intake is associated with adverse lipid levels, insulin resistance, fatty liver disease, type 2 diabetes and cardiovascular disease (Yoshida et al., 2007). These findings clearly suggest that intake of this drink may not be safe for patients suffering from diabetes, obesity and cardiovascular diseases.

The group that took Eviron, showed normal levels of glucose, and it showed a slight increase in the fasting blood glucose level when day 1 and day 7 values were compared. It showed an insignificant increase level of HDL, triglyceride and VLDL, where as it showed a decrease in the levels of cholesterol and LDL. This implies that the eviron contents did not contain any concentrations of sugar sweeteners or other flavoring agents that may significantly affect blood glucose level and lipid profile. Thus, drinking of eviron should not be considered a predisposing factor for diabetes and other cardiovascular diseases.

In the case of the group given water, there was a significant decrease in total cholesterol, low density lipoprotein, triglyceride and normal level of high-density lipoprotein. A slight, but not significant increase in fasting blood glucose was recorded. These results suggest water intake as being relatively safe. There are a lot of water products that natural sweeteners have been added which would be a better option for carbonated drink lovers rather than taking carbonated drinks. This type of water could be encouraged in place of carbonated drinks. Eradication of carbonated drinks should be a critical issue that should be handled because it affects humans of all age.

5.2 Conclusion

The findings of this study have shown that carbonated drink may likely predispose one to some diseases. Carbonated drinks increased the concentration of blood glucose, which is a marker of diabetes mellitus. Also, the increased total cholesterol and LDL- cholesterol showed may reflect lipid profile abnormality. Eviron did not show any negative effect on some of the lipid profile parameters, hence might be a better option for drink intake for public consumption. Water showed no negative effects too and is the best option for drink intake.

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APPENDIX

Appendix I: Consent form

My name is Akunne Precious, an undergraduate student of the Department of Biological sciences, Mountain Top university. I am soliciting for your participation in a research study titled Comparative effects of carbonated drink (bigi tropical) and health drink (eviron) on some biochemical indices in humans Involvement in the study is voluntary, so you may choose to participate or not.

I am interested in learning more about the effects of a common brand of carbonated drink and a health drink sold in Nigeria on blood glucose and lipid profile in humans. If you decide to participate you will be assigned to a group of your choice, either to take water, carbonated or the health drink for some days, after which you will have to give 5mLof your blood. The sample collection will take approximately 10 minutes. All information and data will be kept confidential; I will assign a number to your responses and data, and only I will have the key to indicate which number belongs to which participant.

The benefit of this research is that you will be helping me to understand the effects of these drinks on health. This information will help in advising Nigerians on their choice of drinks. There is no compensation for your participation and you may personally not experience benefits from participating in this study. However, others may benefit in the future from the information found in this study. There are little or no risks involved in participating in this study except for a slight discomfort during collection of blood sample. This will be minimized by involving a trained phlebotomist. The drinks are non-alcoholic and have not been proven to be of harm to anyone.

If you do not wish to continue, you have the right to withdraw from the study, without penalty, at any time. Please call 08024321285 or email akuunebueb20@gmail.com if you have questions about the study, any problems, unexpected physical or psychological discomforts, any injuries, or think that something unusual or unexpected is happening.

Participant - All of my questions and concerns about this study have been addressed. I choose, voluntarily, to participate in this research project.

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Name of Participant Signature/Date

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Name of Researcher Signature/Date

Appendix II: Fasting blood glucose concentrations (mg/dL) of volunteers.

	WATER (GROUP 1)		EVIRON (GROUP 2)		BIGI-TROPICAL (GROUP 3)	
	Day 1	Day 7	Day 1	Day 7	Day 1	Day 7
a	60	74	64	94	89	92
b	66	93	82	90	91	90

c	74	84	85	81	86	92
d	85	73	68	90	75	90
e	78	85	76	90	63	81
Mean	**72.60**	**77.80**	**75.00**	**89.00**	**80.80**	**89.00**
SD	**9.84**	**6.14**	**8.94**	**4.80**	**11.71**	**4.53**
SEM	**4.40**	**2.75**	**4.00**	**2.15**	**5.24**	**2.01**

Appendix III: Plasma Triglycerides concentration of volunteers.

TRIGLYCERIDES								
	GROUP 1(WATER)				GROUP 2(EVIRON)			
	DAY 1		DAY 7		DAY 1		DAY 7	
	Abs	Concentration	abs	Concentration	abs	Concentration	abs	Concentration
		Mg/dL		Mg/dL		Mg/dL		Mg/dL
A	0.257	1504.2	0.333	1949.0	0.036	210.7	0.367	2148.0
B	0.229	1340.3	0.318	1861.2	0.739	4325.3	0.851	4980.9
C	0.084	491.6	0.109	637.9	0.539	3154.7	0.137	801.9
D	0.335	1960.7	0.060	351.2	0.102	597.0	0.092	538.5
E	0.098	573.6	0.071	415.6	0.372	2177.3	0.454	2657.2
	Mean	**1129.64**		**1042.98**		**2093.0**		**2480.46**
	SEM	**307.29**		**355.43**		**771.32**		**616.76**

	GROUP 3(BIGI-TROPICAL)			
	DAY 1		DAY 7	
	Abs	Concentration	abs	Concentration
		Mg/dL		Mg/dL
A	0.296	1732.5	0.033	193.1
B	0.042	245.8	0.853	4992.6
C	0.128	749.2	0.416	2434.8
D	0.128	304.4	0.228	1334.5
E	0.281	1644.7	0.332	1943.2
	Mean	**935.32**		**2179.64**
	SEM	**319.91**		**796.69**

Appendix IV: Plasma Cholesterol concentrations of volunteers.

CHOLESTEROL(CHOL)				
	GROUP 1(WATER)		GROUP 2(EVIRON)	
	DAY 1	DAY 7	DAY 1	DAY 7

	Abs	Concentration Mg/dL	Abs	Concentration Mg/dL	Abs	Concentration Mg/dL	Abs	Concentration Mg/dL
A	0.393	2992.8	0.204	1553.5	0.319	2429.3	0.356	2711.1
B	0.119	906.2	0.249	1896.2	0.592	4508.3	0.151	1149.9
C	0.316	2406.5	0.272	2071.4	0.459	3495.5	0.477	3632.5
D	0.162	1233.7	0.047	357.9	0.297	2261.8	0.091	693
E	0.653	4972.8	0.138	1050.9	0.132	1005.2	0.538	4097.1
	Mean	**2502.4**		**1385.98**		**2740.02**		**2456.72**
	SEM	**724.91**		**310.44**		**592.98**		**669.18**

	GROUP 3(BIGI-TROPICAL)			
	DAY 1		DAY 7	
	Abs	Concentration Mg/dL	abs	Concentration Mg/dL
a	0.079	601.6	0.036	274.2
b	0.120	913.8	0.273	2079
c	0.052	396	0.085	647.3
d	0.058	441.7	0.185	1408.8
e	0.097	738.7	0.261	1987.6
	Mean	**618.36**		**1279.38**
	SEM	**95.58**		**358.30**

Appendix V: Plasma HDL-Cholesterol concentrations of volunteers.

HDL-CHOLESTEROL (HDL)								
	GROUP 1(WATER)				GROUP 2(EVIRON)			
	DAY 1		DAY 7		DAY 1		DAY 7	
	Abs	Concentration Mg/dL	Abs	Concentration Mg/dL	Abs	Concentration Mg/dL	Abs	Concentration Mg/dL
A	0.017	70.8	0.132	550	0.093	387.5	0.057	237.5
B	0.048	200	0.064	266.7	0.062	258.3	0.093	387.5
C	0.094	391.7	0.009	37.5	0.026	108.3	0.045	187.5
D	0.128	533.3	0.053	220.8	0.047	195.8	0.057	237.5
E	0.326	1358.3	0.028	116.7	0.035	145.8	0.070	291.7
	Mean	**510.82**		**238.34**		**219.14**		**268.34**
	SEM	**226.18**		**87.57**		**49.03**		**36.05**

	GROUP 3(BIGI-TROPICAL)	
	DAY 1	DAY 7

	Abs	Concentration Mg/dL	Abs	Concentration Mg/dL
a	0.299	1245.8	0.045	187.5
b	0.018	75	0.102	425
c	0.042	175	0.122	508.3
d	0.023	95.8	0.407	1695.8
e	0.051	212.5	0.097	404.2
	Mean	**360.82**		**644.16**
	SEM	**222.67**		**268.19**

Appendix VI: Plasma VLDL-Cholesterol concentrations of volunteers.

VLDL				
	GROUP 1(WATER)		GROUP 2(EVIRON)	
	DAY 1	DAY 7	DAY 1	DAY 7
	Concentration Mg/dL	Concentration Mg/dL	Concentration Mg/dL	Concentration Mg/dL
a	300.8	389.8	42.1	429.6
b	268.1	372.2	865.1	996.2
c	98.3	127.6	630.9	160.4
d	392.1	70.2	119.4	107.7
e	114.7	83.1	435.5	531.4
Mean	**234.8**	**208.58**	**418.6**	**445.06**
SEM	**56.24**	**71.09**	**154.27**	**159.11**

	GROUP 3(BIGI-TROPICAL)	
	DAY 1	DAY 7
	Concentration Mg/dL	Concentration Mg/dL
a	346.5	38.6
b	49.2	998.5
c	149.8	486.9
d	60.9	266.9
e	328.9	388.6
Mean	**187.06**	**435.9**
SEM	**63.97**	**159.34**

Appendix VII: Plasma LDL-Cholesterol of volunteers.

LDL				
	GROUP 1(WATER)		GROUP 2(EVIRON)	
	DAY 1	DAY 7	DAY 1	DAY 7
	Concentration Mg/dL	Concentration Mg/dL	Concentration Mg/dL	Concentration Mg/dL
a	2601.2	613.7	1999.7	2044
b	438.1	1257.3	3384.9	-233.8
c	1916.5	1906.3	2756.3	3284.6
d	308.3	66.9	1946.6	293.6
e	3499.8	851.1	423.9	3274
Mean	**1752.78**	**939.06**	**2102.28**	**1732.48**
SEM	**616.99**	**309.12**	**496.21**	**735.49**

	GROUP 3(BIGI-TROPICAL)	
	DAY 1	DAY 7
	Concentration Mg/dL	Concentration Mg/dL
a	159.3	48.1
b	789.6	655.5
c	71.2	-347.9
d	285.0	954.4
e	197.3	1194.8
Mean	**300.48**	**500.98**
SEM	**127.01**	**288.06**

Appendix VIII: Atherogenic index of plasma (AIP) and Atherogenic coefficient (AC) of volunteers

AIP				
	GROUP 1(WATER)		GROUP 2(EVIRON)	
	DAY 1	DAY 7	DAY 1	DAY 7
a	1.3	0.5	-0.2	0.9
b	0.8	0.8	1.2	1.1
c	0.1	1.2	1.5	0.6
d	0.6	0.2	0.5	0.4
e	-0.4	0.5	1.2	0.9
Mean	**0.48**	**0.64**	**0.84**	**3.9**
SEM	**0.28**	**0.17**	**0.31**	**1.57**

	GROUP 3(BIGI-TROPICAL)

	DAY 1	DAY 7
a	0.1	0.01
b	0.5	1.1
c	0.6	0.7
d	0.5	-0.1
e	0.9	0.7
Mean	**0.52**	**0.48**
SEM	**0.13**	**0.22**

Atherogenic index of plasma (AIP) = $\log \frac{\text{(Triglycerides)}}{\text{(HDL-chol.)}}$

AC				
	GROUP 1(WATER)		GROUP 2(EVIRON)	
	DAY 1	DAY 7	DAY 1	DAY 7
A	-	1.8	5.3	10.4
B	3.5	6.1	16.5	2.0
C	5.1	-	31.3	18.4
D	1.3	0.6	10.6	1.9
E	2.7	8.0	5.9	13.0
Mean	**3.15**	**4.1**	**13.9**	**9.1**
SEM	**0.65**	**1.10**	**4.79**	**3.21**

	GROUP 3(BIGI-TROPICAL)	
	DAY 1	DAY 7
A	-0.7	2.2
B	26.7	1.2
C	2.7	-0.2
D	13.7	-0.7
E	8.4	0.8
Mean	**10.2**	**0.7**
SEM	**4.74**	**0.52**

Atherogenic coefficient (AC) = $\frac{\text{Total Cholesterol (TC) - HDL}}{\text{HDL}}$