

**SYNERGISTIC EFFECT OF FRUITS EXTRACTS OF *Balanites aegyptiaca* AND *Adansonia digitata* ON LIPIDERMIC PARAMETERS OF DIABETICS ALBINO RAT****\*<sup>1</sup>Baba Gabi, <sup>2</sup>Hauwa Umar, <sup>2</sup>Hauwa Haruna, <sup>3</sup>Aminu Ado**<sup>1</sup>Department of Biochemistry, Kaduna State University, Kaduna State<sup>2</sup>Department of Applied Chemistry, College of Science and Technology, Kaduna Polytechnic<sup>3</sup>Department of microbiology, Federal University Dustin-Ma Katsina State\*Corresponding authors' email: [babagabi1969@gmail.com](mailto:babagabi1969@gmail.com)**ABSTRACT**

Lipidemia is a significant risk factor for cardiovascular disease, particularly in people with diabetes mellitus. High plasma triglyceride concentration, low HDL cholesterol concentration, and elevated concentration of tiny dense LDL cholesterol particles are some of its distinguishing features. Fruits from *Balanites aegyptiaca* and *Adansonia digitata* have the ability to decrease blood pressure. Therefore, the purpose of this study is to examine the combined effects of the two fruits on the lipidemia of alloxan-induced albino rats. Utilizing the glucose dehydrogenase method, glucometer accu-check was used for the evaluation of blood glucose levels. Investigations were also conducted on alloxan-induced diabetic albino rats to measure total cholesterol, triglycerides, high density lipoprotein (HDL), and low density lipoprotein (LDL). According to the results, *B. aegyptiaca* and *A. digitata* fruit extracts significantly reduced the lipid profiles of the treated groups when used at concentrations of 80:20, 160:40, and 240:60 mg/Kg body weight, respectively. While the lipid profiles were significantly reduced in groups that received treatment at dosages of 100, 200, and 300 mg/kg body weight. However, the groups treated with the highest doses of the extracts demonstrated the most significant effects on the levels of triglycerides (47mg/dl), HDL (79mg/dl), and LDL (40mg/dl). The fruits have shown cumulative effects on their potential to lower the lipid profile of diabetic rats, validating their folkloric usage in the management of cardiovascular illnesses.

**Keywords:** Lipidemia, Blood glucose, Total cholesterol (TC), High Density Lipoprotein (HDL), Low Density Lipoprotein (LDL), *B. aegyptiaca*, *A. digitata*

**INTRODUCTION**

Since the dawn of the human race throughout the millennia, lipidemia disease has a lengthy history. According to coronary heart epidemiology research published in 2002, there is a substantial association within cultural contexts Gordon *et al.*, (2007), between blood lipids, hyperlipidemia, and coronary heart disease (CHD) complications. According to Ginsberg *et al.* (2001), situations considered to be hyperlipidemia occur when the concentration of triglyceride- and cholesterol-carrying lipoproteins in plasma exceeds a predetermined normal limit. Artherosclerosis is the term used to describe the process in which lipoproteins accumulate in the intestinal arteries that branch off the aorta and impede the blood flow to the heart, frequently referred to as a heart attack (Srikanth and Deedwania, 2016). Myocardial infarction (MI) on the other hand is a condition marked by excessive lipoprotein accumulation that prevents the heart's blood flow (Smelt, 2010). One of the main risk factors for cardiovascular disease in people with diabetes mellitus is lipidemia, which is marked by high plasma triglyceride levels, low HDL cholesterol levels, and a rise in the concentration of tiny (Nelson, 2013), dense LDL cholesterol particles. The increased free fatty acid flow brought on by insulin resistance is thought to be the cause of the lipid alterations linked to diabetes mellitus. Numerous cholesterol-lowering medications and dietary supplements are now readily available, giving patients fresh chances to reach desired lipid levels (Smelt, 2010). However, the variety of therapeutic options poses series of challenges subjecting the patient to prioritizing therapy. The prevalence of hypercholesterolemia in patients with diabetes mellitus may not be the issue, but mortality associated to coronary heart disease which has increases exponentially (Whelton *et al.*, 2020), a function of serum cholesterol levels and lowering of cholesterol with

reduced diabetic patients' relative cardiovascular risk with concurrent individualization of dyslipidemia drugs. Most individuals with diabetes mellitus are candidates for statin therapy, and often need treatment with multiple agents to achieve therapeutic goals (Kannel *et al.*, 2012). Diabetics' mellitus (DM) is one the most common endocrinal diseases characterized by hyperglycemia and altered metabolism of lipids, carbohydrates and proteins with an increased risk of complications (Li *et al.*, 2017; Arnold *et al.*, 2020).

*B. aegyptiaca* Del., also referred to as the "Desert date" in English, is a member of the *Zygophyllaceae* family and is one of the most widespread trees in Senegal. It is a native of much of Africa and parts of the Middle East. It is a multibranched, spiny shrub or tree that can grow to a height of 10 m. It has a short trunk and frequently branches from the base. The branches are armed with thick yellow or green thorns that can measure up to 8 cm in length (Khare, 2007). African baobab trees, on the other hand, are often referred to as *A. digitata* L. (*Malvaceae*) trees. According to Rahul, *et al.* (2015), it is regarded as a multi-purpose tree that offers protection in addition to supplying food, clothes, medicine, and raw materials for numerous useful goods. The fruit pulp's rich vitamin C, calcium, phosphorus, carbohydrate, fiber, potassium, protein, and fat content makes it ideal for flavoring, preparing drinks, and serving as an appetizer. Significant amounts of phosphorus, magnesium, zinc, sodium, iron, and manganese are present in seeds, whereas lysine, thiamine, calcium, and iron are present in high concentrations (Malabadi, *et al.*, 2021). The plants have a wide range of biological qualities, including, among others, antiviral, antioxidant, anti-inflammatory, anti-malarial, diarrheal, anemia, asthma, and antibacterial capabilities (Deora & Shekhawat, 2019; Siddiqui *et al.*, 2020). An analysis of the plant's phytochemicals found flavonoids,

phytosterols, amino acids, fatty acids, vitamins, and minerals (Braca *et al.*, 2018; Ismail *et al.*, 2019). Plant extracts were traditionally utilized to manage blood sugar levels, particularly in diabetic patients, without taking other disease-related adverse effects into account. The goal of the research is to ascertain the combined impact of *B. aegyptiaca* and *A. digitata* on the lipidermic parameter of alloxan induced diabetes mellitus rats.

## MATERIALS AND METHODS

### Sample Collection and Preparation.

*B. aegyptiaca* and *A. digitata* fruits were purchased from central markets Kaduna, identified and authenticated at herbarium units of Ahmadu Bello University and Kaduna State University with the respective voucher numbers as ABU06123 and KASU/PCG/HERB/101 for *B. aegyptiaca* and *A. digitata*. The pericarp of the fruits were cleaned, dry and then prepared in the form of coarse powder. Cold mass extraction method was used to obtain the extract using aqueous. One hundred (100g) of the macerated powder of the fruits were mixed with 1000ml of distilled water for 60mins under continuous stirring. The aqueous extract was maintained in 4°C until required for used.

### The Experimental Animals

The investigation was conducted using 18 male and female Wistar rats, aged 4 to 8 weeks. The rats were obtained from the Department of Applied Biology's biological garden at Kaduna Polytechnic in Kaduna. The rats were housed in cages with ordinary brackets that were suspended. Before the trial began, the animals were housed in typical experimental conditions and given two weeks to get acclimated to their surroundings. They were given a conventional diet of important feed ground cereals, ample water, and a 12-hour day/night cycle.

### Induction of Diabetes

Prior to receiving an alloxan injection to induce diabetes, albino rats were starved for 16 hours. 150mg/kg of the alloxan solution was administered intraperitoneally to the groups of the experimental model after being dissolved in 5ml of distilled water. The diabetic group of rats was defined as those with a fasting plasma glucose level of higher than 200 mg/dl.

### Animals Grouping

The rats were initially weighed, marked for easy identification, and then randomly divided into 6 groups of three. The groups were as indicated, with the Treatment groups receiving *B. aegyptiaca* and *A. digitata* fruit extracts. Group 1(Normal control): Rats not diabetic induced not treated with drugs/extracts but administered distilled water and feed only.

Group 2(Negative control): Rats diabetic induced with alloxan but no treatment.

Group 3(Positive control): Rats diabetic induced and treated with standard diabetic drugs.

Group 4(Test low dose): Rats diabetic induced and treated with 100 mg/Kg body weight of fruits extract of *B. aegyptiaca* and *A. digitata*.

Group 5 (Test medium dose): Rats diabetic induced and treated with 200 mg/Kg body weight of fruit extracts of *B. aegyptiaca* and *A. digitata*

Group 6 (Test high dose): Rats diabetic induced and treated with 300mg/Kg body weight of fruit extracts of *B. aegyptiaca* and *A. digitata*.

These treatments lasted for 28days with their fasting blood sugar (FBS) being determined for 3days.

### The Drug (fruits extracts) Preparation and Administration

The treatment for the period of 21days was initiated after alloxan was administered. The extracts doses administered were 100mg/kg, 200mg/kg and 300mg/kg body weight dose to group 4, 5 and 6, low, medium and high doses respectively via oral route. The dose were administered daily for a period of 21days. The preparation of the drugs were based on ratio of (0.8:0.2g), for low dose (1.6:0.4g) for, medium dose and (2.4:0.6g) for high doses of *A. digitata* and *B. aegyptiaca* respectively.

### Blood Analysis

After the animals were sacrificed, the blood was collected into sterile, dry centrifuge tubes and allowed to coagulate for 30 minutes at room temperature in a water bath. To separate the serums, the blood was centrifuged for one minute at 3000 rpm. Serums were carefully aspirated, placed in sterile, plastic tubes, and stored there until analysis.

### Total Cholesterol Test

Enzymatically, the 3-OH group of cholesterol was oxidized and hydrolyzed in a sequence of linked reactions in serum or plasma to assess cholesterol. In a peroxidase-catalyzed process that results in color, H<sub>2</sub>O<sub>2</sub> is quantified as one of the reaction byproducts. At 500nm, absorbance is measured. The concentration of cholesterol has a direct relationship with color intensity.

### HDL-Cholesterol Test

In order to measure HDL-cholesterol in serum or plasma while simultaneously taking other lipoprotein particles into account, the HDL-cholesterol test employs a homogeneous two-reagent system. The test is divided into two phases; Cholesterol oxidase and peroxidase solubilize and consume free cholesterol in non-HDL lipoproteins in the first stage, creating a colorless end product. The selective solubilization of HDL-lipoproteins with a unique detergent during phase two freed HDL cholesterol for reaction with cholesterol esterase, cholesterol oxidase, and a chromogen system to form a blue-colored complex that can be detected at 600 or 700 nm. Inversely and directly correlated with the subsequent increase in absorbance is the HDL-C content in the sample.

### LDL – Cholesterol Test

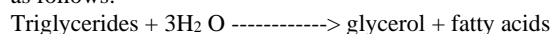
LDL – cholesterol is calculated from the measured values of total cholesterol, triglycerides and HDL – cholesterol according to the relationship.

$$[LDL - chol] = [total chol] - [HDL chol] - [TG]/5$$

Where [TG]/5 is an estimate of VLDL – cholesterol and all values are expressed in mg/dl.

### Triglycerides Estimation

Triglycerides are measured enzymatically in serum or plasma using a series of coupled reaction in which triglycerides are hydrolyzed to produced glycerol. Glycerol is then the oxidized using glycerol oxidase, and H<sub>2</sub>O<sub>2</sub>, one of the reaction products is measured as described above for cholesterol. Absorbance is measured at 500nm. The reaction sequence is as follows:



### Statistical Analysis

Every value was expressed as the mean less the standard deviation. To find the significant differences between the variables: glucose concentration and lipid profiles of the animal groups, the data were subjected to descriptive analysis

and analysis of variance (ANOVA) using the SPSS 17 software package. The treatment means were compared using the (LSD) test, where a significant p-value of 0.05 was regarded as significant.

## RESULTS AND DISCUSSION

### Results

The results of the average weight of the experimental animals before and after treatment on induction of diabetics were represented in Table 1, Considerable loss of weight was observed among the group that where diabetics induced. There is a general loss of weight between 4.5% for the group treated with highest dose to 54.6% for the negative group induced but not treated, which might be due to external stress as a result of the induction and treatment (Table 1).

Table 2 displays the outcomes of blood glucose monitoring following the induction of diabetes. The blood glucose levels of the induced animal groups showed a significant rise, rising from 62.0% for the group receiving the highest doses (300mg/Kg) of extract to 78.2% for both groups receiving the lowest doses (100mg/Kg) and for the group receiving no drug at all i.e. negative control. While the positive control group and medium doses (200mg/Kg) both significantly increased to 74.0±0.03% and 69.2±0.05% respectively. The non-induced control group, on the other hand, did not exhibit any increase in the percentage level of serum glucose because they did not have diabetes. With the exception of the normal

controls, who weren't at all induced, the induction of the diabetics and the rise in glucose levels were successfully accomplished across the board. Figure 1 to 4 represent the results of lipid profile of the alloxan induced groups of rats Figure 1 displays the total cholesterol levels in the treated groups, with the group that got the highest treatment dosages exhibiting the greatest reduction. The average triglyceride profile for the induced and treated groups is shown in Figure 2. As the doses increased, the average triglyceride showed inversely proportionate impacts on all of the treated groups, obviously lowering the triglyceride concentration, with more noticeable effects on the groups receiving the highest dose. Low-density lipoproteins (LDL) levels are evidently falling in all treatment groups, with the benefits being more prominent in the groups given the maximum dose (Figure 3). As can be seen in Plate 1, the liver from the group of animals given the greatest extract concentration has a level of LDL that is higher than the typical normal amount of anticipated fat deposits from that group. The high-density lipoprotein concentration in Figure 3 demonstrates that the effect of the extracts on all treatment groups is supported by the fact that the group treated with a medium dose had relatively the greatest HDL levels, and the high and low doses were almost equal. In contrast, Plate 2 shows the liver from the group of animals treated with the lowest concentration of extracts, clearly indicating tissue without any fat deposits.

**Table 1: Average Weight and the % weight loss of the Albino Rats Before and After Treatment with *B. aegyptiaca* and *A digitata***

Treatment	Average weight before treatment (gm.)	Average weight after treatment (gm.)	Average (%) weight loss	Percentage
Normal control	298.322 ± 0.05 <sup>a</sup>	234.5 ± 0.03 <sup>a</sup>	21.3±0.03	
Positive control	213.056 ± 0.05 <sup>b</sup>	121.2 ± 0.04 <sup>ab</sup>	43.1±0.01	
Negative control	303.941 ± 0.03 <sup>b</sup>	137.8 ± 0.02 <sup>c</sup>	54.6±0.04	
Low dose	168.291 ± 0.04 <sup>b</sup>	144.6 ± 0.02 <sup>c</sup>	14.0±0.02	
Medium dose	154.93 ± 0.06 <sup>b</sup>	123.6 ± 0.03 <sup>ab</sup>	20.2±0.04	
High dose	236.964 ± 0.01 <sup>b</sup>	226.3 ± 0.04 <sup>bc</sup>	4.50±0.03	

**Table 2: Average serum Glucose Level and % increase in the level of the Albino rats, before and After Alloxan Induction of Diabetics**

Treatment	Before induction mMol/dl	After induction mMol/dl	Average percentage % Increase in Serum Glucose (%)
Normal control	6.6 ± 0.01 <sup>a</sup>	7.1 ± 0.02 <sup>a</sup>	7.0±0.05
Positive control	6.0 ± 0.03 <sup>b</sup>	23.1 ± 0.01 <sup>ab</sup>	74.0±0.03
Negative control	6.3 ± 0.03 <sup>b</sup>	29.0 ± 0.01 <sup>c</sup>	78.2±0.03
Low dose	6.3 ± 0.06 <sup>b</sup>	29.0 ± 0.04 <sup>c</sup>	78.2±0.05
Medium dose	6.0 ± 0.07 <sup>b</sup>	19.5 ± 0.04 <sup>ab</sup>	69.2±0.05
High dose	6.0 ± 0.06 <sup>b</sup>	15.8 ± 0.09 <sup>bc</sup>	62.0±0.07

The Data are presented as mean ±SD and n=5; the values with different superscript differ significantly (p<0.05)

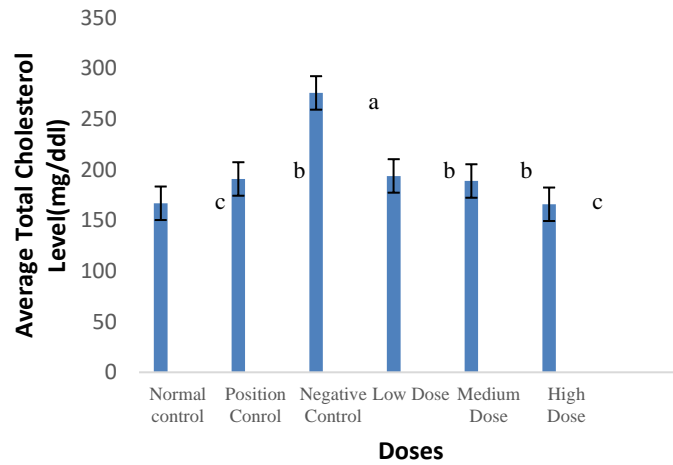


Figure 1: Average Total Cholesterol Level of Albino Rats Treated with *B. Aegyptiaca* and *A. digitata*. \*The values with different superscript differ significantly (p<0.05)

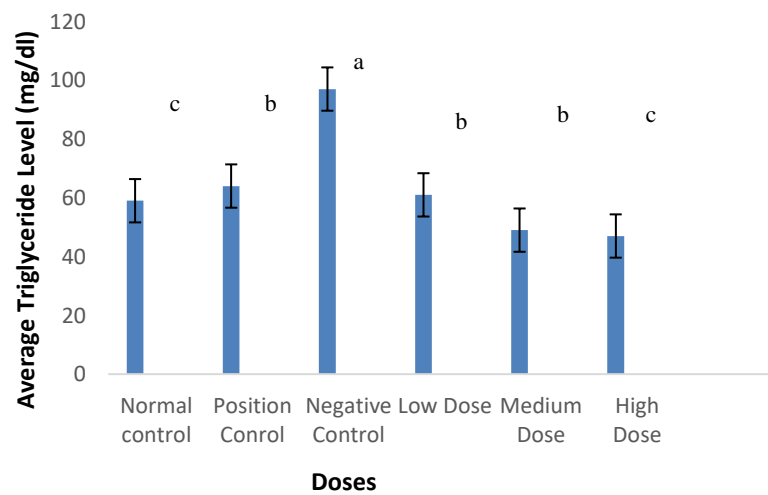


Figure. 2: Average Triglyceride Level of Albino Rats Treated with *B. aegyptiaca* and *A. digitata*. \*The values with different superscript differ significantly (p<0.05)

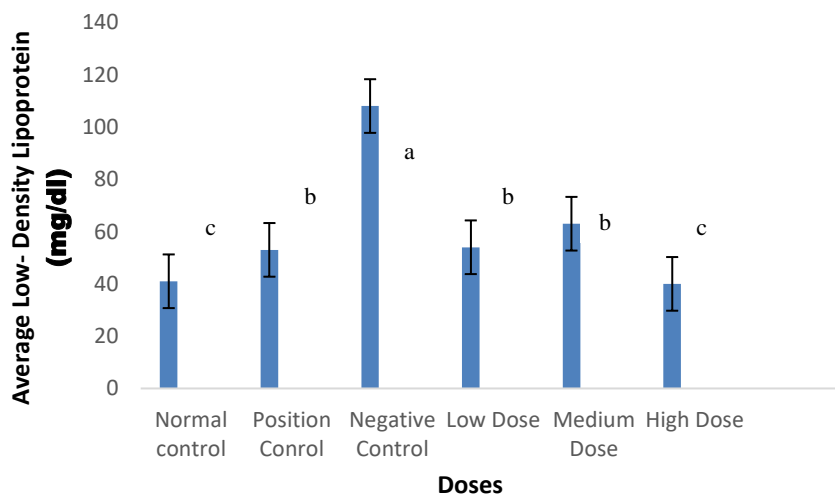


Figure 3: Average Low- Density Lipoprotein Level of Albino Rats Treated with *B. aegyptiaca* and *A. digitata*. \*The values with different superscript differ significantly (p<0.05)

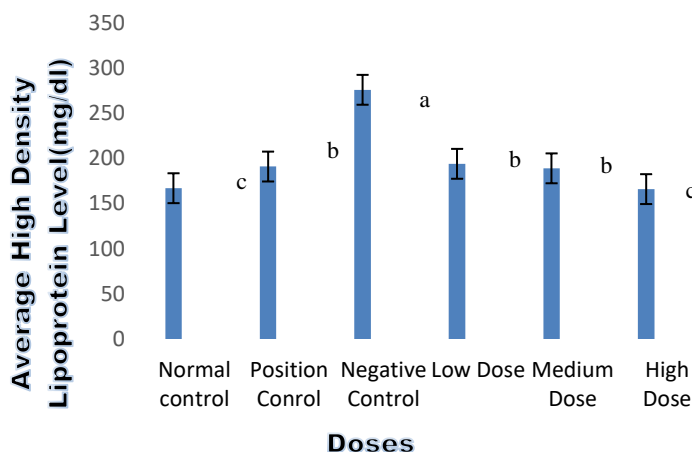


Figure 4: Average High Density Lipoprotein Level of Albino Rats Treated with *B. aegyptiaca* and *A. digitata*  
\*The values with different superscript differ significantly ( $p < 0.05$ )



Plate 1: Fat Deposits (white patches) on the Liver of Normal Control Group



Plate 2: Fat Deposits (white patches) Liver of Albino Rats From Treatment Group

### Discussion

Loss of weight is a hallmark of diabetes. Rats' body weight significantly decreased after an alloxan induction. According to Obidah *et al.*, (2009), alloxan monohydrate is known to cause diabetes by partially destroying pancreatic beta cells in the islet of Langerhan. This happens as a result of hyperglycemia and low insulin levels, which are symptoms of type 1 diabetes mellitus. As a result, the alloxan-treated rats seem to be a promising laboratory model for experimental diabetes with residue or lingering insulin secretion by the pancreatic beta cells. As daily insulin treatment is not necessary for survival in Alloxan-treated diabetic animals, the diabetic condition of diabetic rats treated with Alloxan is consequently different from that obtained by complete pancreatectomy (Akah *et al.*, 2004). Recent studies demonstrate that the administration of *B. aegyptiaca* seed extract for 3 days considerably lowers serum levels of glucose, cholesterol, triglycerides, low density lipoproteins, and very low density lipoproteins (Gordon *et al.*, 2020). The fruits of *A. digitata* exhibit significant levels of flavonoids, according to a different study by Osman (2004), which lower mice's lipid profiles and blood sugar levels. The combined effects of these two extracts are more potent than what was discovered in earlier research on either of the two plants separately.

When compared to non-diabetic rats that were not given any treatment (Table 1), diabetic rats demonstrated a considerable weight loss following the establishment of diabetes (2 weeks following the administration of alloxan solution). But the experimental rats displayed a remarkable recovery in their

body weight two weeks following the start of the aqueous extract treatment. The capacity of the extracts to lower hyperglycemia, which in turn corrects irregularities including loss of body weight (Gad *et al.*, 2006), may be the cause of the subsequent increase in the average body weight of the rats. The glycemic profiles of the rats before and after being induced with 150 mg/kg body weight of alloxan were shown in Table 2. There were notable variations in glucose levels, which demonstrate a significantly elevated glucose level ( $p < 0.05$ ) following the induction of alloxan. Although Szkudelski (2001) reported that rats were injected with an alloxan solution (150 mg/kg body weight) to produce hyperglycemia, the normal control group treated with only distilled water and fed after induction did not exhibit any variation in the blood glucose test before the induction, which is consistent with the literature. According to Szkudelski (2001), diabetic animals were those whose fasting plasma glucose levels were more than 200 mg/dl.

The amount of cholesterol in the blood is known as total cholesterol, which is made up of both high-density lipoprotein (HDL, or "good" cholesterol), and low-density lipoprotein (LDL, or "bad" cholesterol). However, cholesterol, a chemical that resembles waxy fat, is present in every cell of the body and a healthy level is 200 mg/dl. In accordance with Figure 1 of the research findings, the low dose group (treated with fruit extract at a concentration of 100 mg/kg body weight) and positive control (induced with alloxan but treated with a standard diabetic drug) had cholesterol levels that were nearly identical at 191 mg/dl and 194 mg/dl, respectively. The highest lipidemia profile was found in the negative controls,

which were alloxan-induced but not otherwise treated, as neither an extract nor a regular medication was given to the groups. The group that received the most fruit extracts overall, or 300 mg/kg body weight, showed the lowest total cholesterol level, which was 166 mg/dl. This is important to notice as it demonstrates that increasing dosages of fruit extract can have a diminishing impact on lowering overall cholesterol levels.

The blood contains a particular kind of fat (lipid) called triglycerides. Any calories consumed that the body cannot utilize immediately away are transformed into triglycerides by the digestive system. Triglycerides are kept in fat cells for later use and its typical healthy range is below 100 mg/dl. According to the research findings in Figure 2, the triglyceride levels of all the groups were within the normal range, but the negative control group still had the greatest concentration, at 97 mg/dl, followed by the positive control group, at 64 mg/dl. It was discovered that normal controls that had only water and food as well as low doses (treated with 200 mg/kg body weight) had triglyceride levels that were quite similar to each other, at 59 mg/dl and 61 mg/dl, respectively. The lowest triglyceride level was found to be 53 mg/dl in the groups fed the most fruit extract, at a dosage of 300 mg/kg body weight. This demonstrates that high doses of the fruit extract have a reducing effect on triglyceride levels.

LDL cholesterol is usually referred to as "bad" cholesterol since it congregates in the blood vessel walls and raises the risk of health problems like a heart attack or stroke. It is common for the result to fall between 0 and 70 mg/dl. According to the research finding in Figure 3, the lipid profile of the three test groups was significantly lower than that of the negative control group (induced but untreated). The difference is enormous; the negative control's 108 mg/dl level is much greater than the accepted standard threshold. With values of 40 mg/dl and 41 mg/dl, respectively, the highest dose value even comes extremely near to the standard control group's level, which were administered simply water and food. This demonstrates that fruit extract can lower LDL levels (Hajat et al., 2004).

Absorbing cholesterol and transporting it back to the liver is HDL cholesterol, also known as "good" cholesterol, and then the liver excretes it from the body. The risk of heart disease and stroke can be reduced by high HDL cholesterol levels. The typical range is 60 to 150 mg/dl. The HDL value for normal controls that were simply provided water and food was discovered from the research results in Figure 4 to be 62 mg/dl. With respect to the test groups' respective levels for the low dose, medium dose, and high dose, which were 75 mg/dl, 82 mg/dl, and 79 mg/dl, respectively, the HDL level increased in each group, including the negative control, showing that high doses of the fruit extract equally have a reducing effect on HDL levels.

This impact can be related to the potential presence of several phenolic compounds in *B. aegyptiaca* and *A. digitata* fruit, which have the power to significantly reduce lipid profiles and include large amounts of flavonoids that may act as antioxidants and free radical scavengers. According to earlier research on the antioxidant activities of *B. aegyptiaca* and *A. digitata* fruit (Dougar et al., 2007; Gaur et al., 2008), it has been observed that they lowered the levels of lipid profiles.

## CONCLUSION

Fruit extracts from *B. aegyptiaca* and *A. digitata* have shown additive effects in their potential to decrease the lipid profile of diabetic rats, validating their traditional usage in the management of cardiovascular illnesses.

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