World Journal of
Advanced Science and Tehnology

Journal homepage: https://zealjournals.com/wjast/

(RESEARCH ARTICLE)

Hypolipidemial potentials of selected seed oils on high-fat fed adult albino rats

Fatai Adebayo Kareem [1, 2, *], Yesiru Adeyemi Kareem [3], Adeola Oladayo Osifeso [1], Mutiu Adewunmi Alabi [4] and Olasunkanmi Adedoyin Adetutu [5]

[1] *Department of Science Laboratory Technology, School of Science and Technology, Gateway Polytechnic, Saapade, Ogun State, Nigeria.*
[2] *Department of Medical Biochemistry, Lagos State University, Ikeja campus, Lagos State, Nigeria.*
[3] *Department of Clinical Services, Neuropsychiatric Hospital, Aro, Abeokuta, Nigeria.*
[4] *Department of Medical Biochemistry and Pharmacology, Faculty of Pure and Applied, Sciences, Kwara State University Malete, P.M.B. 1530 Ilorin, Nigeria.*
[5] *Department of Science Laboratory Technology, Federal Polytechnic, Ilaro, Ogun State, Nigeria.*

World Journal of Advanced Science and Technology, 2024, 06(01), 025–034

Publication history: Received on 25 May 2024; revised on 02 July 2024; accepted on 05 July 2024

Article DOI: https://doi.org/10.53346/wjast.2024.6.1.0039

Abstract

Annually, cardiovascular diseases (CVDs) claim the lives of more than 17 million individuals. In this study, hypolipidemia potential of *Persea americana, Moringa oleifera, Chrysophyllum albidum* and *Capsicum annuum* seed oils were evaluated. Fifty adult Wistar rats of both sexes weighing between 120 and 170 g were grouped into ten, with five animals in each group. The animals were stabilized for one week prıor to the induction period. The seed oils were administered in their meal for fourteen days. Blood samples were collected through a cardıac puncture into heparinized tubes centrifuged at 5000 rpm for 10 minutes and were used for haematological and lipid profile assays. Comparatively between the analyte and control groups, the result showed no significant difference ($p \geq 0.05$) in haemoglobin (HB) with a significant increase ($p \leq 0.05$) in the packed cell volume (PCV) level of the rats. In addition, there were significant reductions in the levels of triglycerides, total cholesterol, and low-density lipoprotein-cholesterol. Among the male rats of the analyte and control group, there was a significant increase ($p<0.5$) in the level of high-density lipoprotein-cholesterol ın their blood samples. This study reveals that the rich essential fatty acid reported in the seed oils has hypolipidemic effect potential.

Keywords: Cardiovascular diseases; Hypolipidemic; Packed cell volume; Seed oil; Triglycerides; Total cholesterol

1 Introduction

Cardiovascular disease has emerged as a significant worldwide and public health issue within the category of non-communicable diseases [1,2]. Cardiovascular diseases (CVDs) have been the primary cause of death worldwide in recent decades [3]. CVDs were responsible for approximately 17.7 million fatalities in 2015, accounting for 31% of all global deaths [4]. Additionally, it is worth mentioning that over 75% of cardiovascular disease (CVD) fatalities occur in countries with low and intermediate incomes [4]. Cardiovascular disease risk factors refer to specific traits, which may be modified or cannot be modified, that elevate the likelihood of having cardiovascular diseases. These risk factors are becoming more prevalent in developing countries worldwide [3]. Hyperlipidemia, sometimes referred to as dyslipidemia, has been extensively documented as a risk factor for cardiovascular disease (CVD) [5]. Hyperlipidemia is a long-term metabolic disorder characterised by excessive levels of lipids in the blood, specifically high levels of total cholesterol, total triglycerides, and LDL-c, and low levels of HDL-c. This condition can lead to a variety of chronic metabolic disorders [6].

* Corresponding author: Fatai A. Kareem

As a common risk factor for CVDs, dyslipidemia is defined as high blood levels of lipids (cholesterol, triglycerides, or both) carried by lipoproteins; symptoms include high cholesterol >200 mg/dL, low HDL < 40 mg/dL, and high TG ≥ 150 mg/dl [7]. The use of lipid-lowering medications may have certain unfavourable effects [8].

The use of plants in the treatment of hyperlipidemia and cardiovascular disorders has grown in recent years, and there is a growing consumer demand for fresh, natural, safe, and healthful foods that need little work or time to prepare [9]. Actually, because they have a low to medium energy density and are significant providers of major nutrient deficiencies, dietary guidelines worldwide advocate consuming more fruit and vegetables. Various fruit and vegetable varieties are also being reevaluated and categorised as excellent sources of nutraceuticals [10].

Therefore, this study was designed to evaluate the effect of Avocado pear, Moringa, African star apple, and pepper seed oil on their composition by assessing their fatty acid composition, hypolipidemic effect, and chemical properties in high-fat diet-induced adult Albino rats of both sexes.

2 Materials and methods

2.1 Plant Materials

Dust was removed from the fruits by washing them in clean water. The outer skin was peeled off and the seeds were extracted from within. After being chopped into pieces, the *Persea americana* (avocado) and the *Chrysophyllum albidum* (African star apple) were sun-dried for approximately a week in order to remove any remaining moisture. Fruits were manually opened and seeds were removed after sorting and cleaning. The seeds were washed and even dried at 45°C using a vented air-drying for two days. *Moringa oleifera seeds and* Pepper *(Capsicum annuum)* seeds oil were also processed for extraction using n-hexane.

2.2 Extraction

In order to prepare the soxhlet extractor for the first run, 350 g of each powdered seed was weighed individually into a thimble (a semipermeable membrane) and 350 ml of hexane was added. To obtain the recovered lipids, the solid particles were eliminated using filtration. The device was set up on the heater and left to operate for two hours. A test tube containing the extracted oil was set aside for distillation and characterization. This oil extraction technique was demonstrated quite well by Okene and Evbuomwan [11].

2.3 Chemical Characterization of Seeds Oil

The peroxide value of the seed oils was obtained following the spectrophotometric method (2010). The iodine value essay was carried out according to the Association of Official Analytical Chemists (AOAC) [12] method. The acid value was evaluated as described by Okene and Evbuomwan [11].

2.4 Animal Model and Experimental Design

Fifty Albino rats were used in this study. They were bred at the animal house of the Department of Biochemistry of Gateway Polytechnic Saapade. The rats comprised of fıfty albıno rats weighing between 120 and 170 g. Each rat was housed alone in a cage and exposed to a 12-hour cycle of light and darkness for the whole duration of the experiment. The animals were allocated randomly into 10 groups, each consisting of five rats. They were provided with a conventional food and unlimited access to water for a period of one week in order to stabilise their metabolism before the start of the experiment. Additionally, certain groups were provided with a high-fat diet. The rats were given seed oils orally once a day for a period of 14 days, using a gastroesophageal tube. The studies were conducted in accordance with the regulations and ethical permission of the Experimental Animal Welfare and Ethics Committee of the institution.

Briefly, they were distributed as follows:

- GROUP 1: Standard diet + 2 ml Distilled water (CTR)
- GROUP 2: Standard diet + 2 ml Turkey oil (TOSD)
- GROUP 3: Standard diet + 2 ml avocado pear seed oil (AOSD)
- GROUP 4: High-fat diet + 2 ml avocado pear seed oil (AOSHFD)
- GROUP 5: Standard diet + 2 ml Pepper seed oil (POSD)
- GROUP 6: High-fat diet + 2 ml Pepper seed oil (POSHFD)
- GROUP 7: Standard diet + 2 ml Moringa seed oil (MOSD)
- GROUP 8: High-fat diet + 2 ml Moringa seed oil (MOSHFD)

- GROUP 9: Standard diet + 2 ml African star seed oil (ASASD)
- GROUP 10: High-fat diet + 2 ml African star seed oil (ASHFD)

The rats were provided unrestricted access to food and water.

2.5 Diet Composition

A broiler finisher was purchased in the market. The composition of the high-fat diet used for experimental diet is shown in the Table below.

Table 1 Composition of High Fat Diet

Ingredients	Hyperlipidemic diet
Protein	20%
Fats	3.5%
Fibre	3.5%
Calcium	1.0%
Phosphorus	0.5%
Lysine	1.1%
Methionine	0.45%
Net. Energy	2800 Kcal/kgmin

2.6 Serum Collection and Lipid Assessment

After the last dose of gastric substances, rats were anaesthetized and blood samples were obtained from the abdominal aorta. The blood samples were then transferred into laboratory test tubes without any material to prevent clotting. The animals were euthanized by immediately dislocating their cervical vertebrae, and the critical organ (heart) was then isolated. Following a 30-minute incubation period at ambient temperature, blood samples undergoing coagulation were subjected to centrifugation at a force of 3000 g for a duration of 15 minutes. The liquid portion (sera) was obtained using micropipettes and preserved in Eppendorf tubes at a temperature of -20°C for biochemical analysis. The photometric approach using Chronolab commercial kits was used to test total cholesterol (TC), triglycerides (TG), and high density lipoprotein cholesterol (HDL-c) [13]. The concentration of very low-density lipoprotein cholesterol (VLDL-c) was calculated by dividing the concentration of triglycerides (TG) by 5 ([VLDL-c] = [TG]/5). The concentration of low-density lipoprotein cholesterol (LDL-c) was approximated using the Friedewald equation ([LDL-c] = [TC] − ([HDL-c] + estimated [VLDL-c])). The concentrations were denoted in milligrammes per deciliter (mg/dl).

2.7 Haematological Test

The blood samples were analysed for the following haematological parameters: Haemoglobin Determination and Packed Cell Volume (PCV) [14].

2.8 Statistical Analysis

Values are expressed as Mean of 5 replicates. SEM (Standard Error of Mean). Data were analyzed using graph pad

3 Results

3.1 Physicochemical Characteristics and Extraction Yield of Seed Oils

The avocado pear seed oil obtained after extraction was reddish brown in colour, densely liquid at room temperature, with an odour characterizing the avocado fruit. The moringa seed oil was yellowish in colour, liquid at room temperature, with an odour characterizing the moringa fruit. The pepper seed oil obtained after extraction was reddish in colour, densely liquid at room temperature, with an odour characterizing the pepper seed while the African star apple showed a light yellow colour.

Table 2 Chemical Characterization of Seed Oils

The results of the chemical characterization of seed oils are presented in the table below.

Oil Characteristics	Avocado	Pepper	Moringa	African Star	AOAC Standard
Oil yield (%)	9.47	9.85	7.7	22.7	
Iodine value (gl_2/100 g)	71.91	67.68	101.52	105.75	80-100
Peroxide value (meq/kg)	3.27	3.13	3.27	3.47	2-10
Free fatty acid (mg/kg)	4.3	2.08	5.83	7.24	≤1.30
Acid value (mgKOH/g)	8.80	4.30	11.22	14.03	≤4.00
SAP value (mg/KOH/g)	168.3	112.2	149.6	168.3	≥180

The results are means of duplicate.

Table 3 Effect of Seed Oils on Packed Cell Volume and Haemoglobin

S/N	Groups	PCV (%)	Haemoglobin (g/l)
1.	CTRL	51.60±1.08	16.70±0.33
2.	TOSD	55.00±1.70	18.60±0.59
3.	AOSD	52.00±2.50	16.98±1.09
4.	AOSHFD	55.00±0.95	18.40±0.10
5.	POSD	49.40±0.50	16.44±0.36
6.	POSHFD	53.80±`1.24	18.02±0.55
7.	MOSD	46.80±1.59	17.16±0.88
8.	MOSHFD	50.40±1.91	17.68±0.88
9.	ASASD	56.80±4.74	20.34±2.05
10.	ASAHFD	50.00±0.71	17.32±1.50

Values are expressed as mean ± SEM of five replicates.

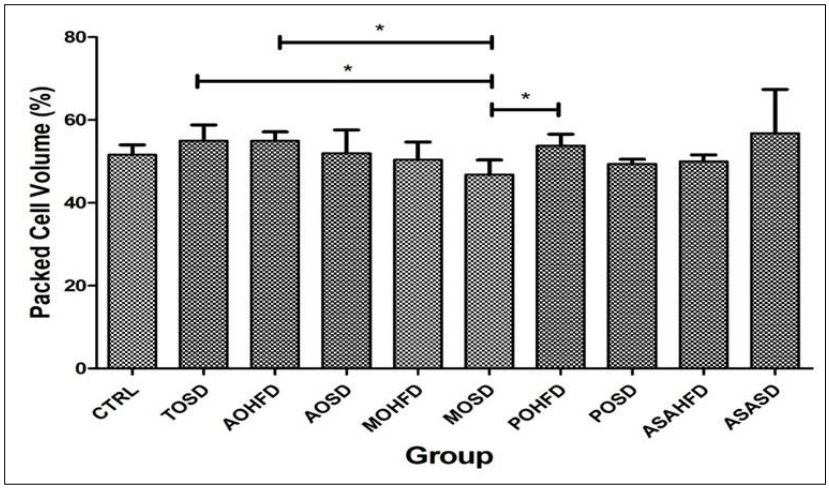

Figure 1 Packed Cell Volume (%) in treatment animal

There is a significant difference in PCV %: in the animal fed with standard diet plus Turkey oil when compared with those fed with standard diet plus Moringa oil; in the animal fed with high fat diet plus Avocado oil when compared with those fed with standard diet plus Moringa oil; and in the animal fed with standard diet plus Moringa oil when compared with those fed with high fat diet plus Pepper oil.

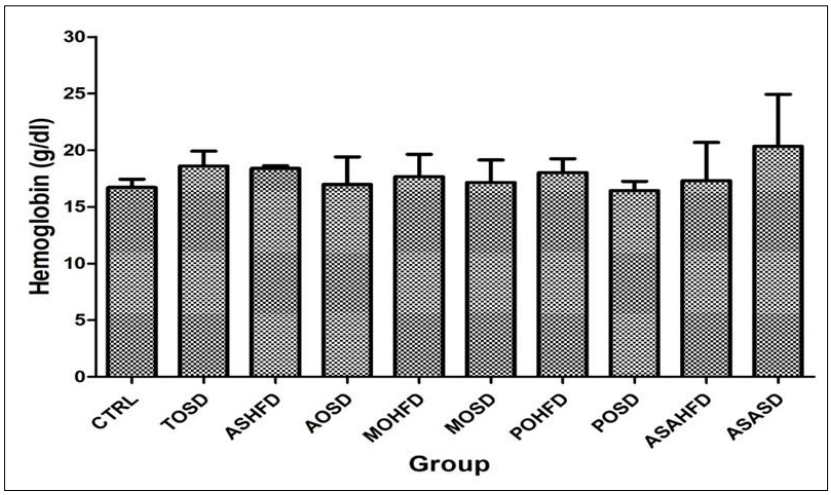

Figure 2 Hemoglobin (g/dl) in treatment animal

There is no difference in the concentration of hemoglobin in the treated animal when compared among themselves and control.

Table 4 Effect of Seed Oils on Blood Lipid Profile

S/N	Groups	TC (mmol/l)	TG (mmol/l)	HDL (mmol/l))	LDL (mmol/l)
1.	CTRL	1.70±0.29	0.42±0.11	0.40±0.10	1.20±0.18
2.	TOSD	1.80±0.23	2.78±0.95	1.12±0.15	1.46±0.27
3.	AOSHFD	1.48±0.86	2.72±0.85	0.72±0.10	0.70±0.17
4.	AOSD	1.30±0.13	4.62±0.99	1.72±0.09	1.56±0.74
5.	MOHFD	1.42±0.18	4.46±2.32	0.86±0.34	1.76±0.79
6.	MOSD	1.20±0.07	1.98±0.40	0.52±0.10	1.40±0.71
7.	POHFD	1.20±0.07	0.94±0.27	0.42±0.15	0.30±0.07
8.	POSD	1.54±0.21	0.50±0.16	0.98±0.12	0.40±0.16
9.	ASAHFD	1.90±0.31	0.56±0.10	0.96±0.21	1.26±0.52
10.	ASASD	1.50±0.23	0.34±0.09	1.00±020	1.10±0.23

Values are expressed as mean ± SEM of five replicates.

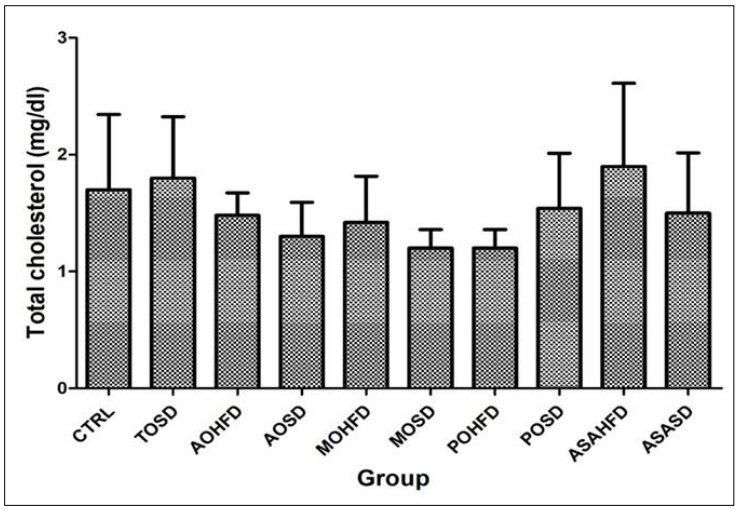

Figure 3 Total Cholesterol (mg/dl) in treatment animal

There is no difference in the level of Total Cholesterol in the treated animal when compared among themselves and control.

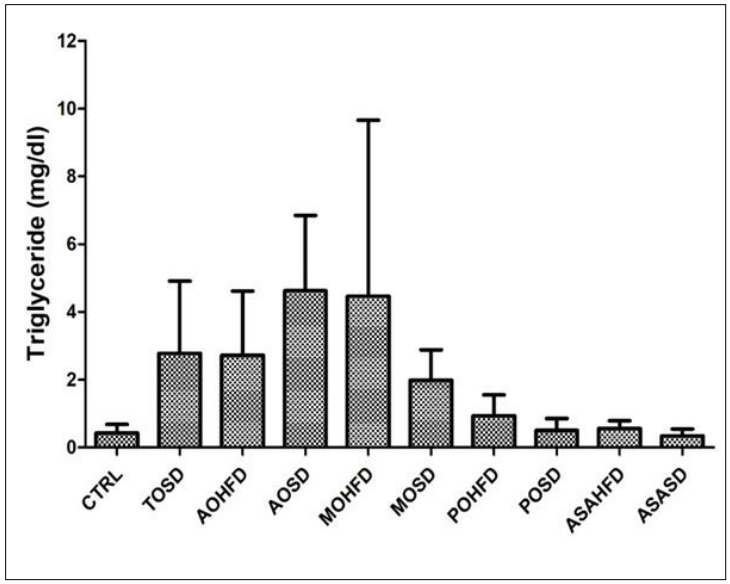

Figure 4 Triglyceride (mg/dl) in treatment animal

There is no difference in the level of Triglyceride in the treated animal when compared among themselves and control.

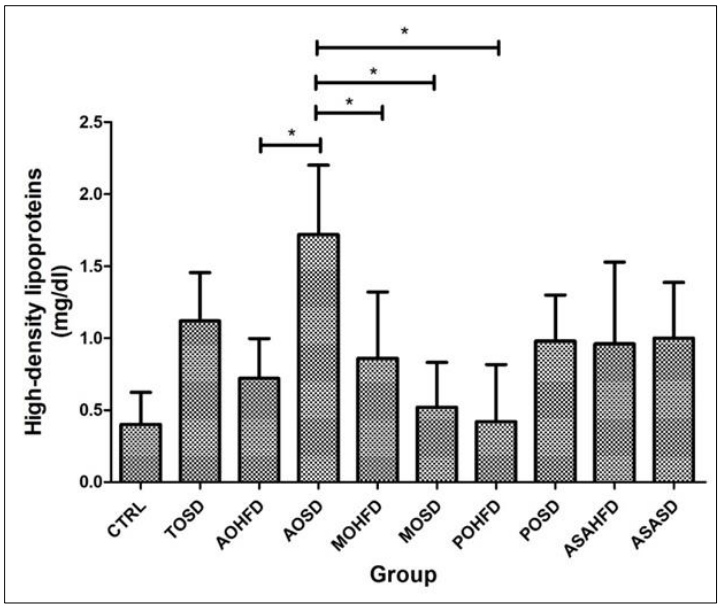

Figure 5 High-density lipoprotein (mg/dl) in treatment animal

There is a significant difference in the level of High-density lipoprotein: in the animal fed with standard diet plus Avocado oil when compared with the animal fed high fat diet plus Avocado oil; in the animal fed with standard diet plus Avocado oil when compared with the animal fed high fat diet plus Moringa oil; in the animal fed with standard diet plus Avocado oil when compared with the animal fed standard diet plus Moringa oil; and in the animal fed with standard diet plus Avocado oil when compared with the animal fed high fat diet plus Pepper oil.

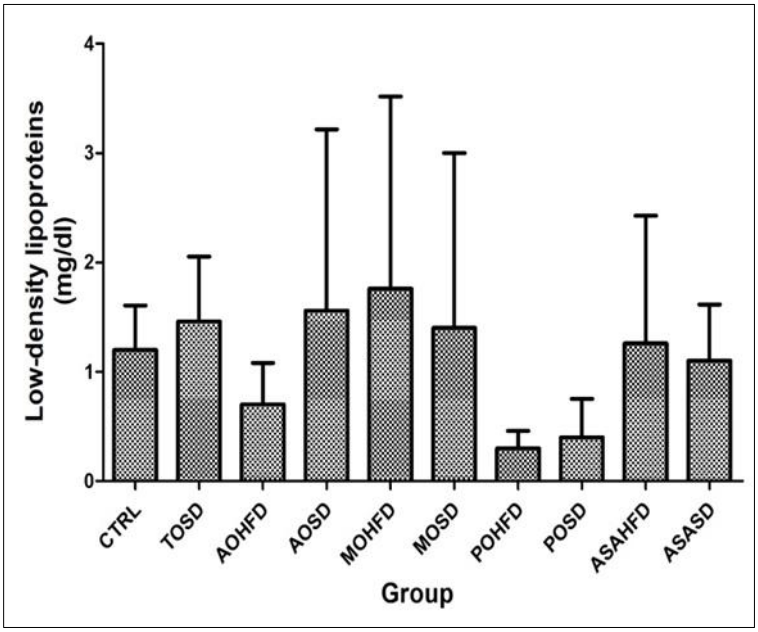

Figure 6 Low-density lipoprotein (mg/dl) in treatment animal

There is no difference in the level of Low-density lipoprotein (mg/dl) in the treated animal when compared among themselves and control.

4 Discussion

Most fruits are considered to be abundant in nutrients like vitamins and carbs, but just a few are good providers of lipids

From this study, an oil fraction with a saponification value of at least 180 mg KOH/g has been documented to have low molecular weight fatty acids [12]. The saponification values of oil derived from moringa seeds, avocado seeds, pepper, and African star seeds are 149.6 mgKOH/g, 168.3 mgKOH/g, 112.2 mgKOH/g, and 168.3 mgKOH/g, respectively. The saponification value serves as a quantitative measure of the extent of oxidation that occurs during storage, as well as an indicator of the degradation of the oils. An elevation in the saponification value of oil leads to an augmentation in the volatility of the oils. The presence of smaller molecular weight components in 1 g of the oil boosts its quality by increasing the energy yield upon combustion [14]. The poor saponification value suggests that the oil may not be appropriate for soap production, oil-based ice cream, and shampoos. Pearson [14] observed that oils with high saponification values had a significant amount of lesser fatty acids. Thus, the oils with poor saponification values, as shown in the review, contain a significant amount of higher fatty acids and can be classified as inedible oils.

The peroxide value (PV) is the predominant biomarker of lipid oxidation. The peroxide values of moringa seed, pepper seed, avocado seed, and African star apple seed are 3.27 meqKOH/g, 3.13meqKOH/g, 3.28 meqKOH/g, and 3.47 meqKOH/g, respectively. All of these values indicate a low peroxide content. The values reported by AOAC [13] for all these fell within the standard range of 2-10 meqKOH/g. This suggests that the oils may exhibit greater resistance to oxidative destruction. Free fatty acid refers to the weight percentage of a certain fatty acid, such as oleic acid. Elevated levels of unbound fatty acids are unfavourable in raw vegetable oils due to their association with significant losses of the neutral oil throughout the refining process. Free fatty acids in crude fat serve as an indicator of the oil that will be lost during refining processes aimed at eliminating fatty acids. The Moringa seed, pepper seed, avocado seed, and African star apple seed have low levels of free fatty acids, with content measured at 5.83 mgKOH/g, 2.07mgKOH/g, 4.32 mgKOH/g, and 7.24 mgKOH/g, respectively. All of these readings exceed the acceptable limit of < 1.3 mgKOH/g, as stated in the AOAC study from 1990. The iodine value is a quantitative measure of the characteristics of unsaturated organic compounds [14]. It denotes the level of reactivity exhibited by a double bond. The iodine value of avocado seed is 71.91gI2/100g, while the iodine value of pepper seed is 67.68gI2/100g. Both values are below the standard range of 80-100gI2/100g, as published by AOAC [12]. These oils contain a low level of unsaturation and are categorised as non-drying oils. Moringa and African star apple seed have iodine values of 101.52 gI2/100g and 105.75 gI2/100g, respectively. According to Atasie *et al.* [16], the oil samples show a high level of unsaturation and are classed as drying oil. A notable disparity is evident in the iodine value of all the seed samples ($p<0.05$).

There is a significant difference in PCV %: in the animal fed with standard diet plus Turkey oil when compared with those fed with standard diet plus Moringa oil; in the animal fed with high fat diet plus Avocado oil when compared with those fed with standard diet plus Moringa oil; and in the animal fed with standard diet plus Moringa oil when compared with those fed with high fat diet plus Pepper oil. There is no difference in the concentration of hemoglobin in the treated animal when compared among themselves and control.

There is no significant difference in the level of Low-density lipoprotein, Total cholesterol, Triglyceride in the treated animal when compared among themselves and control.

5 Conclusion

This study reveals an appreciable physico chemical characteristics for the seeds oils with a remarkable PCV and Hb content. The oils were found to have a moderate lipid profile content. Conclusively, the rich essential fatty acid reported in the seed oils has hypolipidemic effect.

Compliance with ethical standards

Acknowledgments

Mr Oseni Usman of the Department of Science Laboratory Technology, Gateway Polytechnic, Saapade is highly appreciated for his immense contributions.

Disclosure of conflict of interest

No conflict of interest associated with this work.

Statement of ethical approval

The studies were conducted in accordance with the regulations and ethical permission of the Experimental Animal Welfare and Ethics Committee of the institution. Thus, the study was carried out after obtaining ethics clearance from the Research and Ethics Committee (REC) with clearance number OOUREC/23/084.

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Authors short Biography

	**Fatai Adebayo Kareem** is a versatile and renowned Biochemist. He had his BSc and MSc in Biochemistry from the Federal University of Agriculture and Olabisi Onabanjo University respectively. He is a dedicated Senior lecturer who has spent several years in the teaching of Biochemistry.
	**Dr. Yesiru Adeyemi Kareem** is a fellow of the West African College of Physicians. He had his Medical Residency Training and is presently a Consultant Psychiatrist at Neuropsychiatric Hospital, Aro Abeokuta. He is an ambitious multitasking Psychiatrist, early career researcher, thoroughbred clinician, and an astute mobiliser with research interests in Old Age, Forensic, Consultation-Liaison, and Academic Psychiatry. He has over 60 Academic seminar presentations, presented papers in over 15 Scientific conferences, and collaborated in several studies with about 20 Publications in local and International Journals.
	**Mutiu Adewunmi Alabi** (PhD) is an associate professor in the Department of Biochemistry, Kwara State University, Malete. He graduated from the Federal University of Agriculture for his BSc and proceeded to Olabisi Onabanjo University for his MSc and PhD. He was trained at ITSI Bioscience LLC, Johnstown, United States for his PhD fellow and Manipal Academy of Higher Education, Manipal, India for his Postdoctoral fellow.
	**Adeola Oladayo Osifeso** had her Bachelor of Science and Master of Science degrees in Biochemistry from Obafemi Awolowo University, Ile Ife, Osun State and Federal University of Agriculture, Abeokuta, Ogun State.
	**Olasunkanmi Adedoyin Adetutu** attended Ladoke Akintola University Ogbomoso, Oyo state where she obtained B.Tech (Hons) and M.Tech in Biochemistry She is a member of Science Association of Nigeria and presently a lecturer II at Federal +Polytechnic Ilaro, Ogun State.