ISSN No:-2456-2165

Comparative Effects of Some Common Vegetable Oils on Lipid Profile and Liver Function in Male Wistar Rats

Umezulike Anulika Jacinta, Maduka Stephen Ozoemena, Njoku-Oji Nancy Njideka, OkonudoPeter O, Nwaefulu Kester Eluemunor, Mmaju ChidimmaIfeyinwa, Chukwukaeme Chidimma Winifred, Ezeani Emmanuel Ifeakandu.
Department of Physiology, Faculty of Basic Medical Sciences, Nnamdi Azikiwe University,
Awka, Nnewi Campus, Anambra State, Nigeria

Abstract:- Vegetable oils, a major source of lipids form essential part of the human diet, It is second source of energy after carbohydrates. Dietary oils high in unsaturated fatty acids have been correlated with the incidence of cardiovascular disease.

This study investigated the comparative effects of some common vegetable oils on lipid profile and liver function in male wistar rats. 25 male wistar rats were divided into 5 groups (n=5). Group 1 served as control and were given rats feed and water only, Groups 2, 3, 4 and 5 served as the test groups and were administered 2ml/kg/day of palm, coconut, olive and soya oils respectively. All animals had feed and water *ad libitum* for 8 weeks. Body weight, relative liver weight, lipid profile, serum liver enzymes and histopathological examination of the liver tissue were compared among different groups. Results revealed a significantly ($p < 0.05$) elevated body weight in all the groups when initial and final weight was compared. Group 4 significantly elevated relative liver weight and triglyceride level, while group 3 significantly decreased total cholesterol and LDL-C. However, HDL-C and liver enzymeswere insignificantly changed in all treated groups compared to control and with one another. Histopathological examination revealed a moderate impairment of hepatic tissues in all the oil treated groups compared to control. This study concludes that consumption of these vegetable oils for longer duration increase body weight, and impairs liver function. Coconut oil however, improved lipid profile while olive oil showed a therogenic property than other oils.

Keywords:- *vegetable oil, lipid profile, coconut, palm, liver.*

I. INTRODUCTION

Cholesterol is an essential part of cells in the human body which is required for the formation of new and repair of older cells (Abuoa*et al*.,2009). However, increased levels of cholesterol or triglycerides are most often attributed to disorders of lipid metabolism (Edem and Akpanabiatu.,2006). These lipids may also be increased by some other disease conditions such as diabetes mellitus (DM), hypothyroidism, kidney and liver disease (Oladapo*et al*.,2017). In addition, diets high in fats, which is the basis of this study also affects serum cholesterol and TG concentrations (Dauqan*et al*.,2011).

Edible oils mainly composed of triacylglycerol and cholesterol form an essential part of the human diet, apart from adding desirable taste and flavours to human diet, they are also rich sources of energy for the body's metabolic processes second tocarbohydrate (Edem.,2009).They provide the body with essential fatty acids and fat soluble vitamins (Bester *et al*., 2010; Mohammed and Luka., 2013). However, fats and oils that contain high levels of saturated fatty acids increase blood cholesterol levelsand hence the risk of cardiovascular disease (Imafidon and Okunrobo.,2012). Although oils rich in unsaturated fatty acids are particularly susceptible tooxidation, intake of food containing oxidized lipidincrease the concentration of secondaryperoxidation products in the liver (Nna*et al*.,2014).

Palm oilis extracted from the fruit of oil palm (*Elaesisguineensis*) is most commonly used cooking oil in the world (Ebong*et al* .,1999).

It is rich in mono unsaturated fatty acids, antioxidant and vitamins (Dauqan*et al*.,2011). Some of its beneficial effect include reduction in oxidative stress, arterial thrombosis and arterial blood pressure (Chandrasekharan*etal*.,2000). Palm oil is a good source of carotenoids mainly beta-carotene that gives it the reddish colour which can make up 0.08% of the crude oil (Mukherjee & Mitra .,2009).

Coconut oil extracted from coconut (*Cocosnucifera*) is a colourless often pale brownish yellow oil,rich in saturated fats containing medium chain fatty acid mostly lauric, mystric and palmitic acid(Arlee*et al*.,2013). Coconut oil have been shown to inhibit lipo-protein oxidation, and has hypolipidemic effects. It has also been proven to be beneficial in hair and skincare, weight loss, increased immunity and dental care(Sankararaman&Sferra.,2018). These health benefits have been attributed to the presence of lauric acid, capric acid and caprylicacid, and the possession of antimicrobial, antioxidant, antifungal and antibacterial properties (Marina.,2009).

Soybean oil is derived from the seed of glycine max.It contains approximately 60% of polyunsaturated fatty acids (PUFA), 24% MUFA, and 16% of saturated fatty acids (Mensink*et al*.,2003, Lawrence.,2013). it has been reported to improve the blood lipid profile status due to the high level of PUFA (Warner.,2005).

Olive oil is a yellow-coloured oil, extracted from olive fruits produced from olive tree (Oleaeuropaea) widely

ISSN No:-2456-2165

cultivated in Mediterranean basin and some Asian parts (Owen *et al.*,2000). It consists of mainly monounsaturated fatty acid (MUFA) including oleic acid and increased availability of antioxidants which increases the stability of olive oil (Alhazza, 2007). Some of these antioxidant micronutrients are polyphenols and squalene that have the ability to scavenge single oxygen radicals, also it's fatty acid composition of may play a role in improving the serum lipid profile (Bester *et al.*,2010: Haggag*et al*.,2014).

Dietary oils remain the major source of lipid in diets. However, due to the fact that elevated serum concentration of cholesterol is a major risk factor for coronary artery disease, it became necessary to investigate the effect of these oils on lipid profile and liver function in male wistar rats.

II. MATERIALS AND METHODS

A. *Purchase of vegetable Oils*

Vegetable oils used for this study including palm oil, coconut oil, olive oil and soybean oil were purchased from Nkwo market in Nnewi, Anambra State, Nigeria.

B. *Experimental animals*

Twenty five male wistar rats weighing 160-190grams were used for this study. The animals were purchased from the Department of Physiology Animal House, College of Health Sciences, Nnamdi Azikiwe University, Nnewi campus, Nigeria. Ethical approval was obtained for the progress of this study from the Institutional Animal Ethics Committee (NAU/AREC/TEMP/2021/00019). Animals handling and treatment conform to the guidelines of the Nnamdi Azikiwe University Animal Research Ethics Committee (NAU-AREC) for laboratory animal care and use.

C. *Experimental design*

Animals were randomly grouped into 5 (n = 5), after which they were allowed to acclimatize for 2 weeks. All animals were allowed access to food and water *ad libitum* and 12 hours light/dark cycle.

Group 1 served as control and were served food and water only, group 2 palm oil - treated group were administered 2ml/kg/day of palm oil, group 3 coconut oil - treated group were served 2ml/kg/day coconut oil, group 4 olive oil -treated group were served 2ml/kg/day olive oil, group 5 soybean oil - treated group were served 2ml/kg/day soybean oil. All the oils were administered through oralgavage for 8 weeks. Initial and final measurements of bodyweight were recorded.

D. *Collection of Blood and Liver Samples*

At the end of 8 weeks, The rats were subjected to an overnight fast after which they were anesthesizedusing chloroform vapour (soaked in cotton wool) and blood collected by cardiac puncture. The samples were collected (using 5mls syringe) into plain capped containers without anticoagulant. The liver were harvested, weighed and placed in 10% formalin for histopathological analysis.

E. *Serum analysis*

The serum total cholesterol TC level was estimated by enzymatic colorimetric method described by Naito, (1984). The serum high density lipoprotein cholesterol HDL-C level was estimated by precipitation and CHOD-POD enzymatic colorimetric reaction, according to the method described by Grove, (1979) and Naito, (1984). The serum triglycerides TG level was estimated by GPO-POD enzymatic colorimetric reaction, according to the method described by Fossati*et al*., (1982).

The Low density lipoprotein cholesterol LDL-C was estimated, according to the method described by Friedewald*et al.,* (1972).

LDL-cholesterol (mmol/L) = Total Cholesterol – (HDL-C + VLDL-C)
VLDL-cholesterol = Triglyceride/2.2.

Serum Aspartate amino transferase AST and Alanine aminotransferase ALT were determined using the spectrophotometric method of Bergermeyer*et al*., (1978).While Alkaline phosphatase ALP was assayed using the spectrophotometric method of Schlebusch*et al*., (1974).

F. *Histological examination*

Histopathological examination was done using the method of Humason (1962). The liver tissues were fixed in 10% neutral formalin, dehydrated, embedded in paraffin, sectioned and stained with hematoxylinand eosin.

G. *Statistical Analysis*

All data are represented as mean ± SEM. One – way analysis of variance (ANOVA) was used for analysis of Differences between groups, followed by least square difference (LSD) post hoc, for multiple comparison, student t-test was used for body weight comparison within groups using SPSS software version 20. Significant difference was employed at $P < 0.05$.

III. RESULTS

A. *Comparative Effect of the various vegetable oils on body weight of the rats*

There was a significant(p>0.05) increase in weight gain between the initial and final body weight in all the vegetable oil treated groups and the control.

S/No	Treatment	Initial weight(gm)	Final weight(gm)
1	Control	176.00±6.20^{a}	220.00±11.9^{b}
2	Palm oil	188.40±3.51^{a}	228.68±6.52^{b}
3	Coconut oil	192.00±6.33^{a}	227.70±4.65^{b}
4	Olive oil	187.20±5.85^{a}	224.22±2.89^{b}
5	Soya oil	188.20±4.03^{a}	222.06±4.93^{b}

Table 1: Effect of the various vegetable oils on initial and final body weights

ISSN No:-2456-2165

Values are mean±SEM, n=5. Mean on the same row with different

letter superscript are statistically significant(P<0.05). Data analysis was done using paired t-test

B. Comparative Effect of the various vegetable oils on relative liver weight of the rats

The relative Liver weight wasn't significant when the vegetable oil treated groups were compared to the control. However group 4 was significantly increased(p<0.05) when compared with group 2 and 3.

S/No	Treatment	Relative liver weight
1	Control	0.031±0.0019[ab]
2	Palm oil	0.027±0.0021[a]
3	Coconut oil	0.026±0.0032[a]
4	Olive oil	0.036±0.0026[b]
5	Soya oil	0.028±0.0035[ab]

Table 2: Effect of the various vegetable oils on relative liver weight

Values are mean±SEM, n=5. Mean on the same column with different

letter superscript are statistically significant(P<0.05)

C. Comparative Effect of the various vegetable oils on lipid profile of the rats

On Total cholesterol there was no significant difference in groups 2, 4 and 5. However, group 3 significantly reduced when compared with the control (P>0.05). No significant difference was recorded when TC level was compared among the oil treated groups.

On TG levels, no significant difference was recorded in all the various oils groups when compared with the control (P<0.05). However, group 4 showed a significant increase when compared to group 3.

No significant difference was recorded on HDL-C in the various oils treated groups (groups 2-5) when compared with control (P>0.05). Also, when compared among the different oils treated groups.

On LDL-C there was no significant difference in groups 2, 4 and 5 while group 3 had a significant reduction when compared with the control (P<0.05).

S/No	Treatment	TC (mmol/L)	TG (mmol/L)	HDL-C (mmol/L)	LDL-C (mmol/L)
1	Control	3.67±0.33[a]	0.78±0.05[abc]	0.82±0.06	2.50±0.35[a]
2	Palm oil	2.96±0.20[ab]	0.71±0.04[abc]	0.92±0.09	1.72±0.10[ab]
3	Coconut oil	2.73±0.24[b]	0.67±0.07[b]	0.86±0.07	1.55±0.28[b]
4	Olive oil	2.89±0.28[ab]	0.85±0.06[c]	0.81±0.05	1.69±0.27[ab]
5	Soya oil	3.18±0.33[ab]	0.68±0.07[abc]	0.88±0.07	1.95±0.38[ab]

Table 3: Comparative Effects of Various vegetable oils on Lipid Profile of the rats

Values are mean±SEM, n=5. Mean on the same column with different letter superscript are statistically significant (P<0.05)

D. Comparative Effect of the various vegetable oils on liver enzymes of the rats

On liver enzymes there was no significant difference in serum ALT, ALP and AST level in all the various oil treated groups (groups 2, 3, 4, and 5) when compared with the control (P>0.05) and among the various oil treated groups.

S/No	Treatment	ALT	ALP	AST
1	Control	7.50±1.6	54.25±5.5	11.00±1.0
2	Palm oil	6.00±2.1	51.33±6.4	8.67±1.5
3	Coconut oil	9.25±1.6	59.25±9.8	9.00±1.5
4	Olive oil	8.25±0.6	63.25±11.2	10.25±0.8
5	Soya oil	7.75±2.1	67.75±9.6	9.25±2.1

Table 4: Effects of Various vegetable oils on Liver enzymes

Values are expressed as mean±SEM, n=5.

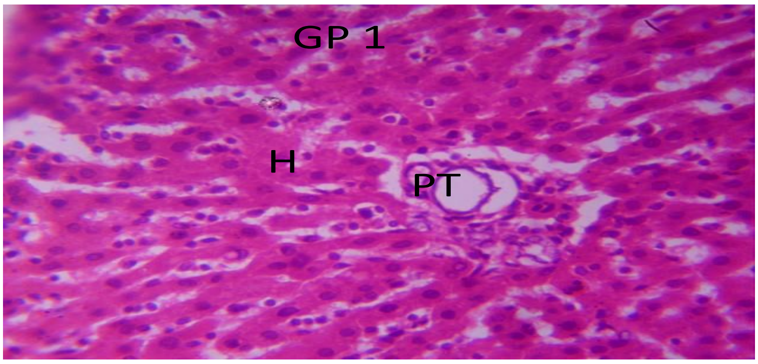

Plate 1: Photomicrographic representation of liver of group 1 (control) showing a normal hepatic architecture with Portal triad(PT) and hepatocyte(H) (x400) (H&E).

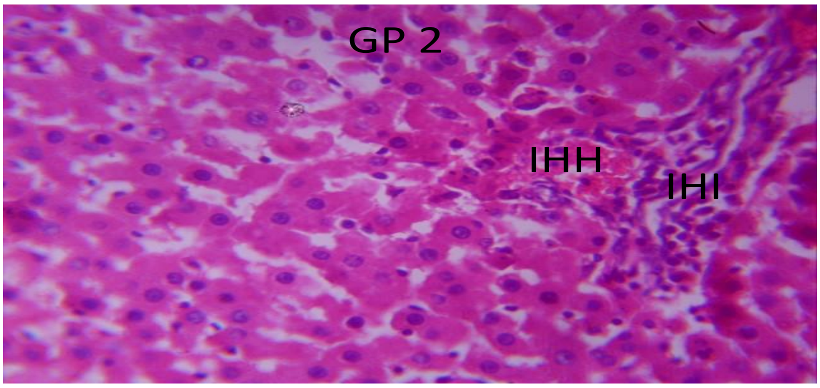

Plate 2: Photomicrographic representation of the liver of group 2 (palm oil administered)rats showing a moderate intra hepatic inflammation(IHI) and mild intra hepatic hemorrhage(IHH) (X400 H & E).

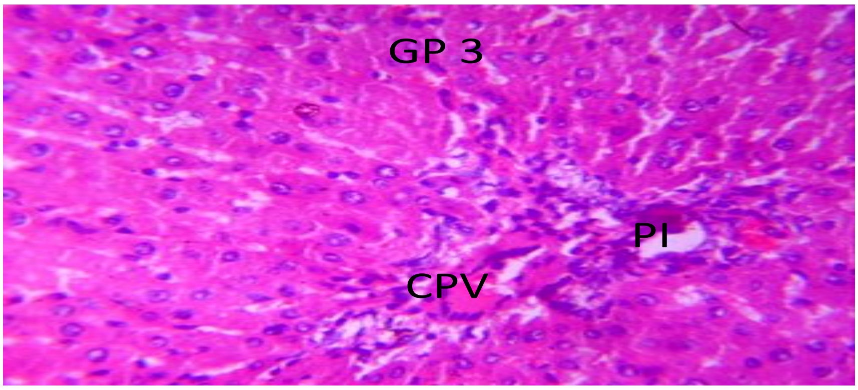

Plate 3: Photomicrographic representation of the liver of group 3 (coconut oil administered)rats showing a moderate Portal inflammation(PI) and congestion of the portal vein(CPV) (X400 H & E).

ISSN No:-2456-2165

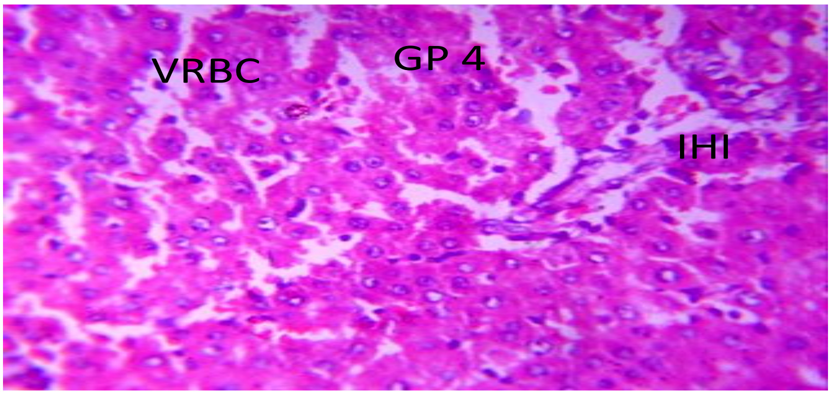

Plate 4: Photomicrographic representation of the liver of group 4(olive oil administered)rats showing a moderate vacoulation, intra hepatic inflammation(IHI) and mild extra vasation of the red blood cells(VRBC) (X400 H & E).

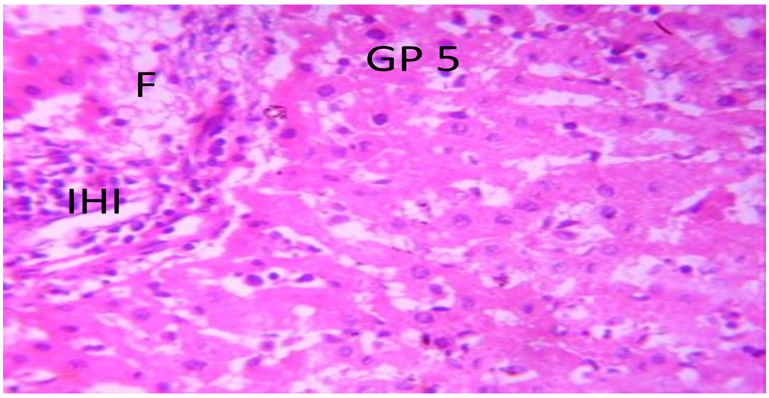

Plate 5: Photomicrographic representation of the liver of group 5 (soyabean oil administered)rats showing a moderate Intra hepatic inflammation(PI) and fibrosis(F) (X400 H & E).

IV. DISCUSSION

The amount and type of fat contained in a diet has long been linked with the risk of Coronary Heart Disease (CHD),with saturated fats adversely affecting health while polyunsaturated fats have protective factors. (Nna*et al.*,2014). The Wister rats used were strictly males since it was reported that female sex hormones (17-βestradiol) has a lowering effect on the plasma cholesterol concentration (Mohammad & Luka.2013).

The present study investigated the comparative effects of some common vegetable oils on lipid profile and liver function of male wistar rats.

The present study revealed a significant increase between the initial and final body weight of all the groups, this is in consistent with the report of Oladapo*et al.*,(2017) who recorded a significant increase in soya and olive oil fed rats, Dauqan*et al.*, (2011) earlier reported that red palm olein and coconut oil caused an increase in body weight of the rats, Stroher*et al.*, (2020) also recently reported an increase in bodyweight of rats fed with coconut oil. The increase in body weight observed in this study may be due to the palatability of the various oils leading to increased food intake and weight gain (Dauqan*et al.*, 2011). There was no significant difference in the relative liver weight in the various oil treated groups when compared with the control. However there was a significant increase in group 4 (olive oil treated group) when compared with group 2 (palm oil treated group) and group 3(coconut oil treated group).This tallies with the report of Imafidon and Okunrobo.,(2012) who reported an increase in liver weight of diabetic rats treated with olive oil. According to Orisakwe*et al.*,(2003) an increase or decrease in absolute or relative organ weight after administration of a chemical or substance is an indication of the toxic effect of the substance.

Liver enzymes are a useful tool in determination of liver function. They are usually increased in acute hepatoxicity but tend to decrease with prolonged intoxication due to hepatic damage (Imafidon and Okunrobo., 2012).

Changes in serum enzymes levels are often early determinant of tissue damage either by toxicant or in disease conditions. Serum alkaline phosphate, alanine amino transferase and as partate aminotransferase are liver biochemical markers. Therefore, increases in ALT activity are always due to hepatocellular damage and it is usually accompanied by AST activity (Sekar*et al*.,1990).

The present study revealed that the mean ALT, AST and ALP in all the various oil treated groups were similar to one another although ALP was increased compared to the control. Previous reports by Imafidon and Okunrobo., (2012) showed an increase in ALT in rats treated with palm oil and increase in AST in rats treated with coconut oil while Mohammed and Luka., (2013) reported a decrease in ALP and AST in coconut oil fed rats, also Ramadan *et al*., (2019) reported a decrease in AST in coconut oil fed rats. However, increase or decrease in serum liver enzymes can be attributed to level of liver toxicity (Imafidon and Okunrobo.,2012). The insignificant difference observed in this study may be due to the long term treatment.

Atherosclerosis is characterized by elevated TG and TC which are risk factors that cause deposition of fats on the inner lining of blood vessels, resulting in endothelial dysfunction, chronic vascular inflammation, dyslipidemia, plaque formation and eventually cause thickening and narrowing of the arteries (Peng., 2013). The present study revealed no significant difference in Total cholesterol in groups 2, 4 and 5 while group 3 had a significant decrease when compared with the control, this tallies with reports of Mohammed and Luka.,(2013) and Nna*et al*., (2014)who reported a significant decrease in coconut oil fed rats. The ability of coconut oil to decrease TC seen in this study may be attributed it's antioxidants content like polyphenols such as ferulic acid, vanillic acid and quercetin (Narayanankutty*et al*.,2018; Santararaman and Sferra.,2018; Arlee *et al*.,2013). The present study revealed no significant difference in triglyceride level in all the oil treated groups when compared with the control. However, group 4 caused a significant increase in triglycerides when compared with group 2 and 3, this is in contrast with reports of Oladapo *et al*., (2017) who previously reported that olive oil had lowest triglycerides level in diabetic treated rats than red palm olein and soya oil, Amir *et al*.,(2015)earlier reported a decrease in TG in olive oil treated hyperlipidemic subjects. The increase in triglycerides observed in this study may due to the fatty acid contents of the olive oil which suggest its atherogenic property since increased serum triglyceride is a risk factor for development of atherosclerosis.

The results from the HDL-C in all the oil treated groups (group 2-5)when compared with the control as well as when compared among the various oil treated groups were similarly higher except olive oil group which was reduced. High concentrations of HDL indicate a cardio protective effect. The mechanism that triggers this effect may be different depending on the characteristics of each oil (Jaarin et al.,2015). Based on our findings, however, the vegetable improved HDL values except olive oil.

The present study revealed no significant difference in the LDL-C level in groups 2,4 and 5 while group 3 caused a significant decrease in LDL-C when compared to the control, this is in consistent with the report of Mohammed and Luka.,(2013) and Nna*et al*.,(2014) which was attributed to its content as reported by Prasad *et al*., (2016) coconut oil contains medium chain fatty acids which do not have cardiovascular risk and also it's polyphenols content (Narayanankutty*et al*.,2018). While Dauqan*et al*.,(2011); Stroher*et al*.,(2020) reported that coconut oil had a significant increase in LDL-C and is associated with risk of cardiovascular disease due to its high content of saturated fatty acids.

This study conclude that consumption of these vegetable oils for longer duration increased body weight, and impairs liver function by distorting the liver architecture. Coconut oil however, improved lipid profile while olive oil proved to be moreatherogenic than other oils.

REFERENCES

[1.] Aboua Y. G. , N. Brooks, D. O. Awoniyi and S. S. Plessis (2009) "Red Palm Oil: A Natural Good Samaritan for Sperm Apo- ptosis," Medical Technology, Vol. 23(1): 8-10.

[2.] Alhazza IM.(2007) "Antioxidant and Hypolipidemic Effects of olive oil in normal and diabetic male rats. Saudi Journal of Biological sciences; 14(1): 69-74.

[3.] Amir EA, Sayed T A, Ietimad M.A. A, Ihab HN, Mohammed HFS (2015)"Lipid Profile and Liver Function Test Estimates in Saudi Females'' *International Journal of Science and Research (IJSR)* vol 5 issue 4 ,2319-7064

[4.] Arlee R, Suanphairoch S, Pakdeechanuan P (2013): "Differences in chemical components and antioxidant-related substances in virgin coconut oil from coconut hybrids and their parents. International Food Research Journal ;20:2103–2109.

[5.] Bester D, Esterhuyse A. J. , Truter E. J. And van Rooyen J. (2010) ''Cardiovascular effects of edible oils: a comparison between four popular edible oils.'' Nutrition Research Reviews 23, 334–348.

[6.] Bergmeyer HU, Scheibe P, Wahlefeld AW. (1979). "Methods for aspartate and alanine amino transferase." Clinical Chemistry. 125 :1487.

[7.] Chandrasekharan N, Sundram K, Basiron Y.(2000) "Changing nutritional and health perspectives on palm oil."Brunei International Medical Journal 2: 417-27.

[8.] Dauqan E, Abdullah A, Abdullah H S.(2011) '' Lipid peroxidation in rat liver using different vegetable oils'' The Malaysian Journal of Analytical Sciences, Vol 17 (2): 300 – 309

[9.] Ebong PE, Owu DU, Isong EU. (1999) "Influence of palm oil (Elaesisguineensis) on health" Plant Foods and HumanNutrition; 53(3): 209-22.

[10.] Edem DO, Akpanabiatu MI (2006). "Effects of palm oil-containing diets on enzyme activities of rats"Pakistan Journal of Nutrition. 5(4): 301-305.

[11.] Edem DO (2009). ''Haematological and histological alterations induced in rats by Palm Oil-Containing Diets". European Journal of Science Research 32: 405-418.

[12.] Fossati P, Prencipe L, Lorenzo P. (1982). "Serum triglycerigle determination colourimetrically with an enzyme that produces hydrogen peroxide."Clinical Chemistry. 28; 2077 – 2080.

[13.] Friedewald WT, Levy RI, Fredrickson DS. (1972) "Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge"ClinicChemistry;18:499–502.

[14.] Grove TH. (1979). "Effect of reagent pH on determination of HDL-C by precipitation with Na-phosphotungsate-magnesium."Clinical Chemistry. 25: 560.

[15.] Haggag M EY, Rafaat M E, Mohamed FR (2014) ''Impact of dietary oils and fats on lipid peroxidation in liver and blood of albino rats" Asian Pacific Journal of Tropical Biomedicine4(1): 52-58

[16.] Humason, G.L. (1962). "Animal Tissue Techniques" Freeman, W.H. and Co. Publishers, London. Pp57-61.

[17.] Imafidon K.E. and Okunrobo L.O. (2012) "Study on Biochemical Indices of Liver Function Tests of Albino Rats Supplemented with Three Sources of Vegetable Oils" Nigerian Journal of Basic and Applied Science 19 (2): 105-110

[18.] Jaarin K, Nor-Aini U, Siti-Aishah MA and Das S (2015). " Palm oil fat diet consumption and its effects on serum liver enzymes and microscopic changes in experimental Rats".Pakistan Journal of Nutrition 14: 575-580.

[19.] Lawrence GD (2013). "Dietary fats and health: dietary recommendations in the context of scientific evidence"Advance Nutrition 4(3): 294-302.

[20.] Marina AM, Che Man YB, Nazimah SAH, Amin I (2009) "Antioxidant capacity and phenolic acids of virgin coconut oil". International Journal of Food Science and Nutrition (Suppl 2):114–123.

[21.] Mensink RP, Zock PL, Kester AD, Katan MB (2003) "Effects of dietary fatty acids and carbohydrates on the ratio of serum total to HDL cholesterol and on serum lipids and apolipoproteins" A meta-analysis of 60 controlled trials. American Journal of Clinical Nutrition77(5): 1146-55

[22.] Mohammed, A1 and Luka, C.D(2013) "Effect of Coconut Oil, Coconut Water and Palm Kernel Oil on Some Biochemical Parameters in Albino Rats"IOSR Journal of Pharmacy and Biological Sciences (IO SR-JPBS) e-ISSN: 2278-3008, p-ISSN:2319-7676. Volume 6, (3), PP 56-59

[23.] Mukherjee S, Mitra A.(2009) "Health effects of palm oil" Journal of Human Ecology26(3): 197-203.

[24.] Naito HK. (1984). "High-density lipoprotein (HDL) cholesterol" Kaplan A . Clinical Chemistry. The C. V. Mosby Co St. Louis, Toronto Princeton. Pp. 437 and 1207-1213.

[25.] Nna V U., Nsima M. E, Stella C. B, Ofem E. O (2014) "Comparative Effect of Chronic Consumption of Some Edible Vegetable Oils on Lipid Profile and Some Haematological Parameters in Rats"Annals of Biological Research, 5 (7):16-21

[26.] Narayanankutty A, Palliyil DM, Kuruvilla K, RaghavamenonAC (2018) "Virgin coconut oil reverses hepatic steatosis by restoring redox homeostasis and lipid metabolism in male Wistarrats"Journal of the Science of Food Agric 98:1757–1764.

[27.] Oladapo OO, Ojora KA, Quadri OM, Ajani RS.(2017) "Lipidemic effects of common edible oils and risk of atherosclerosis in diabetic wistarrats" ARYA Atherosclerosis; 13(1): 14-9.

[28.] Orisakwe, O.E., Hussaini, D.C. and Afonne, O.J. (2003). Testicular effects of sub-chronic administration of Hibiscus sabdariffa calyx aqueous extract in rats. Reproductive Toxicology, 18:295-298.

[29.] Owen RW, Mier W, Giacosa A, Hull WE, Spiegelhalder B, Bartsch H. (2000) Phenolic compounds and squalene in olive oils: the concentration and antioxidant potential of total phenols, simple phenols, secoiridoids, lignansand squalene. Food ChemToxicol 38(8): 647-59.

[30.] Peng, Ye, Irwin, K. C., & Barry, H. (2013). "High-fat diets and pathology in the Guinea pig". Atherosclerosis or liver damage? Biochimica et BiophysicaActa, 1832, 355–364.

[31.] Prasad RM, Urban D'Souza, Shankar Bhat, Kumar M R Bhat (2016) "Effect of vegetable oils on the lipid profile and antioxidant status in Wistar rats" :A comparative study, Journal of International Medicine and Dentistry 3(2): 109-114

[32.] Ramadan M T,M.,Ali,M, Rahman,S,Rahman, M.H (2019) "A study on biochemical properties and effects of different vegetable oils on blood indices in wistarrats"Journal of Pharmaceutical Research International 25(5)1-10

[33.] Sankararaman S, Sferra TJ (2018) "Are we going nuts on coconut oil?" Current Nutrition Report ;7:107–115.

[34.] Schlebusch H, Rick W, Lang H, Knedal M. (1974). "Standards in the activities of clinically important enzymes". Deutsche MedizinischeWochenschrift. 99: 765-766.

[35.] Sekar N, Willam S, Balasubramanniyam N, Karamajam P,(1990) ''Optimization of sodium orthoranadate in streptomycin induced diabetic rats, Journal of Biological science 16:67-75

[36.] Stro¨her DJ , Micaela FO ,Patrı´ciaMO,Bruna C P,, Ma´rciaDP,Cattelan, ER, KalyneB,Paulo B D, Jacqueline da Costa E P, and VanusaM (2020) "Virgin Coconut Oil Associated with High-Fat Diet Induces Metabolic Dysfunctions, Adipose Inflammation, and Hepatic Lipid Accumulation"Journal of Medical Food 23 (7) 689–698

[37.] Warner K. (2005) "Effects on the flavor and oxidative stability of stripped soybean and sunflower oils with added pure tocopherols". Journal of Agric Food Chemistry 53(26): 9906-10.