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COMPARATIVE EFFECTS OF PALM KERNEL OIL, OLIVE OIL, CRUDE OIL AND HONEY ON LIVER FUNCTION OF MALE ALBINO RATS

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ABSTRACT

Background: Comparative effects of palm kernel oil, olive oil, crude oil and honey on liver function of male albino rats was evaluated in this study because of their wide use in traditional medicine. **Methods**: A total of 35 healthy male albino rats were used in this study. The rats were randomly placed into 5 groups with 6 rats in each group. The rats were administered the chemical substances for 21 days before being sacrificed and their blood samples and livers collected for biochemical and histological analysis respectively. Results: ALT reduced nonsignificantly (p>0.05) in groups 2, 4 and 5, but increased non-significantly in group 3 compared to the control. ALP increased non-significantly (p>0.05) in groups 2 and 5, but increased significantly (p<0.05) in groups 3 and 4 compared to the control. AST reduced non-significantly (p>0.05) in group 2, but increased significantly (p<0.05) in groups 3 and 4, and non-significantly (p>0.05) in group 5 compared to the control. Total proteins increased nonsignificantly (p>0.05) in groups 2 and 3, but increased significantly (p<0.05) in groups 4 and 5 compared to the control. Albumin and globulin increased significantly (p<0.05) in all the test groups compared to the control. Bilirubin increased non-significantly (p>0.05) in groups 2, 3 and 5, but increased significantly (p<0.05) in group 4 compared to the control. Photomicrographs of histoarchitectural state of the liver tissues showed some mild alterations in some part of the tissues of the test animals administered olive oil and crude oil when compared with the control. Conclusion: Comparative evaluation of long-term administration of palm kernel oil, clive oil, crude oil and honey as in this study showed that olive oil and crude oil may negatively alter liver function than palm kernel oil and honey. Palm kernel oil and honey may support certain liver functions.

KEYWORDS: Crude oil, honey, olive oil, palm kernel oil, liver function.

BACKGROUND

Oil palm (*Elaeis guineensis*) is mostly grown in tropical regions such as Africa, South East Asia, Latin America and South pacific. [1] It produces two different kinds of oil which are palm oil derived from the fibrous mesocarp of the fruit and palm kernel oil which is obtained from the white kernel flesh. [2] In Nigeria, the process of extraction is done locally by de-shelling the palm kernel using crusher in the inform of moderately heavy object to separate the kernel which is heated in a pot until it turned black, while the oil melts out of the nut into the pot in the process after which the oil is decanted into a clean container for storage. Palm kernel oil can be used alone or in combination with other oil in the production of cocoa-butter substances, confectionary fats, biscuit doughs, filling cream, cake icing, and margarine. [3] Similarly, palm kernel oil can be used in the manufacturing of several non-edible products like candle, cosmetics/cream, soap/detergent, grease/lubricants for machines, plastic products, use in drilling mud for petroleum industry, printing inks and pharmaceuticals products. In traditional medicinal

practice, it is used as an antidote for poison and for wound healing.

Olive tree (*Olea europaea* L.) originated from the upper Mesopotamia and South Front Asia, including a part of the south-eastern Anatolia region of Turkey and Syria. [4] It has been largely cultivated in southern Europe and played a vital role during the early civilizations of Egypt and Greece. [5] About 98% production of olive and its products in the world is concentrated in the Mediterranean basin countries. [6] The nutritional and medicinal importance of olive leaves, fruit, and its oil have been exploited in several households long ago.^[7] The consumption of the fruits and liquid extracted from olive tree was evident in the copper age (6th millennium BC) and in the 9th and 8th century BC. The oil was used for food as well as to take care of burnt skin, dermatitis, stomach problems, intestine problems and sun protection. [8] There has been an increase in the level of consumption of olive oil throughout the world which may be attributed to its antioxidant and antiinflammatory potentials which are essential in preventing diseases in humans. [9] Several studies have implicated the

consumption of olive oil in decreasing the risk of malignant neoplasms, especially cancers of breast, stomach, ovary, colon and endometrium^[10], as well as prevention of cardiovascular and thrombotic diseases.^[11]

Crude oil, also known as petroleum is a complex mixture of hydrocarbons which can exist in several forms such as aliphatic, alicyclic and aromatic compounds. Most of these compounds when exposed to at a given lethal dose are known to be toxic to different biomass in the ecosystem. [12-13] The acute toxicity of crude oil is usually attributed to low molecular weight hydrophobic petroleum hydrocarbons that penetrate into lipid membranes and cause death by narcosis. [14] Larger compounds, such as alkyl PAHs with three or more rings that are less soluble in water contribute less to acute toxicity because they are taken up more slowly than low molecular weight compounds, however, they are associated with chronic and sub-chronic toxicity owe to the fact that, their metabolism usually yields metabolites that are more toxic than the parent compound. [15]

Honey is a natural substance produced by honeybees of the genera Apis and Meliponini when the nectar and sweet deposits from plants are brought together in the honeycomb and subsequently modified by the biological activities of the honeybees.^[16] It contains essentially concentrated aqueous solution of inverted sugars and complex mixture of other saccharides, proteins, enzymes, amino acids, organic acids, polyphenols, and carotenoid like substances, vitamins and minerals.^[17] When honey is orally consumed, its carbohydrate is easily digested and absorbed to generate energy requirements of about 303 kcal/100 g to the human body. [18] It has also been found by Taormina *et al.*^[19] to be effective against enzymatic browning of fruits and vegetables, oxidative degradation of some foods and growth of some food borne pathogens such as E. coli 0157:H7, S. typhimurium, S. sonnei, B. cereus, L. monocytogenes and S. aureus. In general, the therapeutic potential of this substance could be attributed to the several scientific reports of its antimicrobial, antiinflammatory and anti-oxidant properties as well as immune system boosting capacity. [20]

Palm kernel oil, olive oil, crude oil and honey are used for variety of reasons in traditional medicine. They are also believed to be effective for use as antidote for certain poisons. Due to the missing information of the comparative effects of these selected substances on liver function, this study is designed to provide the information on the effects of palm kernel oil, olive oil, crude oil and honey on liver function of male albino rats.

METHODS

Chemical substances used

Four chemical substances were used in this study. Palm kernel oil, olive oil, crude oil and honey were obtained from Umuahia, Wukari, Port Harcourt and Kurmi L.G.A. of Taraba State, Nigeria respectively.

Experimental animals

A total of 30 healthy male albino rats of 7 weeks of age were used in this study. The albino rats were purchased and kept at the animal house, Department of Biochemistry, Federal University Wukari, Nigeria. They were allowed to have access to feed and water *ad libitum* throughout the experimental period. Standard laboratory protocols for animal studies were followed and all methods were performed in accordance with the relevant guidelines and regulations.

Experimental design

The 30 male albino rats were placed (randomly) into 5 different groups of 6 rats each. The group 1 albino rats served as normal control and did not received any chemical substance. The test albino rats were placed in groups 2, 3, 4 and 5, and were administered palm kernel oil (5 ml/kg bw), olive oil (5 ml/kg bw), crude oil (5 ml/kg bw) and honey (5 ml/kg bw) respectively for 21 consecutive days prior to animal sacrifice. The chemical substances were administered to the experimental rats once daily through oral route.

Blood collection

Following the administration of the chemical substances to the test rats, all the rats were starved overnight, anaesthetized with the use of chloroform and sacrificed by cervical dislocation. Blood samples of the rats were collected through cardiac puncture and dispensed into plain sample tubes. The blood samples were allowed to stand for about fifteen minutes to clot and thereafter centrifuged at 4000 rpm for 10 minutes. The serum was aspirated using Pasteur pipette and dispensed into clean tubes for the biochemical analysis. The liver of all the rats were harvested for histological analysis.

Biochemical and histological analysis

Serum biochemistry was carried out on each blood sample. The activities/concentrations of aspartate transaminase (AST), alanine transaminase (ALT), alkaline phosphatase (ALP) and bilirubin were determined using Reflotron Plus. Total protein was determined using the method of Gomall *et al.*^[21], while albumin was determined using the method of Doumas *et al.*^[22] Globulin was estimated using the equation: Globulin = Total protein – Albumin.

The liver of the rats in all the groups were harvested and fixed in 10% formalin. They were processed using an automatic tissue processor and embedded in paraffin wax, and sections cut using a rotary microtome. The liver sections were stained using haematoxylin and eosin (H&E) method. The sections were examined and photomicrographs of the liver sections taken (Magnification: x200).

Statistical analysis

The results of the biochemical analysis were analyzed statistically using One-Way Analysis of Variance with the use of Statistical Package for Social Sciences, version

21. The means of the group results were compared for significance at p≤0.05. The final group results were

presented as mean \pm SD (n=6).

RESULTS

The results of the biochemical analysis are presented in the tables below:

Table 1: Concentrations of selected liver maker enzyme activities in rats administered palm kernel oil, olive oil, crude oil and honey (U/L).

Parameters	Group 1 (Normal control)	Group 2 (Palm kernel oil: 5 ml/kg bw)	Group 3 (Olive oil: 5 ml/kg bw)	Group 4 (Crude oil: 5 ml/kg bw)	Group 5 (Honey: 5 ml/kg bw)
ALT	70.57 ± 11.21^{a}	68.10 ± 18.48^{a}	74.07 ± 15.61^{a}	65.87 ± 4.40^{a}	53.83 ± 11.89^{a}
ALP	404.67 ± 24.17^{a}	476.33 ± 6.03^{a}	620.33 ± 10.26^{b}	813.67 ± 51.16^{c}	419.00 ± 45.35^{a}
AST	151.03 ± 40.47^{a}	141.73 ± 49.18^{a}	$273.00 \pm 22.90^{b,c}$	$346.33 \pm 40.45^{\circ}$	$218.67 \pm 22.32^{a,b}$

Result represent mean \pm standard deviation of group serum result obtained (n=6).

Mean in the same row, having different letters of the alphabet are statistically significant (p<0.05).

ALT reduced non-significantly (p>0.05) in groups 2, 4 and 5, but increased non-significantly in group 3 compared to the control. ALP increased non-significantly

(p>0.05) in groups 2 and 5, but increased significantly (p<0.05) in groups 3 and 4 compared to the control. AST reduced non-significantly (p>0.05) in group 2, but increased significantly (p<0.05) in groups 3 and 4, and non-significantly (p>0.05) in group 5 compared to the control.

Table 2: Concentrations of proteins and bilirubin in rats administered palm kernel oil, olive oil, crude oil and honey.

Parameters	Group 1 (Normal control)	Group 2 (Palm kernel oil: 5 ml/kg bw)	Group 3 (Olive oil: 5 ml/kg bw)	Group 4 (Crude oil: 5 ml/kg bw)	Group 5 (Honey: 5 ml/kg bw)
Total proteins (g/dL)	3.90 ± 0.13^{a}	$4.81 \pm 0.77^{a,b}$	$4.70 \pm 0.48^{a,b}$	5.35 ± 0.43^{b}	5.30 ± 0.55^{b}
Albumin (g/dL)	1.34 ± 0.13^{a}	0.41 ± 0.06^{b}	0.73 ± 0.09^{c}	$0.56 \pm 0.26^{b,c}$	1.65 ± 0.04^{d}
Globulin (g/dL)	2.56 ± 0.25^{a}	$4.39 \pm 0.72^{b,c}$	$3.97 \pm 0.56^{b,c}$	4.78 ± 0.19^{c}	3.66 ± 0.54^{b}
Bilirubin (µmol/L)	13.55 ± 6.21^{a}	$19.40 \pm 10.30^{a,b}$	$23.17 \pm 7.75^{a,b}$	35.10 ± 1.59^{b}	$24.57 \pm 13.01^{a,b}$

Result represent mean \pm standard deviation of group serum result obtained (n=6).

Mean in the same row, having different letters of the alphabet are statistically significant (p<0.05).

Total proteins increased non-significantly (p>0.05) in groups 2 and 3, but increased significantly (p<0.05) in groups 4 and 5 compared to the control. Albumin and globulin increased significantly (p<0.05) in all the test groups compared to the control. Bilirubin increased non-significantly (p>0.05) in groups 2, 3 and 5, but increased significantly (p<0.05) in group 4 compared to the control.

Photomicrographs of the liver sections are presented below:

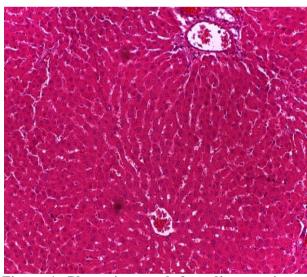


Figure 1: Photomicrograph from liver section of normal rat (group 1) showing normal features of the hepatic tissue.

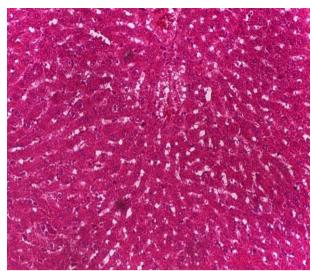


Figure 2: Photomicrograph from liver section of rat administered palm kernel oil (5 ml/kg bw).



Figure 3: Photomicrograph from liver section of rat administered olive oil (5 ml/kg bw).

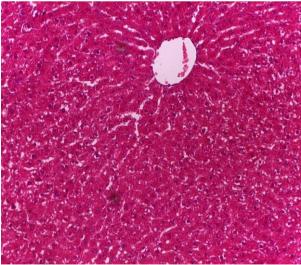


Figure 4: Photomicrograph from liver section of rat administered crude oil (5 ml/kg bw).

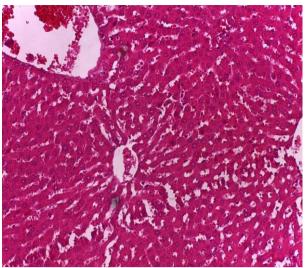


Figure 5: Photomicrograph from liver section of rat administered honey (5 ml/kg bw).

DISCUSSION

Natural substances such as honey, palm kernel oil, olive oil used as component of foods as well as for their therapeutic importance may become toxic to the cell depending on the level and period of consumption while others such as crude oil can exhibit deleterious effects on the body cells at different levels of exposure, especially when the body internal protective mechanism such as endogenous antioxidants is being comprised. Assessment of liver function alteration can be made by estimating the activities of serum ALT, AST and ALP which are enzymes originally present at higher concentrations in cytoplasm of hepatocytes. [23] They are usually present in low concentrations in the serum under normal physiological condition where their functions are not actually known and hence are called non-functional enzymes in the serum.^[24] These enzymes leak into the blood stream when there is hepatopathy and rupture of cell or organelle membranes. Their concentrations in the serum are usually in conformity with the extent of liver tissues damage. [25] Administration of palm kernel oil and honey did not cause any significant change (P<0.05) on the liver enzymes (ALT, ALP and AST), while olive oil and crude oil showed significant increase (P<0.05) in ALP and AST but not on ALT (Table 1). The significant increase in these enzymes (ALP and AST) caused by these chemical substances may be as a result of direct damage to the hepatocytes^[25], but more so that, the very specific liver marker enzyme ALT is not affected. This could mean that the increase may have arisen from the damage done to other cells or organs such as kidney, heart and muscle that can also synthesis ALP and AST. The alteration of these enzymes is in line with the one caused by administration of consumable chemical substances such as acetaminophen as demonstrated by Imo $et\ al.^{[26]}$ and Aluko $et\ al.^{[27]}$ and non-consumable substances as reported by Imo $et\ al.^{[28]}$ and Abiodun $et\ al.^{[28]}$ $al.^{[\widetilde{29}]}$

Abnormal levels of Albumin and globulin have been reported to be associated with haemolysis or liver damage. They are both produced by the liver, if the liver is damaged, it may no longer be able to produce these proteins. The significant increase (P<0.05) in total proteins, albumin and globulin (Table 2) upon administration of palm kernel oil, honey, olive oil and crude oil could be an indication of non-toxic effect exhibited on the liver cells by these compounds whereas, the significant rise (P<0.05) in bilirubin may have resulted from internal haemorrhage since bilirubin is an intermediate metabolic product of haeme group found in haemoglobin or myoglobin breakdown in the liver^[30] or this rise in bilirubin could also be associated with the reduction of its uptake by the hepatocytes. [31] Bilirubin levels in the blood can be elevated due to overproduction, decreased uptake by the liver, decrease conjugation or obstruction of the bile duct. High level of bilirubin in the blood is known as hyper-bilirubinemia, and can cause jaundice. The result of this study showed that further administration of the chemical substances or increase in the volume administered to the test animals may induce hyper-bilirubinemia.

Comparative evaluation of photomicrograph of liver sections of the test rats when compared with the control (Figure 1-5) showed that olive oil and crude oil may negatively alter certain liver functions than palm kernel oil and honey. Palm kernel oil and honey may support certain liver functions. Administration of the olive oil showed mildly enlarged central vein (Figure 3), while administration of the crude oil showed degeneration of some of the hepatocytes with mild alteration of some periportal tissues (Figure 4) when compared with the control. In this study, it is observed that ingestion of crude oil and olive oil may be very dangerous to the liver and even some other tissues in albino rats, therefore, may adversely affect the liver of humans.

CONCLUSION

Comparative evaluation of long-term administration of palm kernel oil, olive oil, crude oil and honey as in this study showed olive oil and crude oil may negatively alter some liver functions than palm kernel oil and honey. Palm kernel oil and honey may support certain liver functions.

COMPETING INTERESTS

The authors declare that they have no competing interests.

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