

# Faecal Composition and Moisture after Chronic Consumption of Different Forms of Palm Oil Diets in Rats

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**Abstract:** The effect of chronic consumption of fresh and thermally oxidized palm oil on faecal composition and moisture of rats was studied. The animals were divided into 3 groups of five rats each. The first group control was fed on normal rat chow only. The second (FPO) and third group (TPO) were fed diet containing either fresh or thermoxidized palm oil (15% wt/wt) for 14 weeks. The rats in the entire group were allowed free access to food and water. At the end of the feeding period, faecal protein, glucose, moisture and electrolyte were determined. The results show that there was no significant difference in faecal weight among the groups. The faecal protein in control, FPO and TPO was  $27.50 \pm 33.42$ g/100ml,  $31.50 \pm 3.67$  and  $51.68 \pm 0.005$ g/100ml respectively. The faecal protein in TPO was significantly higher ( $P < 0.05$ ) than that in control or FPO group. Faecal glucose was significantly lower ( $P < 0.05$ ) in TPO than control or FPO. There was however no significant difference in faecal  $\text{Na}^+$  and  $\text{K}^+$  among the groups. The total moisture loss and total faecal lipids were significantly raised ( $P < 0.05$ ) in TPO than in control or FPO group. In conclusion, chronic consumption of thermally oxidized oil may lead to loss of proteins, lipids and water in faeces. This may be dangerous to health and so thermally oxidized oil should be used with caution.

**Keywords:** Electrolytes, faecal lipids, faecal moisture, palm oil.

## 1. Introduction

Palm oil is derived from the plant *Eleasis guineensis*. It is one of the most important oil producing plants in the tropics. The plant is wide spread throughout West, Central Africa and other tropical countries [1,2]. The fresh form is rich in vitamin A, E and carotenoid [3]. It also contains sterols and squalene [4,5]. These antioxidants confer fresh palm oil same protective status. Palm oil is incorporated as a minor component in fat blends and is used in manufacturing food products. Like other edible fats and oils, palm oil is easily digested, absorbed and utilized to support healthy growth. It is used either in its fresh form or oxidized state but at various levels of oxidation. It is fresh when it is not subjected to frying at very high temperatures. Both forms are commonly used in various food processing industries and many stable diets. When palm oil is exposed to oxygen, spontaneous oxidation is triggered at an accelerated rate [6]. Such stimulants could be thermolytic, chemolytic, photolytic or enzymatic [7]. The oxidation follows a free radical chain reaction which devastates the component fatty acid, especially if it is unsaturated. Thermoxidation is potentially destructive to dietary oils especially when they are exposed on an intermittent basis.

Many symptoms indicative of tissue pathology, functional impairment and toxicity accompany ingestion of oxidized oils. They range from growth retardation which is manifested in the reduced body weight, reduced biliary secretion and damaged liver [8] damage to nucleic acids, fatty acids as well as micro-nutrients deficiency [7]. There is also, alteration in the morphology of tissues [9,10]. These are besides the damage to intestinal mucosa, decreased villus height and disintegrated epithelium [11]. Chronic ingestion

of oxidized palm oil also leads to poor feeding efficiency and reduced gut absorption [11,12]. These tend to exacerbate the state of protein deficiency probably due to carrier protein insufficiency.

Due to generalized tissue damage the cell membranes are not spared. There is leakage of electrolytes ( $\text{Na}^+$ ,  $\text{K}^+$  and  $\text{H}^+$ ). The electrical conductivity of the membrane is therefore amplified and there is increase in motility [8,12]. Because of the distortion in gut morphology there is possibility of the disturbance in the digestion and absorption of food. This may manifest in the faecal composition of the affected rats. This study was therefore conducted to examine the effect of chronic consumption of fresh and thermoxidized palm oil diets on biochemical, nutritional and electrolytes composition of faecal matter in rats.

## Materials and Methods

### 2.1 Preparation of Experimental Diets

Ten litres of fresh palm oil were purchased from a local market in Calabar, Nigeria. The palm oil was certified fresh by virtue of its low oxidation (rancidity) value [13]. The fresh palm oil was divided into two equal parts. One part was used fresh for formulation of fresh palm oil diet while the other part was subjected to thermoxidation and used for thermoxidized oil meals. The rat feed was bought from a local dealer in Calabar. The commercial rat feed contained 3.5% fat, 14% protein and 7% fibre. The approval for the study was obtained from the College of Medical Sciences ethical committee.

## 2.2 Thermoxidation of Palm Oil

Fresh palm oil contained in a stainless steel pot was intermittently heated at a temperature of 150°C for 5 rounds with each round lasting for 20 minutes. The oil was allowed to cool for 5 minutes before the next successive round of heating.

## 2.3 Formulation of Test Diets

Since the level of palm oil in most West African dishes is about 15%, palm oil diet was formulated as described by [11]. Briefly 15g of fresh palm oil was mixed with 85g of animal feed, to produce fresh palm oil diet (FPO). The cooled thermoxidized palm oil was also mixed 15% wt/wt to produce thermoxidized palm oil diet (TPO). The control group had normal rat feed diet. All the rats had free access to food and water *ad libitum*.

## 2.4 Faecal weight and Texture

Faeces were obtained for 2 weeks from each animal in the three groups and weighed using a precision weighing balance (Ohaus, USA). The texture was assessed by rubbing the faeces with the fingers to determine whether it is coarse greasy or wet.

## 2.5 Determination of Faecal Moisture

Fresh faecal materials were collected from all the rats for determination of moisture content. Moisture content was determined by lyophilisation for 24 hours and calculated thus:

$$\frac{\text{Moisture loss} \times 100}{\text{Wet sample (g)}}$$

Faecal content was weighed and dried at 90°C. The difference in weight after drying is the moisture loss. Faecal moisture loss of 80% was considered to indicate occurrence of clinical diarrhoea [14].

## 2.6 Determination of Faecal Electrolytes

Faecal electrolytes were determined after making a homogenate of the faeces using 15ml of distilled water for 1g of faeces. The homogenate was then filtered using Whatman's filter paper. The supernatants was then transferred into plastic containers and frozen at -15°C. The electrolytes, Na<sup>+</sup> and K<sup>+</sup> were determined by the principle of ion selective electrode using flame photometer (Model 410C petracourt LTD England) at a wavelength of 598nm and 767nm for sodium and potassium ions respectively.

## 2.7 Determination of faecal glucose and protein

Faecal protein estimation was by the Buret method of [15]. The faecal glucose was by Dialab method of [16]. The principle is based on the fact that in the presence of glucose oxidase, glucose is oxidized to glucuronic acid and hydrogen peroxide. The hydrogen peroxide reacts in the presence of peroxidase with phenol and 4-amino phenozone to form Quinone dye (pink) whose colour intensity is in proportion to the glucose concentration in the sample.

## 2.8 Determination of Total Faecal Lipids

Total faecal lipids were extracted by the modified method of [17] and by the method of [18]. A small amount of water (0.5ml) was added to 0.5g of frozen dried faeces. 5ml of Methanol followed by 10ml of chloroform were added to each test-tube and heated in a water bath at 60°C for 1hour. The solid residue was re-extracted with methanol and chloroform and heated at 60°C for 1 hour in a water bath. Potassium chloride (8.8gm/L) in water was added to the filtrate in a proportion of one quarter to the total volume. The content was then centrifuged at 265g for 5 minutes to obtain 2 distinct layers. The bottom layer containing the purified lipid was transferred to a test-tube and weighed. It was later dried overnight under nitrogen until a solid lipid residue was obtained. The difference in weight before and after drying was the weight of the total lipids.

## 2.9 Statistical Analysis

All data are expressed as mean ± SEM. Analysis of data was done using graph pad prism Software version 5. (Graph pad software, San Diego California USA) and one-way Anova was used to compare means followed by post hoc Bonferroni test where P-values were significant. A P-value of 0.05 was considered significant.

## 3. Results

### 3.1 Daily Water and Food Intake

The mean daily water intake for control, FPO and TPO were 19.86 ± 0.29ml, 19.17 ± 0.23ml and 18.36 ± 0.23ml respectively. The food intake was 7.71 ± 0.36, 7.53±0.31g and 7.14 ± 0.35g respectively. There was no significant difference in food or water intake among the groups (Table 1).

### 3.2 Body weight changes

The mean change in body weight of control, FPO and TPO group was 172.04 ± 0.08g, 174.0 ± 0.93g and 158.3 ± 0.90g respectively. The change in body weight in TPO was significantly lower (P<0.05) than either control or FPO group. Table 1.

### 3.3 Faecal Weight and Texture

The mean faecal weights for control, FPO and TPO were 5.05 ± 0.29g, 5.13 ± 0.27g and 5.22 ± 0.42g. There was no significant difference between the groups. The texture of the faeces of the control group was solid and rough. It was solid but smooth in FPO group and semisolid in TPO group (Table 1).

### 3.4 Faecal Protein

The faecal protein was measured in g/100ml of the faecal extract. The protein content of the faeces of control rats was 27.50 ± 3.43g/100ml while that for FPO and TPO were 31.50 ± 3.67 and 51.68 ± 0.005 respectively (Table 1). There was no significant difference between FPO and control

group but between TPO and control or FPO there was significant difference ( $P < 0.05$ ).

### 3.5 Faecal Glucose

The mean faecal glucose is as shown on Table 1. The control group was  $0.96 \pm 1.0$ , while the FPO and TPO groups were  $0.96 \pm 1.10$  and  $0.67 \pm 0.05$  mMol/L respectively. There was no significant difference between the control and FPO group, but the mean faecal glucose of TPO was significantly lower ( $P < 0.05$ ) than that for control or FPO group.

### 3.6 Faecal Electrolytes

The mean sodium ion concentration in control, FPO and TPO were  $15.80 \pm 1.51$ ,  $15.05 \pm 1.10$  and  $14.46 \pm 1.19$  mMol/L, respectively. There was no significant difference between any two groups. Conversely, there was also no significant difference in  $K^+$  concentration between the groups as shown on Table 1.

**Table 1:** Faecal protein, glucose and electrolytes

Parameter	Control	FPO	TPO
water intake	19.86±0.29	19.17±0.23	18.36±0.23
Food intake (g)	7.71±0.36	7.53±0.31	7.14±0.35
Body weight changes (g)	172.04±0.08	174.0±0.93g	158.3±0.90
Faecal Weight (g)	5.05±0.29	5.13±0.27	5.22±0.42
Faecal texture	Solid & coarse	Semi Solid with yellow pigments	Semi-solid
faecal protein (g/ 100ml)	27.50±3.43	N.S 31.50±3.67	<sup>a</sup> 51.68±4.05
faecal Glucose (mMol/L)	0.96±0.10	NS 0.97±0.10	0.67 <sup>ab</sup> ±0.05
Na <sup>+</sup> mMol/L	15.80±1.51	15.85±1.8 <sup>NS</sup>	14.46±1.19 <sup>NS</sup>
K <sup>+</sup> mMol/L	10.00±0.39	10.85±0.81	11.51±0.20

n = 5 for all groups, results are presented as mean ± SEM

b =  $P < 0.05$  v FPO

\* =  $P < 0.05$  vs. control

NS = Not significant vs. control

### 3.7 Faecal Moisture Loss

The percentage moisture loss in faeces of the control, FPO and TPO groups were  $22.29 \pm 3.75\%$ ,  $22.15 \pm 2.40$  and  $27.80 \pm 0.89\%$  respectively. The moisture loss in TPO was significantly higher ( $P < 0.05$ ) than either control or FPO group (Table 2). There was no significant difference between control and FPO group.

### 3.8 Total Faecal Lipids

The total faecal lipids for control, FPO and TPO groups were  $0.047 \pm 0.003$ g,  $0.110 \pm 0.002$  and  $0.184 \pm 0.005$ g respectively. The total faecal lipids in TPO was significantly higher ( $P < 0.05$ ) than either control or FPO group. The faecal lipids in FPO was significantly ( $P < 0.05$ ) higher than in control (Table 2).

**Table 2:** Comparison of Faecal Moisture loss and Lipids

Parameter	Control	FPO	TPO
Faecal Moisture Loss (%)	22.29 ± 3.75	22.15 ± 2.40	27.80 ± 0.89*
Total Faecal Lipids (g)	0.047 ± 0.003	0.110 ± 0.002*	0.184 ± 0.005* <sup>a</sup>

\* $P < 0.05$  vs Control; a =  $P < 0.05$  vs Control

## 4. Discussion

Chronic consumption of fresh and thermoxidized palm oil did not produce significant changes in food and water intake in rats. It is likely therefore, that the two forms of palm oil diets were as palatable as the control diet to rats. The rats in control and FPO meals had more weight than that in TPO probably because TPO meals are less nutritious as heat destroys all ingredients capable of good health [7].

There were also no significant changes in faecal weight among the groups which may be a consequence of same amount of diet and water intake among the groups. There was however difference in the texture of the faeces among the three groups. The difference in texture may be due to the treatments of the diets especially the thermoxidized palm oil diets that produced semi-solid coarse faeces in comparison with the solid coarse faeces of control rats. The faeces of fresh palm oil diet fed rats were also semi solid (watery) with yellow pigment which was not identified.

Igiri *et al* [10] and Obembe *et al* [11] have all reported that chronic consumption of oxidized oil leads to distortion in the morphology of the gastrointestinal tract which invariably affects the villi. Damage to the villi could lead to poor digestion and absorption of nutrients. It is possible therefore; that the oils in the test diets were poorly digested and absorbed, thus producing semi-solid faeces in rats fed the two forms of palm oil diets.

In this study, there was significant decrease in the faecal glucose and increase faecal proteins in rats feed on TPO meals when compared with that of the control or FPO group respectively. Alleyne *et al* [19] have documented that reduced intestinal mass as well as villus height due to distortion may lead to protein restriction which invariably affects protein digestion and absorption, hence, its appearance in faeces. The decreased faecal glucose in TPO group when compared with control or FPO, may be the body's way of making up for lost protein because glucose possesses both anabolic (proteogenic) and protein sparing ability, so the decreased glucose in faeces of the TPO may be due to the diversion of glucose by the body to more efficiently meet energy demands of the cells due to protein loss [20].

There was no significant difference in faecal  $K^+$  and  $Na^+$  among the groups. The efficiency of absorption of electrolytes was the same for all the groups studied, chronic ingestion of thermally oxidized palm oil may therefore not grossly affect electrolytes absorption.

The faecal moisture loss was higher in the TPO (27.80 ± 0.89%) than in control (22.29 ± 3.75%) or FPO (22.15 ± 2.40%). However, the percentage loss in the TPO groups was not up to that reported by [14] for the diagnosis of clinical diarrhoea in pigs. The percentage for the diagnosis of diarrhoea in rats is not known. The lower value found in this study may be due to differences in diet and species of animals used.

The high total faecal lipid in the TPO group may be due to poor digestion and absorption of lipids owing to damage of the intestinal mucosa [10]. It may also be due to liver damage following consumption of oxidized palm oil diet [4,21]. The damage to the liver may have affected bile secretion since the bile is required for digestion and absorption of fats. All of these may be responsible for the steatorrhea reported by [12] after ingestion of oxidized oil.

## 5. Conclusion

In conclusion, chronic consumption of thermoxidized palm oil diet may lead to increased loss of faecal moisture, lipids and proteins thereby limiting their bioavailability to tissues, its consumption should therefore be with caution.

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