

COMPARISON OF RADIATION EFFECTS ON WEIGHT OF RABBITS AFTER CONSUMPTION OF FRESH AND THERMOXIDIZED PALM OIL DIETS

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ABSTRACT

This study, investigated the effects of radiation on weight change in rabbit following consumption of diets mixed with fresh and thermoxidised palm oil, common diets among the people in the tropics. Two groups of rabbits each were fed with normal diets (Control group 1), Fresh palm oil diets (FPOD) and thermoxidised palm oil diets (TPOD), respectively, while one batch of each group (Control group 2R, FPOD 2R and TPOD 2R, respectively) were subjected to X-irradiation. Weights of the experimental animals were taken over 12 weeks of feeding, followed by another ten weeks after a break of seven weeks to allow for recovery of the animals. Results showed that FPOD diets produced a weight enhancement effect on the animals, with 14.86% weight increase in 12 weeks of feeding against 13.05% for the control group 1 animals. This reduced after X-irradiation to a weight gain of 8.85% for the control group 2R and 7.08% for the FPOD 2R group. TPOD groups showed poorer weight response at -3.75% for the group 1 and -3.32% for the irradiated group 2R. Repeating the experiments after the rest period showed the same pattern of results but at slightly lower values. The results are statistically significant at $P < 0.0001$, suggesting that X-radiation aggravates weight loss in TPOD subjects, but its effect on FPOD and Control diets subjects are reversible.

KEYWORDS: Radiation, palm oil, weight, thermoxidised, Rabbits

1. INTRODUCTION

Palm oil is a major derivative from the mesocarp of the fruits of the tropical tree -*Elaeis guineensis* and forms an integral part of the diet of the people in the tropical areas, like Nigeria (Ebong et al, 1999), and indeed in many parts of the world for many years (Cotrell, 1991). It is a rich source of beta-carotene, a common precursor of vitamin A, which promotes good night vision (Guyton & Hall, 2000). Fresh palm oil contains both saturated (47%) and unsaturated (53%) fatty acids (Kochar, 1981), while the oxidized form has higher levels of saturated acids (Okiv and Oke, 1986). The utility of the oil is in both fresh and thermoxidized forms for culinary purposes. (Osim et al, 1992).

The traditional method of production by boiling followed by squeezing the fruit and heating the oil obtained, at a low temperature to remove the debris (Ekpa et al, 1994), produced the fresh palm oil. Five rounds of heating of the fresh oil produced the thermoxidized palm oil (Osim et al, 1996).

Oxidation is a part of the normal metabolism in the human body (Salonen, 2000) resulting in the production of radicals and

intermediate forms of reactive species, which can readily oxidize other molecules with which they come in contact. Radiation has been shown to greatly increase the toxicity of the palm oil due to the production, by oxidation, of increased fatty acid, peroxide and iodine levels (Egbe et al, 2003). These results, as well as the production of free radicals have been linked to both heart disease and cancer. (Sun, 1990; Pryor, 1991; Lunec, 1992). Some extraneous factors like alcohol, stress and environmental pollutants have been reported to increase the generation of free radicals in the body (Salonen, 2000)

This study is designed as part of the continuous investigation on the radiation effects on everyday phenomena that may play a part in the design of radiation protection protocol in Medical and industrial practices. Palm oil is widely and regularly consumed in its varied forms across the world, and particularly in the tropics. Occasionally, palm oil consumers present with medical conditions that require the use of ionizing radiation for diagnostic and therapeutic processes. Besides, the oil is a common trade commodity and sometimes is the subject of security scans at entry and exit points of sovereign states.

This study sets out to examine if there are any benefits or otherwise to the consumer from radiation interaction with the oil within the consumer (say during diagnostic examination) and one who consumes the oil that may have been exposed to irradiation while in transit. Rabbits has been used as a model system to assess the radiation effect of X-radiation on palm oil diets following consumption of fresh and thermoxidised palm diets.

2. MATERIALS AND METHODS

Thirty six male, inbred rabbits selected from a herd of *Orytolagus cuniculus*, on the basis of meeting the criteria of weight (0.450 to 0.453 ± 0.001 kg), and age (6 weeks old) from the animal house of the Department of Medical Physiology, University of Calabar, Nigeria, were housed in wooden cages separated from the other animals, for this study. The conditions of housing were drawn from Suckow and Douglas (1997) and the guidelines for animal care from ARENA (1992), with the assistance of a registered Veterinary clinician were followed. This is because there are currently no regulations governing the welfare of laboratory animals in Nigeria. The rabbits were divided randomly into six groups of six rabbits each, and maintained at room temperature range of $26 - 28^{\circ}\text{C}$ for the period of the study. They also satisfied a fitness test by the veterinary clinician before inclusion in the study. The rabbits in each group were weighed prior to and after the commencement of the experiment, and their mean weight and standard deviations recorded.

2.1 Preparation of the Rabbit Feed

Three portions having high fiber, protein and low carbohydrate content were made from animal feed (pellets), obtained from Vital Feeds®, Nigeria, which was the regular feed of the animals before the experiment. One portion was kept as the control, while the other two were mixed with fresh palm oil and thermoxidized palm oil, at the ratio of 15% palm oil to 85% feed, the average percentage concentration of palm oil in traditional tropical diets (Osime, *et al.*, 1992) to make FPOD and TPOD, respectively. After the mixing, which was done in a semi-dark room to reduce the rate of oxidation by light on the fresh oil, the feed were stored in black bags and kept in a darkroom, to reduce continuous oxidation. The different groups of animals were fed with the prepared feed for a period of twelve weeks, in the first instance, and their mean weight taken weekly. Feed was prepared to last only six weeks, after which a fresh consignment was prepared following the same procedure.

2.2 Food and Water intake

One hundred (100g) of food ration was served daily to each group of rabbits, with 2 litres of water in individual water bottles with soft rubber teats, which allowed for release of water only on contact with some mild pressure from the rabbits. This arrangement reduced losses to negligible levels and made it easier to quantify intake per group. The average weekly food and water intake were recorded.

2.3 X-irradiation

After the third week (21 days after commencement) of feeding and weighing, the animals in Control group 2R, FPOD 2R and TPOD 2R were subjected to X-radiation from a high frequency, three-phase generator model R501, at a tube potential of 65 kV, 200 mA and 0.8 secs (160 mAs), which yielded an absorbed dose of 38 mGy, measured with a Radcal® radiation multi-o-metre, at a source to object distance of 90 cm. The tube was calibrated with exposure accuracy of $\pm 3\%$, with the beam collimated to the abdominal portion of each immobilized animal, for a single exposure. The rabbits were returned to their cages after the exposure and fed continuously for the rest of the period of study with their respective groups. The animals were rested for seven weeks with restoration of normal feed during the period. Their weights were continuously taken over the rest period. After this, the experiment was repeated for all the groups for another ten weeks. The animals were fed for three weeks and then irradiated with the same exposure, and their weights taken over the following seven weeks. The weights measured over the period are recorded in Table II. The mean weight of the rabbits from week 20 to week 29 following the repeat of the exposure after a period of 7 weeks discontinuation of the feed were also recorded.

3. RESULTS

The results obtained in the duplicated experiment are presented in Tables I – II. The data show that FPOD diets had a weight enhancement effect on the animals, showing 14.86% weight increase in 12 weeks of feeding against 13.05% for the Control group 1 animals. The results from the groups exposed to X-radiation show a weight gain of 8.85% for the control group 2R and 7.08% for the FPOD 2R group. TPOD groups showed weight losses of -3.75% for the group 1 and -3.32% for the irradiated group 2R. Repeating the experiments after a rest period gave the same pattern of results but at values less than those obtained in the first set of results. The results are statistically significant at $P < 0.0001$ for a two tailed probability test, for all the variables studied

Table I: Percentage weekly food and water consumption.

Stage	Control group		FPO		TPO	
	Food	Water	Food	Water	Food	Water
Before irradiation	71.2 ± 3.26	64.1 ± 3.0	70.2 ± 1.37	76.3 ± 0.1	42.8 ± 0.1	68.6 ± 0.4
After irradiation	59.6 ± 2.96	66.3 ± 0.8	54.4 ± 2.6	88.5 ± 0.5	34.2 ± 4.1	80.1 ± 0.6
% difference	8.84	-1.64	12.68	-7.40	11.17	-7.74

Table II: Mean weight (kg) of rabbits

Group	Wk1	Wk 12	% Difference
Control 1	0.450	0.510	6.25
Control 2R	0.450	0.490	4.23
FPOD 1	0.450	0.518	7.02
FPOD 2R	0.451	0.484	3.52
TPOD 1	0.451	0.432	-2.16
TPOD 2R	0.450	0.432	-2.10

Table III: Mean weight (kg) of rabbits following repeat of the exposure after discontinuation of feed for seven weeks.

Group	Wk 20	Wk 21	Wk 22	Wk 23	Wk 24	Wk 25	Wk 26	Wk 27	Wk 28	Wk 29	Mean
Control 1	0.562	0.564	0.567	0.570	0.572	0.576	0.579	0.582	0.583	0.585	0.574
* Control 2R	0.563	0.563	0.562	0.562	0.564	0.565	0.568	0.569	0.572	0.573	0.566
FPOD 1	0.562	0.563	0.566	0.569	0.570	0.574	0.579	0.583	0.584	0.586	0.574
*FPOD 2R	0.562	0.562	0.559	0.557	0.558	0.561	0.561	0.568	0.569	0.574	0.564
TPOD 1	0.562	0.561	0.561	0.559	0.556	0.553	0.552	0.551	0.551	0.550	0.556
*TPOD 2R	0.563	0.561	0.560	0.558	0.556	0.551	0.551	0.548	0.548	0.547	0.554

* Groups subjected to X-radiation indicated by 'R'

FPOD = Fresh Palm Oil Diets

TPOD = Thermostoxidized Palm Oil Diets

4. DISCUSSION

Palm oil in its fresh state has been reported to have nutritional and beneficial properties (Osim *et al*, 1992; Osim *et al*, 1994; Osim *et al*, 1996; Ebong *et al*, 1999). Instead it exhibits anti-hypertensive effects on the endothelium of blood vessels, as well as prevent oxidative damage due to large amounts of vitamin A precursors, and vitamin E. It also induces hepatic drug metabolizing enzymes (Manorama *et al*, 1993; Owu *et al*, 1998). Fresh palm oil also prevents cellular aging, atherosclerosis, cancer, arthritis and Alzheimer's disease (Walton & Parker, 1980; Hirai *et al*, 1982; Cross, 1987; Elson and Qureshi, 1995). The performance of the FPOD group 1 rabbits seems to support these reports.

On the other hand, thermostoxidized form of the oil has been reported to contain more saturated fats and cause tissue damage in the liver, lungs and kidney (Osim *et al*, 1994). Increased vascular resistance (Yin *et al*, 1991; Kitagawa *et al*, 1992;

Owu *et al*, 1998) liver hypertrophy and elevation of triglycerides (Gabriel *et al*, 1977; Causeret *et al*, 1978; Mouni *et al*, 1986) have also been traced to thermostoxidized palm oil. On reproductive physiology, the reports have cited toxicity producing functional changes in the testes of male animals (Krivenlova and Treschuk, 1978), gestational resorption syndrome in female rats, discoloration in the uterus, ovary and testes of rats (Giassudin (1985), delayed gestation, reduced fertility and embryo-foetal toxicity (Isong *et al*, 1997).

Radiation effects on biological medium have been severally reported to be due to direct or indirect routes, and these may also be immediate or delayed (Yarmonenko, 1988; Salonen, 2000; Egbe *et al*, 2003). It has been shown that radiation does cause oxidation of palm oil (Ekpa, *et al*, 1994; Egbe *et al*, 2003) and since the bulk of the oil palm sold in open markets across the tropical areas is exposed to direct sunlight, it is therefore mostly consumed in oxidized forms. Oxidation products from X-irradiation of palm oil have produced about

25.2% rise in peroxide and 68.3% rise for acid values, respectively, with iodine rising by about 3.5% (Egbe *et al*, 2003).

Despite the low radiation dose used, it is a possibility that natural anti-oxidants in fresh palm oil, tocopherols and tocotrienols are affected by X-radiation resulting in accumulation of some oxidation species (Walton and Packer, 1980; Hirai, *et al*, 1982; Cross, 1987; Elson and Qureshi, 1995). These along with radicals (OH from water in the tissue) and other reactive species produced by irradiation within the rabbit, could have produced the observed results (Yarmonenko, 1988). The situation is much more complicated in the case of thermoxidised palm oil where the expectation of a more drastic weight loss as a result of radiation adding its oxidation products to those of thermoxidised oil in the food, failed to occur. It may seem here that either the system was at its oxidation peak, or perhaps some products of thermoxidation somehow prevented the oxidative action of irradiation. This is the subject of further research. However, it has been shown at higher radiation doses that lipid radio toxins, the complex products of the oxidation of unsaturated fatty acids, like hydro peroxides, epoxides, aldehydes and ketones, can cause haemolysis, among other characteristic effects imitating the biological effects of ionizing radiation (Giassudin, 1985). Irradiation of tissues in the presence of these materials leads to a rise in their concentration owing to the depression of the anti-oxidant system. This promotes accelerated oxidation reactions of the radical type (Giassudin, 1985).

From the results in this study, the effect of radiation alone is easily reversed by the body if the additional oxidation load occasioned by the presence of products of radiation oxidation (from FPOD) and thermoxidation (in TPOD) were not present. The effects of radiation oxidation of fresh palm oil in the FPOD are short-lived as the animals defense system appeared capable of overcoming this effect, evident by the weight increase with feeding time. The radiation and oxidation effects probably contributed to the lowered food intake of the animals, a development that could have significantly contributed to the weight losses. However, we note that the FPOD 2R group weights fell below the Control 2R group weights, compared to the neck-to-neck result obtained in the FPOD 1 and Control group 1 animals. This may be largely due to the presence of more oxidation products as a result of radiation induced oxidation of the palm oil in the FPOD group 2R diet, than were in the Control group 2R animals. The case of the TPOD results could be accounted for by the imposition on the animal of an oxidation burden, which in a sense might have reached saturation points as to make for little and insignificant difference between the weights of TPOD1 and TPOD2R groups. However,

the combined effects of radiation oxidation and thermoxidation could lead to an imbalance between the formation of radicals and the defense capabilities (or lack of it) of the body defense system (Salonen, 2000), which may hinder recovery.

5. CONCLUSION

Ionizing radiation increases oxidation in subjects who have consumed fresh and thermoxidised palm oil diets. A system that is susceptible to oxidation was subjected to X-radiation; the results were a significant ($p < 0.0001$) weight loss, and in the case of a system that is already under the effects of thermoxidation, lead to body weakness, and some degree of hair loss. These results suggest critical implications for the irradiation of subjects after consumption of palm oil diets which are prone to high risk of increased oxidation. Even though the doses used are within the diagnostic range of radiation, these results could find application in the development of criteria for radiation protection, particularly in radiation therapy where higher doses are used.

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