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Lipid profiling of Wistar rats fed with processed breadfruit

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Abstract

Background: Breadfruit is a staple leguminous food that is widely consumed in the tropical and subtropical regions of the world and is an important source of dietary nutrients for adults, infants and children.

Aim: This study is aimed at identifying the effect of feed fortification with processed breadfruit on the bodyweight and Lipid profile of Wistar rats.

Methods: The Lipid profile was determined using standard diagnostic methods.

Results: The results gotten from the research showed a continuous increase in bodyweight in all groups with exception of groups fortified with cooked breadfruit. The total cholesterol level of the experimental rats decreased in all the groups with a significant decrease (p < 0.05) recorded in the group fortified with 50% parboiled breadfruit. All groups except groups fortified with cooked breadfruit showed a significant increase (p < 0.05) in their High-Density Lipoprotein (HDL) level when compared with the control group while the Low-Density Lipoprotein (LDL) level of the experimental rats recorded a significant decrease (p < 0.05) in the group fortified with 50% parboiled breadfruit. Also, a significant increase (p < 0.05) in Triglyceride (TRIG) and very low-density lipoprotein (VLDL) levels were only recorded in the group of rats fortified with 50% of grilled breadfruit.

Conclusion: The result of this research however suggests no adverse effect of feed fortification with processed breadfruit on the lipid profile of the experimental animals but suggests that appropriate caution should be applied in the use of grilled breadfruit for feed fortification.

Keywords: Parboiled; Bodyweight; Breadfruit; Total cholesterol; Cooked; Triglyceride; Grilled; Lipoprotein

1. Introduction

According to the National Tropical Botanical Garden [1], all parts of the breadfruit tree is said to yield latex which is useful for boat caulking. It is similar in appearance to breadnuts and jackfruit and has lots of varieties and thousands of common names varying according to its geographic distribution [1]. It is propagated mainly by seeds, although seedless breadfruit can be propagated by transplanting suckers that grow off the surface roots of the tree. Breadfruit which is a known leguminous plant is known to be an excellent source of dietary macronutrients and minerals such as phosphorus,

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potassium, magnesium, sodium, nitrogen, and calcium [2] of which when consumed helps the body to build a strong immune system, and improve the body's health and nutritional status [3]. Research studies reveal that breadfruit contains a considerable amount of anti-nutrients such as Phytate, Tannins, Oxalate, and consumption of these anti-nutrients at high levels often results in toxicity and eventual death [4,5]. According to the research carried out by Nwozo *et al* [6], the levels of anti-nutrient increased significantly (p<0.05) in the raw fraction of African breadfruit seeds with respect to the processed fractions which included steaming, roasting, and particularly boiling thus proper processing before consumption is very necessary to reduce the presence of anti-nutrient [7]. Breadfruit is a staple food in some parts of Africa and contains a substantial amount of minerals, vitamins, and phytochemicals. However, roasting reduces the vitamin content of breadfruit [8].

The seeds of Breadfruit are not considered to be oil seeds but contain about 9-15% of oil [9] which is of higher oil content than those of corn from which oil has been extracted over a long period of time [10], this, however, makes breadfruit relevant in the bio-lubricant and biodiesel segments [11,12] and makes the scientific evaluation of this research on the bodyweight and lipid profile of the experimental animal essential.

2. Material and method

2.1. Sample Collection and Identification

The breadfruit used for this study was purchased from Orie market, Abagana in Njikoka Local Government Area, Anambra State, Nigeria. The sample was identified by a taxonomist in the Department of Botany, Nnamdi Azikiwe University, Awka. The voucher number as deposited in the herbarium of Nnamdi Azikiwe University, Awka is NAUH-77B.

2.2. Processing of Sample

The breadfruit was properly washed and mashed with water to remove its slippery nature and was then dried at room temperature for seven days. After the drying, the breadfruit was shared into three portions for processing.

2.3. Cooked Breadfruit

The first portion of the breadfruit was parboiled for 45 mins in a pot containing only water. The pods were then separated from the chaffs with the help of a corona manual grinding machine. The breadfruit was then cooked using a kerosene stove for a period of 2 hours until it was soft and edible for consumption. Next, the cooked breadfruit was dried under room temperature and pulverized using a corona manual grinding machine, and the dried powdered cooked breadfruit was stored inside a well-labelled airtight plastic container until use.

2.4. Parboiled Breadfruit

The second portion of the breadfruit was parboiled by boiling it inside a pot containing water, on a kerosene stove for 45 minutes, till it was partially cooked. The pods were then separated from the chaffs with the help of a corona manual grinding machine, after which it was dried for one week at room temperature. Next, the pods were pulverized using a corona manual grinding machine, and the powdered parboiled breadfruit was stored inside a well-labelled airtight plastic container until use.

2.5. Grilled Breadfruit

The third portion of breadfruit was grilled on a frying pan using a kerosene stove. The seeds were then separated from the chaffs of the pods and were pulverized using a corona manual grinding machine. The now powdered grilled breadfruit was stored inside a well-labelled airtight plastic container until use.

2.6. Composition of the Rat Feed

The standard feed used was a product of Novum Agric Industries. It was purchased from a Feed dealer in Awka. The ingredients used in the compounding of the standard feed include grains and cereals, vegetables, protein meals, vitamins, minerals, essential amino acids, anti-toxins, enzymes. The composition of the ingredients is as follows: Oil (6%), Protein (16%), Fibre (7%), Ash (10%), Calcium (0.95%) and Phosphorus (0.65%). The feed was fortified with the processed grilled, parboiled, and cooked breadfruit in the following percentages: Using an analytical weighing balance, the feed and respective breadfruits were each measured. To 70 g of feed, 30 g of grilled breadfruit was added; to 70 g of feed, 30 g of parbolied breadfruit was added; to 50 g of feed, 50 g of parbolied breadfruit was added; and to 50 g of feed, 50 g of grilled breadfruit was added; and to 50 g of feed, 50 g of parbolied breadfruit was added; to 50 g of feed, 50 g of parbolied breadfruit was added; to 50 g of feed, 50 g of parbolied breadfruit was added; to 50 g of feed, 50 g of parbolied breadfruit was added; to 50 g of feed, 50 g of parbolied breadfruit was added; to 50 g of feed, 50 g of parbolied breadfruit was added; to 50 g of feed, 50 g of parbolied breadfruit was added; and to 50 g of feed, 50 g of feed, 50 g of parbolied breadfruit was added; to 50 g of feed, 50 g of parbolied breadfruit was added; and to 50 g of feed, 50 g of feed, 50 g of parbolied breadfruit was added; and to 50 g of feed, 50 g of feed, 50 g of parbolied breadfruit was added; and to 50 g of feed, 50 g of feed, 50 g of parbolied breadfruit was added; and to 50 g of feed, 50 g of parbolied breadfruit was added; and to 50 g of feed, 50 g of feed, 50 g of parbolied breadfruit was added; and to 50 g of feed, 50 g of feed, 50 g of parbolied breadfruit was added; and to 50 g of feed, 50 g of feed, 50 g of parbolied breadfruit was added; and to 50 g of feed, 50 g of feed, 50 g of parbolied breadfruit was added; and to 50 g of feed, 50 g of feed, 50 g of parbolied breadfru

cooked breadfruit was added. These formulations were repeated until enough feed was prepared which lasted for a period of one month.

2.7. Study Design

A total of 35 Wistar rats weighing between 120 g-150 g were purchased from Chris Experimental Animal Farm and Research Laboratory, Awka, Anambra State, and randomized into seven groups of five rats each and used for the study. They were maintained and housed in cages under standard environmental conditions (27°C ± 3°C, 12-hour light/dark cycle) in Chris Experimental Animal Farm and Research Laboratory, Awka. The rats were weighed, marked, and put into labelled cages. Their random blood glucose levels were also checked. The groupings were as follows:

Group A – Normal Control

Group B – 70% Standard Feed fortified with 30% cooked breadfruit.

- Group C 70% Standard Feed fortified with 30% parboiled breadfruit.
- Group D 70% Standard Feed fortified with 30% grilled breadfruit.
- Group E 50% Standard Feed fortified with 50% cooked breadfruit.
- Group F 50% Standard Feed fortified with 50% parboiled breadfruit.
- Group G 50% Standard Feed fortified with 50% grilled breadfruit.

2.8. Feeding of the Experimental Animals

The experimental rats were fed accordingly using the feed prepared for each of the groups. The feeding was done for a period of four weeks after which the rats were fasted and anesthetized with chloroform before blood collection. Blood was collected by cardiac puncture and put in plain bottles for lipid profile analysis. The carcasses were properly disposed of by burying them.

2.9. Determination of Weight

The weight of the experimental subjects was checked using an electronic weighing scale. The weight of the rats was monitored before, during, and after the experiment to know whether the chloroform fraction has an effect on the bodyweight of the experimental rats.

2.9.1. Lipid Profile

The lipid profile (Total Cholesterol, Triglycerides, High-Density Lipoprotein-cholesterol, Low-Density Lipoprotein-cholesterol) were determined using Randox test kits [13,14]. Low-density Lipoprotein-cholesterol (LDL-c) was calculated using the standard formula [15]. The procedure used was according to the manufacturer's instructions provided in the manual.

2.10. Statistical Analysis

Data obtained from the experiments were analyzed using the Statistical Package for Social Sciences software for windows version 23 (SPSS Inc., Chicago, Illinois, USA). All the data collected were expressed as Mean ± SEM. Statistical analysis of the results obtained was performed by using Analysis of Variance (ANOVA) to determine if a significant difference exists between the mean of the test and control. The limit of significance was set at p<0.05.

3. Results

A continuous increase in bodyweight was observed in all groups throughout the 4 weeks of the experiment with the exception of the groups fortified with cooked breadfruit. The normal control group and groups fortified with 50% grilled breadfruit showed a significant increase (p<0.05) in bodyweight on the third week when compared to week 0 while a significant increase (p<0.05) with respect to week 1 was observed only in their fourth week. The group fortified with 30% grilled breadfruit when compared with week 0 showed a significant increase (p<0.05) from week 2 to week 4 while a significant increase (p<0.05) was observed from week 3 to week 4 when compared with week 1. The fourth week of the groups of rats fed with feeds fortified with 30% cooked breadfruit and 30% parboiled breadfruit showed a significant increase (p<0.05) in bodyweight when compared with week 1 and week 0 respectively (figure 1).



Figure 1 Bodyweight of normal rats fed with feed fortified with processed breadfruit

Table 1 Results of the weekly bodyweight of the rats fed with feed fortified with cooked, parboiled and grilled Breadfruitseed expressed as mean ± SEM

| | Bodyweight (g) | | | | | | |
|--|----------------|-------------|--------------|---------------|---------------|--|--|
| Groups | Week 0 | Week 1 | Week 2 | Week 3 | Week 4 | | |
| Normal Control | 126.32±3.07 | 131.50±2.00 | 140.62±1.87 | 145.96±2.34a | 156.76±3.31ac | | |
| 70% Std Feed + 30% cooked Breadfruit | 130.50±3.81 | 124.92±3.60 | 131.88±3.86 | 129.08±5.14 | 133.14±4.54c | | |
| 70% Std Feed + 30% Parboiled Breadfruit | 131.72±2.87 | 134.32±1.80 | 138.32±2.20 | 141.10±3.02 | 144.70±2.95a | | |
| 70% + Std Feed + 30% grilled Breadfruit | 128.74±2.70 | 129.00±2.77 | 142.86±2.77a | 146.66±2.88ac | 148.06±2.97ac | | |
| 50% Std Feed + 50% cooked Breadfruit | 129.70±3.06 | 124.04±1.34 | 126.26±12.16 | 124.66±3.64 | 120.86±2.97 | | |
| 50% Std Feed + 50% parboiled Breadfruit | 133.58±3.24 | 138.00±4.24 | 140.48±5.98 | 141.54±9.90 | 143.48±10.56 | | |
| 50% Std Feed + 50% grilled Breadfruit | 125.76±1.54 | 133.38±1.19 | 135.58±2.86 | 137.14±3.61a | 140.08±3.13ac | | |

^aSignificant increase with respect to week 0; ^cSignificant increase with respect to week 1.





| Groups | TCHOL (mg/dl) | HDL (mg/dl) | TRIG (mg/dl) | LDL (mg/dl) | VLDL (mg/dl) |
|--|------------------|---------------------|----------------|----------------------|---------------|
| Normal Control | 340.16±113.75 | 11.13±0.55 | 228.67±98.90 | 283.31±130.86 | 45.73±19.78 |
| 70% Std Feed + 30% cooked Breadfruit | 227.90±51.26 | 16.27±3.37 | 228.04±81.29 | 166.11±55.57 | 45.60±16.26 |
| 70% Std Feed + 30% Parboiled Breadfruit | 336.55±32.03 | 28.53±3.98a | 359.34±204.87 | 236.15±36.31 | 71.87±40.98 |
| 70% + Std Feed + 30% grilled Breadfruit | 318.45±49.79 | 23.97±0.66 <i>a</i> | 236.67±38.61 | 247.12±52.25 | 47.37±7.72 |
| 50% Std Feed + 50% cooked Breadfruit | 271.41±46.67 | 37.09±3.97 <i>a</i> | 294.00±118.53 | 175.52±73.06 | 58.80±23.71 |
| 50% Std Feed + 50% parboiled Breadfruit | 162.85±45.54b | 42.23±2.33a | 367.50±172.01 | 47.12±42.91 <i>b</i> | 73.50±34.40 |
| 50% Std Feed + 50% grilled Breadfruit | 173.70±30.71 | 37.66±3.12a | 571.67±144.56a | 22.21±31.99b | 114.34±28.91a |

Table 2 Effect of feed fortified with cooked, parboiled, and grilled Breadfruit seeds on lipid profile of Wistar rats expressed as mean ± SEM.

^aSignificant increase with respect to normal control; ^bSignificant decrease with respect to normal control.

The total cholesterol level of the experimental rats decreased throughout the groups with significant decrease (p < 0.05) recorded in the group fortified with 50% parboiled breadfruit (figure 2).



Figure 3 High-density lipoprotein concentration of normal rats fed with feed fortified with processed breadfruit.

With the exception to the group fortified with 30% cooked breadfruit which showed a non-statistical increase (p > 0.05) in its HDL levels, all other experimental groups showed significant increases (p < 0.05) in their High-Density Lipoprotein level when compared with the normal control group (figure 3).



Figure 4 Triglyceride concentration of normal rats fed with feed fortified with processed breadfruit.

A significant increase (p< 0.05) in Triglyceride level was only recorded in the group of rats fortified with 50% grilled breadfruit (figure 4).



Figure 5 Low-density lipoprotein concentration of normal rats fed with feed fortified with processed breadfruit.

The Low-Density Lipoprotein level of the experimental rats decreased throughout all the groups with a significant decrease (p< 0.05) recorded in the group fortified with 50% parboiled and grilled breadfruit (figure 5).



Figure 6 Very low-density lipoprotein concentration of normal rats fed with feed fortified with processed breadfruit.

A significant increase (p< 0.05) in Very low-density lipoprotein levels was only recorded in the group of rats fortified with 50% grilled breadfruit (figure 6).

4. Discussion

The continuous increase in bodyweight as observed in all groups could be because of the availability of more nutrients and proteins in their diet leading to a better nutritional status. The lipid profile shows the amount of cholesterol, triglycerides, and lipoproteins present in the blood. These lipids are very important for healthy cells but may be harmful when present in high amounts in the blood. High levels of LDL-*c*, TRIGS, and VLDL-*c* in the blood could lead to inflamed arteries, stroke, and several heart diseases thus these lipids should be at lower concentrations in the body.

The high-density lipoprotein cholesterol (HDL-C) also known to be the good cholesterol is better at a high level in the body because it helps remove excess cholesterol from the body by transporting it to the liver to be flushed out of the body thereby preventing the occurrence of any heart disease [16]. Medicinal plants are known to improve lipid profile parameters which has made them beneficial for consumption for the prevention, management, and treatment of various diseases [17,18,19].

The result of this study however showed a decrease in total cholesterol and low-density lipoprotein levels. This decrease might be a result of the phytomedicinal, mineral, and vitamin constituents of breadfruit. It has earlier been revealed that medicinal plants contain sufficient phytochemicals responsible for their beneficial properties both in nutrition and disease management [20,21].

Triglyceride and very low-density lipoprotein levels were significantly increased (p < 0.05) in the group of rats fortified with 50% grilled breadfruit. This, however, suggests that methods other than grilling should be used in the processing of breadfruit to help minimize the TRIGS and VLDL-*c* levels and insure healthier cells. The HDL levels of all experimental groups increased (p < 0.05) significantly apart from the group fortified with 30% cooked breadfruit when compared with the normal control. This finding agrees with the research carried out by [22,23].

5. Conclusion

The nutritional benefit of any food is maintained by a suitable processing method which may have a certain effect on some biochemical parameters. The result of this research however suggests no adverse effect of feed fortification with processed breadfruit on the lipid profile of the experimental animals but infers caution in the use of grilled breadfruit for feed fortification.

Compliance with ethical standards

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Disclosure of conflict of interest

The authors state that there is no conflict of interest.

Statement of ethical approval

All experiments were approved and supervised by the Nnamdi Azikiwe University Animal Research Ethics Committee (NAU-AREC) in line with the principles of Animal Care and Use in Research, Education and Testing. The ethical approval number as issued by the NAU-AREC is NAU/AREC/2021/00040.

Author's Contributions

O.C.E. and C.F.E designed the study. O.C.E, O.P.A. and G.N.O. performed the literature search and wrote the manuscript. O.C.E, E.L.I, U.V.C and V.N.S. performed the data analysis. All authors read and approved the final manuscript.

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