EFFECTS OF BEAN CAKE COOKED IN DIFFERENT PACKAGING MATERIALS ON HEMATOLOGICAL INDICES AND KIDNEY FUNCTION OF WISTAR ALBINO RATS

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ABSTRACT

This research assessed the effect of bean cake packaged with different materials (Thaumococcus danielle leaf, aluminum foil and plastic plate) on the proximate composition, and the hematological indices and kidney function of Wistar rats. Twenty-five Wistar rats were grouped into five (5 rats per group). Group one was fed with cerelac, group two with commercial rat feed, group three with bean cake packaged in T. danielli leaf, group four with bean cake packaged in aluminum foil and group five fed with bean cake packaged in plastic plate and; all the groups were fed ad libitum for twenty-eight days. The results of the proximate analysis showed that bean cake is rich in carbohydrate (51%), protein (20%) and is a good source of Minerals. There was a significant difference among the Potassium, Sodium, Urea and Creatinine levels of the test animals and the control at p<0.05. However, there was no remarkable difference in the haematological indices of the Control and Test groups. The results of this research showed that packaging materials had no significant effect on haematological indices and kidney function of Wistar rats in 28 days and plastic retained more nutrients than other packaging material.

Keywords: Bean cake (moi-moi), packaging materials, hematological parameters, kidney function, wistar albino rats,

INTRODUCTION

The need for food packaging has existed for as long as humans have prepared and stored food. Initially, humans consumed whatever food they gathered from their surroundings but as they began staying in sheltered areas, the need arose for food storage. Historically, shells, gourds and leaves of plants were used as basic form for food packaging; also grasses, wood and bamboo were used to weave baskets (Risch, 2009). However, due to the limited shelf life of food packaged in leaves, there was a need for more effective packaging materials. As the food packaging industries evolved, so did the materials used to package food, leading to the wide range of options available today.

Modern food packaging materials not only contain food but also provide consumers with important information about the food within, including nutritional content and ingredients. Furthermore, packaging materials can serve as means of advertising to consumers.

Despite the benefits of these materials, many modern food packaging materials are made with synthetic chemicals that can negatively impact human health. These chemicals can migrate from the packaging material to the food in a process known as chemical under migration, especially certain environmental conditions (Muncke, 2013). For instance, certain chemicals such as per/polyfluoroalkyl substances (PFAS), phthalates, and bisphenol Aused in plastic food containers have been linked to various hormonal health concerns (Giuliani et. al. 2020).

Bean cake (moimoi), a popular Nigerian food made from ground bean paste and other ingredients, requires packaging to ensure its preparation, preservation and transport. The ground paste is mixed with other ingredients such as ground pepper, seasoning cubes, crayfish, ground onion etc, then it is put into packaging materials such as plastic, aluminum foil, leaves, and then cooked with steam. In the past, leaves of broad-leaved plants like bananas, Thaumatococcus. daniellii and plantain were used as packaging materials. However, these materials had a short shelf life. Consequently, people have turned to more modern packaging materials, such as aluminum foil and plastic plates, which are more hygienic and efficient. However, using these materials raise concerns over their potential impact on human health, given their propensity to leach harmful chemicals into food. Therefore, there exists, the need to explore alternative packaging materials that are more health-friendly and environmentally sustainable.

MATERIALS AND METHOD

Experimental Design

Twenty-five Wistar albino rats weighing between 50-60g were purchased from the Department of Pharmacology Animal House at the University of Port Harcourt, Choba, Rivers State. The Wistar rats were equalized as nearly as possible with respect to body weight and grouped into five groups with five rats per group and acclimatized for seven days. Group 1 was fed with cerelac and served as the positive control (+ve control), group 2 was fed with commercial rat feed and served as the negative control (-ve control), group 3 was fed with bean cake cooked in T. daniellii leaves, group 4 was fed with bean cake cooked in aluminum foil, group 5 was fed with bean cake cooked in plastic plate. The rats were fed ad libitum for twenty-eight days. At the end of the twenty-eight days, the rats were sacrificed, and biochemical analysis (haematology, kidney function and proximate analysis) were carried out using their blood.

Groups	Feed
1 (positive control)	Cerelac
2 (negative control)	Commercial rat feed
3	Bean cakepackaged in Thaumatococcusdaniellii leaves
4	Bean cakepackaged in aluminum foil
5	Bean cakepackaged in plastic plate

Table	1:	Ex	perimental	Groups
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Preparation of bean cake

Raw bean seeds (*Vignaunguiculata*) were purchased from Choba market, Choba, Rivers State. The method of Ezeocha et al. (2021) was used in the preparation of the bean cake with slight modification. Dirt was removed from the beans and the latter were soaked in water for 10 min, the bean coat was removed manually by rubbing against the palm. The beans (600g) were then wet-milled with pepper (20g) and onions (25g) into a paste, and then salt (1.5g), seasoning cube (2g), vegetable oil (25ml) and water were mixed into the paste. This mixture is the unpackaged paste. The mixture was put into the different packaging materials (aluminum foil, plastic plate and *Thaumatococcusdanielli* leaves) and steamed in a pot for 30 min at 80° C. the mixture of the ground beans with the

Proximate Analysis

Protein content was determined using the kjedahl method, carbohydrate content was determined using the Cleganthrone method, moisture content was determined through the Air oven method, determination of lipid content was done by soxhletextration method, crude fibre was determined through the Weende's method and determination of ash contentwas done by furnace method.

Determination of energy content

The energy content of the packaged and unpackaged bean cakes was calculated by multiplying the mean values of crude protein, crude fat and total carbohydrate by the At water factors of 4, 9, 4 respectively, taking the sum of the products and expressing the results in kilocalories per 100g sample as reported by Onyeike and Acheru (2002)

Determination of Haematological indices

The PCV was read using a microhematocrit reader.To obtain the hemoglobin value, the PCV value was divided by 3 HB units (HB unit: mg/dL).

Determination of Total white blood cell (WBC) count using Neubauer

Diluting fluid of 0.38ml was pipetted into test tubes, then 0.02ml of well-mixed EDTA blood was added and then mixed. The dilute blood sample was remixed using a Pasteur pipette, then one of the grids of the chamber was filled with sample. The chamber was left undisturbed for 20 minutes to allow the white blood cells to settle. The cells were observed and counted using x10 objective lens.

WBC count (per litre) =
$$\frac{N \times Df \times 10^6}{A \times D}$$

where;

N represents the number of cells counted.

DF denotes the dilution factor.

A indicates the area that was counted.

D represents the depth of the chamber

Determination of Platelet count using Neubauer

Diluting fluid (ammonium oxalate) of 0.38ml was pipetted into test tubes, then 0.02ml of well-mixed EDTA blood was added and then mixed. The counting chamber was filled with the sample. The chamber was left unperturbed for 20 min, to prevent drying of the fluid, then; the chamber was placed in a Petri dish on dampened tissue and covered with a lid. The cells were observed and counted using a x10 objective lens.

platelet count (per litre)
=
$$\frac{N \times 20 \times 10^{6}}{0.2 \times 0.1}$$

Determination of Red blood cell (RBC) count using Neubauer

Formal citrate (diluting fluid) of 4ml was dispensed into a test tube; 0.02ml of wellmixed EDTA blood was added and mixed. The sample was added to the counting chamber and subsequently left without any disturbance. The cells were observed and counted using a x10 objective lens

$$RBC \ Count = \frac{N \times 201 \times 10^9}{0.2 \times 0.1}$$

where;

N represents the number of cells counted. 201 is the dilution factor.

 0.2 mm^2 is the area that was counted.

0.1 mm is the depth of the chamber.

Determination of differentials using Leishman staining technique

Procedure: the slide was covered with undiluted Leishman stain and allowed for 2 min. Twice the volume of pH 6.5 buffered water was added to the stain, the dilute stain was blown on to ensure the water mixes well with the stain. Then it was allowed for 8 min after which, the slide was washed off with tap water and left in a draining rack for the smear to dry. The slide was then examined using oilimmerse objective lens.

Kidney Function Test

The direct end point method of creatinine determination was adopted as described by Ochei and Kolhatkar (2008). Determination of bicarbonate (HCO₃) concentration was done using Back titration method. The serum concentration of sodium was determined with Randox sodium test kit using the method of Maruna and Trider as described by Ochei and

Kolhatkar (2008). The concentration of serum potassium was determined using Randox test kit. The method of TietsN.W was adopted as described by Ochei and Kolhatkar (2008). The serum chloride concentration was determined using Randox chloride test kit. The method of Levinaon S. S. was adopted as described byOchei and Kolhatkar (2008). The serum urea concentration was determined with Randox urea test kit based on the method of Berthelot described bvOchei and Kolhatkar as (2008).All procedures were carried out as outlined by the manufacturers of the test kit.

RESULTS AND DISCUSSION

 TABLE 2: Proximate composition of the bean cake samples

Sample packaged with	Moisture (%)	Ash (%)	Carbohydrate (%)	Crude Protein (%)	Lipid (%)	Crude Fibre (%)	Energy content (Kcal/100g sample)
Unpackaged	$74.04^{a}\pm0.50$	$0.59^{b}\pm0.88$	$16.76^d\pm0.15$	$5.73^{c}\pm0.22$	$2.29^{d} \pm 0.04$	$0.58^{\rm c}\pm0.09$	110.57
Leaves	$6.10^{b}\pm2.0$	$2.38^{a}\pm0.53$	$54.39^b\pm0.06$	$19.84^{b}\pm0.25$	$5.00^{\rm c}\pm0.00$	$12.95^{a}\pm2.08$	341.92
Aluminum	$5.30^{b} \pm 1.20$	$2.40^{a}\pm0.80$	$51.01^{\rm c}{\pm}~0.00$	$20.13^{ab}{\pm}\ 0.00$	$8.30^{a}\pm0.005$	$12.19^{a}\pm2.30$	359.26
foil Plastic plate	$3.40^b \pm 0.20$	$3.26^{a}\pm0.49$	$59.72^a\pm0.00$	$20.37^a\pm0.22$	$7.10^b \pm 0.005$	$6.49^{b}\pm0.99$	384.26

Values are means \pm standard deviation of triplicate determinations. Values in the same column having the same superscript letters are not significantly different at the 5% level.

TABLE 3: Result of haematology profile of test and control rats

	L(%) = E(%) = M(%)
$ 2.30 11.42^{b} \pm 0.78 6.04^{b} \pm 0.20 7.22^{b} \pm 1.24 490.00^{a} \pm 110.14 13.00^{a} = 10.14 13.00^{a} = 10.14 $	$\pm 1.00 \qquad 82.20^{a} \pm 3.11 \qquad 2.20^{a} \pm 0.44 \qquad 4.00^{a} \pm 1.22$
$2.07 12.70^{a} \pm 0.75 6.60^{a} \pm 0.29 13.76^{a} \pm 2.45 516.00^{a} \pm 165.14 14.80^{a}$	$\pm 5.71 \qquad 79.40^{a} \pm 9.28 \qquad 1.80^{a} \pm 1.09 \qquad 3.20^{a} \pm 2.48$
$1.58 \hspace{0.5cm} 12.90^{a} \pm 0.53 \hspace{0.5cm} 7.02^{a} \pm 0.48 \hspace{0.5cm} 9.82^{ab} \pm 3.09 \hspace{0.5cm} 543.00^{a} \pm 87.34 \hspace{0.5cm} 12.20^{a}$	$\pm 1.64 \qquad 83.40^{a} \pm 4.72 \qquad 1.40^{a} \pm 0.54 \qquad 3.20^{a} \pm 2.16$
$3.04 13.08^{a} \pm 0.64 6.90^{a} \pm 0.22 10.88^{ab} \pm 2.95 476.60^{a} \pm 12.05 16.60^{a}$	$\pm 2.50 \qquad 77.40^{a} \pm 3.43 \qquad 1.60^{a} \pm 0.54 \qquad 4.40^{a} \pm 0.54$
$2.07 12.44^{ab} \pm 0.54 6.86^{a} \pm 0.28 9.00^{ab} \pm 3.36 475.40^{a} \pm 35.71 14.80^{a}$	$\pm 1.64 \qquad 79.60^{a} \pm 1.14 \qquad 1.60^{a} \pm 0.54 \qquad 3.40^{a} \pm 1.34$
2.30 $11.42^{b} \pm 0.78$ $6.04^{b} \pm 0.20$ $7.22^{b} \pm 1.24$ $490.00^{a} \pm 110.14$ 13.00^{a} 2.07 $12.70^{a} \pm 0.75$ $6.60^{a} \pm 0.29$ $13.76^{a} \pm 2.45$ $516.00^{a} \pm 165.14$ 14.80^{a} 1.58 $12.90^{a} \pm 0.53$ $7.02^{a} \pm 0.48$ $9.82^{ab} \pm 3.09$ $543.00^{a} \pm 87.34$ 12.20^{a} 3.04 $13.08^{a} \pm 0.64$ $6.90^{a} \pm 0.22$ $10.88^{ab} \pm 2.95$ $476.60^{a} \pm 12.05$ 16.60^{a} 2.07 $12.44^{ab} \pm 0.54$ $6.86^{a} \pm 0.28$ $9.00^{ab} \pm 3.36$ $475.40^{a} \pm 35.71$ 14.80^{a}	$\begin{array}{cccccccccccccccccccccccccccccccccccc$

Values are means \pm standard deviation of triplicate determinations. Values in the same column having the same superscript letters are not significantly different at the 5% level.

 Table 4: Serum kidney function of Wistar albino rats fed with bean cake packaged in leaf, aluminum foil and plastic plate

Sample	Potassium	Sodium (mmol)	Urea	Creatinine	Chloride	Bicarbonate
	(mmol)		(mmol)	(mmol)	(mmol)	(mmol)
+ve	$6.16^{a} \pm 0.24$	$169.60^a\pm5.31$	$4.52^{b}\pm0.48$	$91.40^b\pm 6.87$	$66.80^a\pm 6.37$	$22.20^{a}\pm1.92$
control						
-ve control	$3.76^{c}\pm0.16$	$121.40^{\circ} \pm 1.94$	$4.32^b\pm0.13$	$88.60^b \pm 1.81$	$58.20^{ab}\pm3.42$	$26.00^{a}\pm1.58$
Leaf	$4.26^{bc}\pm0.43$	128.80 ^{bc} ±	$7.00^{ab}\pm1.73$	$142.60^{a} \pm 18.84$	$55.80^{b}\pm1.92$	$24.80^a\pm3.03$
		10.56				
Foil	$4.94^b\pm0.56$	$146.20^{b} \pm 11.30$	$6.36^{a}\pm0.25$	$126.00^a\pm4.18$	$63.80^{ab}\pm5.16$	$25.40^a\pm3.36$

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Plastic	$4.22^b\pm0.19$	$128.40^{b} \pm 2.70$	$6.62^a \pm 0.43$	$129.60^{a} \pm 8.96$	$61.20^{ab} \pm 5.26$	$25.60^{a} \pm 2.30$	

Values are means \pm standard deviation of triplicate determinations. Values in the same column having the same superscript letters are not significantly different at the 5% level.

DISCUSSION

The result of the proximate content of the samples is shown in Table 1.

Proximate content of food refers to the basic components or nutrients present in the food that are essential for the body's functioning and energy needs. They include; carbohydrates, lipid, protein, moisture and ash.

The unpackaged bean paste had very high moisture content, 74.04 %, compared to the packaged bean cake; the bean cake packaged in leaf had a moisture content of 6.10%, foil 5.30% and plastic with the lowest moisture content of 3.40%, this large difference in moisture content was as a result of the cooking and drying process the packaged bean cake underwent. There was no significant (p > 0.05)difference in the moisture content of the packaged bean cake. The low moisture content of the sample indicates that it has a long shelf life. Okwunodulu et al. (2019) reported a moisture content of 55.10% and 50.10% for bean cake wrapped in aluminum foil and bean cake wrapped in Thaumatococcusdaniellii leaf respectively. This difference is likely due to the state of the sample at the point of testing (dry versus fresh sample).

Ash refers to the inorganic minerals that remain after a food sample is completely burned (Nielsen, 2017). It is a representation of the food's mineral content, which includes important substances like calcium, magnesium, potassium, and others (Sarmila, 2023).

The ash content of bean cake packaged in leaf was the lowest 2.38%, though it was not significantly (p > 0.05) different from the other packaged bean cake; a similar observation was reported by Ezeocha et al. (2021). This could be a result of minerals leaching out of the leaf

into the cooking water as Onwuka, 2018 (cited by Ezeocha et al. 2021) mentioned, ash content represents the mineral content of the sample after oxidation of organic matter.

Carbohydrate provides energy to the body. The carbohydrate content of the packaged bean cake ranged from 54.39-59.72% and that of the unpackaged bean cake was 16.76%. There was a significant (p < 0.05) difference between the various samples with the unpackaged sample having the lowest carbohydrate content of 16.76% and bean cake packaged in plastic plate having the highest content of 59.72%. This is higher than 14.74-16.07% reported by Ezeocha et al. (2021). This could be as a result of a difference in recipe and difference in the specie of beans used. The carbohydrate content of bean cake packaged in aluminum foil, (51.01%), was close to 52.06% reported by Emelike et al. (2020).

Crude protein content of the unpackaged beans paste was 5.73% which was much lower than that of the packaged bean cake. Protein content of the packaged bean cake varied from 19.84 to 20.37%. There was a significant difference between bean cake packaged in leaf, (19.84%), and plastic plate, (20.37%). The value obtained was similar to the 21.89 reported by Akusu and Kiin-Kabari (2012), for bean cake wrapped in aluminum foil.

Lipid content of the packaged sample ranged from 5.00-8.30%, with aluminum foil having the highest value and leaf having the lowest value and each sample was significantly (p <0.05) different from the other. The low lipid content in the bean cake packaged with leaf is also attributable to the poor barrier properties of the leaf, thus water can easily leach out of the sample taking along some fat with it. Okwunodulu et al. (2019) also reported lipid content of baked bean packaged in aluminum foil (4.64%) being greater than those packaged in Thaumatococcusdaniellii (uma) leaf (4.24%).

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Crude fiber analysis gives details on the sample's dietary fiber content, which is crucial for evaluating the sample's nutritional value, digestive health advantages, and regulatory compliance with food labeling requirements (Sarmila, 2023b). Crude fibre content of bean packaged in plastic cake plate was significantly lower (6.49%) than the other packaged bean cake samples (12.95% for leaf and 12.19% for aluminum foil). Ezeocha et al. (2021) also reported a similar trend of the fibre content of bean cake packaged in plastic plate being less than those packaged in aluminum foil and Thaumatococcusdaniellii leaf.

The packed cell volume of the animals in both the test and control groups was well within the normal range. Delwatta et al. (2018) reported the packed cell volume of both male and female rats as 39.38 ± 7.6 and 36.54 ± 8.5 respectively. Hemoglobin levels from this study (12.70 g/dl – 12.44 g/dl) were within the normal range of 13.63 g/dl for male, 12.86 g/dl for female and 14.6 g/dl for male and 13.8 g/dl for female reported by Delwatta et al. (2018).

Red blood cell count of the test animals $(6.86 \times 10^{12} - 7.02 \times 10^{12})$ were not significantly different from the control (6.04×10^{12}) and 6.60×10^{12}). Red blood cells, also known as erythrocytes, are an important component of blood that transports oxygen throughout the body; since these cells are biconcave discs with no nucleus, they have a higher surface area for oxygen exchange (Thiagarajan et al., 2021). Delwatta et al. (2018) reported a red blood cell count of $5.26 \times 10^6 / \mu L$ for male rats and $5.16 \times 10^6 / \mu L$ for female rats which is also similar to the results of this study.

The white blood cell count obtained in this study $(7.22 \times 10^{12} - 13.76 \times 10^{12})$ is in corroboration with that of Delwatta et al. (2018) who reported 9142 (per mm³) for male rats and 7733 (per mm³) for female rats. There was no significant difference among the groups. White blood cells (WBCs), also known as leukocytes, are an important component of the immune system that helps the body fight illnesses and foreign substances (National Cancer Institute, 2023).

The platelet count of the leaf group was the highest at 543.00×10^9 while plastic group had the lowest platelet count at 475.40×10^9 . No significant difference was observed, (p>0.05) among all the groups. Delwatta et al. (2018) reported $3.44 \times 10^5/\mu$ L for male rats and $3.28 \times 10^5/\mu$ L for female rats, these values are lower than those obtained in this study.

The neutrophil counts ranged from 12.20-16.60%, this value is lower than those reported by Delwatta et al. (2018) (22.64% for male rats and 24.79% for female rats). Neutrophils are a type of white blood cell, often known as leukocytes, which play an important role in the immune system. They are the most common form of white blood cell and serve an important function in protecting the body from bacterial and fungal illnesses (Tigner, 2022).

The values of the lymphocyte count ranged from 77.4- 83.40%, foil had the lowest while leaf had the highest. The values of the lymphocyte count is within range compared to Delwatta et al.'s work (2018) which reported 75.17% for male rats and 72.87% for female rats. Lymphocytes are a type of white blood cell that plays an important role in the immune system. They are responsible for providing immunity to certain pathogens such as bacteria, viruses, and other foreign substances and play a critical role in the body's defense against diseases (Tigner, 2022).

The values of eosinophil count ranged from 1.4% to 2.20%, and these values are similar to those reported by Delwatta et al. (2018) 2.17% for male rats and 2.04% for female rats. There was no significant difference among the entire group. Eosinophils are white blood cells that play an important part in immunological response to allergen and parasite. They are distinguished bv characteristic granules containing certain enzymes and proteins that give them a brilliant red or orange appearance when stained with eosin dye, thus the name "eosinophil." (Saladin, 2012).

The values from this research (3.20% - 4.40%) were far from that reported by Delwatta et. al (2018), 0.18% for male rats and 0.16% for

female rats. Monocytes are a type of white blood cell; they play a crucial role in defending the body against infections and contribute to the body's inflammatory response (Tigner, 2022). There was no significant difference among all the groups.

The kidney's primary duties are to eliminate metabolic wastes, and preserving water, pH, electrolyte balance, as well as producing calcitriol and erythropoietin (Damodaran et al., 2016). Electrolytes are necessary for the fundamental processes of life, which includes the initiation and conduction of action potentials in nerves and muscles, as well as the preservation of electrical balance within cells (Shrimanker & Bhattarai, 2022). In this study, the levels of serum potassium, chloride and sodium was evaluated, as well as the levels of serum urea, creatinine and bicarbonate. The results obtained are discussed below.

The level of serum potassium was highest in the group fed with cerelac (positive control), the test animals all had similar potassium levels (4.22 mmol - 4.94 mmol), and this could be as a result of the difference in the diet, therefore it can be implied that the packaging materials had no effect on the serum potassium levels of the Wistar rats. The primary function of potassium in the body is to support the maintenance of normal fluid levels inside cells, thus it is an intracellular ion (Shrimanker & Bhattarai, 2022). Increasing the intake of potassium can help reduce blood pressure and thus reduce the risk of cardiovascular diseases such as hypertension and stroke ("Sodium, Potassium and Health," 2022). The range of normal serum potassium concentrations is 3.6 - 5.0 mmol/L; ingesting about 4700 mg of potassium each day is necessary to maintain appropriate potassium levels (Kowey, 2002; Reid, 2017).

The sodium level of the test rats ranged from 128.80 mmol - 146.20 mmol. However, no significant difference was observed among the test groups. Since the sodium levels of the test animals was not significantly different, the kind of packaging must have had no effect on the sodium level, the distinction between the

test groups and the control can be attributed to the difference in diet. Sodium is an extracellular ion; it controls the modulation of the membrane potential of cells as well as the extracellular fluid volume (Shrimanker & Bhattarai, 2022). Serum sodium levels below 135mmol would lead to hyponatremia while sodium levels above 145mmol would cause hypernatremia (Shrimanker & Bhattarai, 2022).

It was observed that the concentration of urea in the blood of the test rats (6.36 mmol - 7mmol) was significantly higher than those of the control (4.32 mmol - 4.52 mmol). This could be as a result of the high protein content of beans which the test animals were fed with. Urea is a waste product of protein metabolism. Deamination of amino acids from protein catabolism produces ammonia which is then converted to urea via the urea cycle (Salazar, 2014). Therefore, protein consumption affects the concentration of urea in the blood.

Theserum concentration of creatinine obtained from this study (88.6 - 142.6 µmol/l) is way higher than that from the study carried out by Delwatta et al. (2018) which reported a reference value of 43.36 µmol/l for male rats and 46.01 µmol/l for female rats. This difference could be as a result of a difference in environmental conditions, hormonal status and breeding. It is again observed, that the test groups were not significantly different and their blood creatinine concentration was significantly higher than that of the control. The fact that the creatinine concentration was similar for the test rats implies that the type of packaging material has no effect on the creatinine level. Serum creatinine is not affected by diet as it is dependent on the total muscle mass of the body (Damodaran et al., 2016).

There was no significant difference in the serum chloride concentration of the test groups and the control groups, except for leaf group (55.80 mmol) which was significantly different from positive control (66.80 mmol). This is an indication that the packaging material did not affect the serum chloride concentration of the Wistar albino rats. Chloride is the primary anion in the extracellular fluid of the body; it is mainly diet (McCallum, 2013: gotten from Pfortmueller et al., 2018). Chloride is excreted by the kidneys and the amount excreted into the urine (i.e., not reabsorbed by the tubules of the nephron) is variable and depends on the objective of the kidney, whether it is trying to conserve or eliminate chloride; despite significant daily fluctuations in chloride intake, the kidneys' capacity to modify daily chloride excretion maintains total body chloride values roughly constant and serum chloride concentrations within a specific range (Berend et al., 2012). The normal serum concentration of chloride is 96-106 mmol/L (Berend et al., 2012).

The concentration of bicarbonate in the sera of both test and control animals was not significantly different (22.20 mmol - 26.00 mmol); this implies that the packaging material had no effect on serum bicarbonate concentration of Wistar albino rats. The second-largest component of anions in plasma, after chloride, is bicarbonate. This fraction also contains the carbamino compounds and carbonate (CO^{2}_{3}) , in addition to bicarbonate (HCO_3^{-}) (McCallum, 2014). When the body is unable to maintain acid-base balance by excreting organic acids and hydrogen ions while retaining bicarbonate ions, metabolic acidosis develops (Beynon-Cobb, 2023). The normal range of bicarbonate in the serum is 23 mmol – 29 mmol.

CONCLUSION

In conclusion, the proximate composition, the haematological indices and kidney function of the Wistar rats fed with packaged bean cake was not significantly different from each other and in some cases not significantly different from the control. This suggests that the type of packaging material had no impact on these parameters of the Wistar rats in 28 days.

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