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PRELIMINARY ASSESSMENT OF THE ACUTE TOXICITY EFFECTS OF FRIED CAT FISH EXTRACT ON BIOCHEMICAL, HEMATOLOGICAL AND HISTOPATHOLOGICAL PARAMETERS OF WISTAR RATS

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ABSTRACT

Some food processing techniques have been linked with formation of toxic chemicals known as Polycyclic Aromatic Hydrocarbons (PAHs), which are found in some processed foods and have been widely attributed to the prevalence of cancer of liver and stomach in Nigeria. However, the toxicological effects of the consumption of fried fish have not been adequately documented. Therefore, this study investigated the toxicological effects of PAHs in fried cat fish on Wistar rats. Fifty five rats were used in the study and they were divided into control and ten treatment groups with daily administration of fried fish extract at dosage of 200, 400, 600, 800 and 1000 mg/kg body weight to the rats for 14 days. The results showed that the activities of serum, alanine aminotransferase (ALT), alkaline phospherase (ALP), asparte aminotransferase (AST) as well as urea and creatinine concentration were affected across the treated groups of animals which were indications for hepatorenal damage. White blood cell (WBC) and mean corpuscular volume (MCV) showed significant changes in response to the administered dosage compared to the control. The liver showed no significant histopathological changes between the test rats and control. The architecture of sections of kidney treated groups was similar to that of the control in which the renal corpuscles maintained their normal size of urinary space and normal tubular structures with no necrosis observed. The result revealed that there can be potential toxicity if there is repeated indulgence to fried cat fish at the temperatures between 160 – 230 ºC.

Key words: frying, toxicological, wistar rats, dosage, hepatorenal damage

INTRODUCTION

Food pollution by PAHs is due to deposition of these compounds from air or water or products formed from preservation, drying and cooking processes [1]. The level of PAHs in food increases as a result of cooking steps such as roasting, frying and smoking being employed [2]. Health effects from chronic or long-term exposure to PAHs may include decreased immune function, cataracts, kidney and liver damage (e.g. jaundice), and breathing problems, asthma – like symptoms, and lung function abnormalities. And repeated contact with skin

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may induce redness and skin inflammation [3]. Naphthalene, a specific PAH, can cause the breakdown of red blood cells if inhaled or ingested in large amounts. Many PAHs are only slightly mutagenic or even non-mutagenic in vitro. However, their metabolites or derivatives can be potent mutagens [4].

It is difficult to ascribe observed health effects in epidemiological studies to specific PAHs because most exposures involved PAH mixtures. Increased incidences of lung, skin, and bladder cancer are associated with occupational exposure to PAHs [4]. Epidemiologic reports of PAH-exposed workers have noted increased incidences of skin, lung, bladder, and gastrointestinal cancer. These reports however provide only qualitative evidence of the carcinogenic potential of PAHs in humans because of the presence of multiple PAH compounds and other suspected carcinogens. Some of these reports also indicate the lack of quantitative monitoring data [4].

Embryotoxic effects of PAHs have been described in experimental animals exposed to PAHs such as benzo (a) anthracene, benzo (a) pyrene, and naphthalene. The laboratory studies conducted on mice have demonstrated that ingestion of high levels of benzo (a) pyrene during pregnancy resulted in birth defects and decreased body weight in the offspring. It is not known whether those effects can occur in humans. However, the centre for children's environmental health reports studies that demonstrated that exposure to PAH pollution during pregnancy is related to adverse birth outcomes including low birth weight, premature delivery, and heart malformations. High prenatal exposure to PAH is also associated with lower 1Q at age three, increased behavioral problems at ages six and eight, and childhood asthma. Cord blood of exposed babies shows DNA damage that has been linked to cancer. [5].

The high level of contamination by PAHs when smoking is done using the traditional methods is as a result of exposure to the wood/charcoal combustion fumes [6]. In Nigerian, necessary attention has not been given to the various methods of processing fish and the attendant health challenges to the consumers of fish. Many factors attached to the methods of processing of fish alter the composition of smoked fish as well as the total PAHs present in the products, with the processing temperature playing critical role [7, 8, 9]. To the best of our knowledge there is little or no investigation into the toxicological effects of frying fish on health of consumers of fried fish, yet processed fishes are one of the major delicacies of Nigerian diets. It was on this basis that, this study investigated the acute toxicity effects of PAHs in fried cat fish on biochemical, hematology and histopathology of liver, heart and kidney of Wistar rats.

MATERIALS AND METHODS

Collection of Samples and Preparation

The samples of locally consumed *Arius heudeloti* (cat fish) used in this study were bought from

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local vendor in Ogbomoso, Oyo State, Nigeria. The fish were authenticated in the Department of Pure and Applied Biology Ladoke Akintola University of Technology, Ogbomoso. The fish sample was degutted and washed thoroughly with deionized water. The samples were fried at two different temperatures of 160 and 230 ^{o}C for 12 mins using vegetable cooking oil.

Animals and Animal Husbandry

Fifty five (55) healthy outbred Wistar albino rats used in this study were obtained from the primate colony of the Department of Biochemistry, Federal University of Technology Akure, Nigeria. The rats were about 9 to 10 weeks old when obtained. The animals were housed in polypropylene cages lined with hard wood bedding. Rats were fed on commercial pelleted diet and drinking water through 1-qt gravity-fed feeders and waterer. The rats were allowed to acclimatize for 7 days before the toxicity testing, after which the animals were assigned into eleven groups of five rats per group (n=5), making up of one control and ten experimental groups. The room temperature for the animals was maintained at 29^{o}C and overhead incandescent illumination was maintained on 12-h light – dark cycle. Daily maintenance was conducted during the first quarter of the light cycle.

Extract Administration

For acute toxicity test, each group of animals was given different doses of aqueous extract of fried cat fish (FCF) orally using intra-gastric catheter. The extracts were given after the animals were fasted of food for 16 h with free access to water. After the period of fasting, the animals were weighed and the dose calculated according to the body weight, the extract was administered according to the method described by Dapar *et al.*, [10].

Acute toxicity study

The lethal dose for fifty percent (LD_{50}) for FCF was determined using male Wistar rats. A total of five animals per group were used for each dose level investigated. Ten test and one control groups were used for the study. The dosages delivered to treatment animals groups by oral administration were 200, 400, 600, 800 and 1000 mg kg^{-1} body weight of the fried cat fish (FCF), fried at 160 ^{o}C and 230 ^{o}C. Throughout the duration of the test, animal body weight, food and water consumption, behavioral alterations and clinical signs were monitored daily

Body weight measurement

Body weight of all experimental animals were taken by using digital electronic balance before commencing the administration treatment and till the last day of oral administration of the extracts.

Observations of the animals

All the study animals were observed individually during the first 30 mins after dosing and periodically during the first 24 h and then daily before and immediately after administration of the extract.

Blood collection

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At the end of treatment for fourteen days, all the experimental animals were fasted overnight and were sacrificed by cervical dislocation and blood samples were collected by cardiac puncture into tube with anticoagulant ethylene di-amine tetra acetic acid (EDTA) for hematology and into a tube without anticoagulant for the blood chemistry. Blood samples in test tubes containing EDTA were processed for hematological examination. Packed cell volume (PCV), differential leucocytic counts (DLC), red blood cell count (RBC), the hemoglobin (Hb) concentration, mean corpuscular volume (MCV), mean corpuscular Hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) were determined.

For biochemical analysis, the blood samples in the plain test tubes were allowed to stand for 3 h for complete clotting and then centrifuged at 4500 rpm for 10 mins. The plasma was withdrawn and transferred into another clean vial. The sera were kept in the refrigerator until analysis for clinical biochemistry measurements. Concentrations of alanine aminotransferase (ALT), aspartate aminotransferases (AST,), alkaline phosphatase (ALP), albumin, total protein, urea and creatinine were determined using diagnostic kits (Randox Laboratories Limited).

Organ weight and tissue sample

After sacrificing, the animals were dissected, and the target organs of study (liver, kidney and heart) were removed from the body and were kept in 1 % normal saline for a few mins to clean off any extraneous tissue. These organs were then weighed with precision balance, and the tissue samples taken from them. The sample tissues were placed in a test tube containing 10 % buffered formalin for 24 h and thoroughly rinsed over running water. Thereafter the fixed tissues were dehydrated and cleared in a graded series of ethanol and xylene respectively. The tissues were infiltrated with molten paraffin wax and embedded in paraffin blocks. Ribbons of the tissue sections were gently collected using forceps and laid onto the surface of a water bath heated at 30 to 40 ^{o}C. After the sections were thoroughly spread on the water bath, they were placed over tissue glass slides. The slides were then arranged in slides racks and placed in an oven at a temperature of 30 to 40 ^{o}C overnight to facilitate the fixation of the specimen onto the glass slides. The thin sections were passed through different stages of xylene and alcohol, being stained with haematoxylin and eosin (Forysth, 2005). The slides were coded and examined by a histopathologist who was blinded to the treatment groups.

Histopathology examination

The stained sections of the liver, kidney and heart were carefully examined under binocular compound light microscopic. Tissue sections from the treated groups were examined for any evidence of histopathological changes with respect to that of the control. After examination, photomicrograph of selected slides

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from both the treated and control groups were taken under a magnification of X40 and X20 objectives by using automated built in digital photo camera.

RESULTS AND DISCUSSION

Clinical signs in animals

No adverse clinical manifestation e.g diarrhea, hematuria, restlessness etc. were seen in the experimental animals during the dosing period.

Body weight gain

All the treated animals recorded a progressive gain in body weight throughout the entire duration of the treatment compared to control; this may be as a result of the dosage being proteinous. These body weights were not significantly different from the control group.

Acute toxicity

Oral administration of fried catfish (FCF) aqueous extract at different doses of 200, 400, 600, 800 and 1000 mg kg^{-1} did not produce any sign of morbidity and mortality in the experimental animals during the period of experiment for acute toxicity.

Extract effects on biochemical parameter

The averages of albumin and total protein levels in the control group were 0.39 ±0.11 mg/dl and 0.83 ± 0.17 mg/dl respectively (Table 1), but significantly increased following treatment with 200 – 1000 mg kg^{-1} of fried catfish (FCF) processed at 160 ^{o}C and 230 ^{o}C. This study showed that albumin and total protein levels were slightly increased due to the feeding of the test rats with extracts from fried catfish. The increment in albumin levels as observed in the present study is in contrast with earlier report of decrease in the albumin levels of catfish fed with oil contaminated diet [11] Increase in the levels of total protein is possibly a reflection of the increased albumin levels which is as a result of the proteinous nature of FCF used for the rat's treatment.

The Hearts of test rats showed a significant decrease in the creatine, while the kidney showed an increase in creatine. Liver showed an increase in cretene level for 200 mg kg^{-1} FCF 160 ^{o}C and 1000 mg kg^{-1} FCF 230 ^{o}C but decreased for all other dosage treatment. The serum showed a decrease in the createne level for dosage treatment. Heart, kidney, liver and serum showed a significant decrease in urea. For AST, the heart, kidney, liver and serum also showed a significant and progressive increase in the AST. Similarly, heart, kidney, liver and serum showed significant increase for ALT and ALP compared to the control group.

There was increase in the weight of treated rats compared to the control showing that the extract from FCF did affect the mean weight of the rats. The organs weights were also increased and this may indicate the toxic

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potentials, [12] reported that the increased liver weight of the high dose rats could be in response to hepatic enzyme induction, which is typically associated with hepatocellular hypertrophy and transient hepatocyte hyperplasia.

Table 1: Results of biochemical analysis of albumin and total protein

Animal group	Albumin	Total protein
1A	0.52 ± 0.18	0.95 ± 0.45
1B	0.35 ± 0.25	0.52 ± 0.13
2A	0.72 ± 0.31	1.13 ± 0.24
2B	0.62 ± 0.12	1.09 ± 0.55
3A	0.95 ± 0.29	1.31 ± 0.10
3B	0.77 ± 0.15	1.22 ± 0.82
4A	1.14 ± 0.42	1.67 ± 0.35
4B	1.08 ± 0.18	1.49 ± 0.21
5A	1.14 ± 0.22	1.73 ± 0.15
5B	1.22 ± 0.50	1.71 ± 0.19
Control	0.39 ± 0.11	0.83 ± 0.17

1A and B 200 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively
2A and B 400 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively
3A and B 600 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively
4A and B 800 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively
5A and B 1000 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

Effects of ALT, ALP, AST Urea and Creatinine of the treated rats

Repeated dosing of extract from FCF to rats did not affect both food and water consumption as food consumption of all the treatment groups were similar with a non-significant suppression observed in the high- dose group. Plasma biochemical analysis indicated that the liver,

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heart and to a lesser extent, the kidney were target organs for the potential toxicity by the FCF dosage.

Sero-clinical markers, including ALT, ALP, AST, Urea and Creatinine were significantly affected across all the treatment groups as shown in Figures 1 to 5. This result is in agreement with [13] that reported an increase in ALP activity in rats fed with diet formulated with simulated bitumen leachate. But this is in contrast with that of [14] which reported that rats gavaged with Alberta crude oil, at doses of 1.2 – 5 ml/kg, did not significantly affects body weight gain, alanine aminotransferase, blood urea nitrogen and creatinine. It is plausible to note that the tissue damage was not sufficient enough to allow the leakage of these marker molecules from the tissues into the blood system [15].

As observed in the result, AST, ALT and ALP were all significantly increased in the treatment rats (Figures 1 to 3). They were all found and appeared in high concentration in the liver, kidney and heart, so it could serve as an index of hepatorenal damage 15, 16]. This finding is consistent with previous study on organ toxicity in rats treated with landfill leachate [17].

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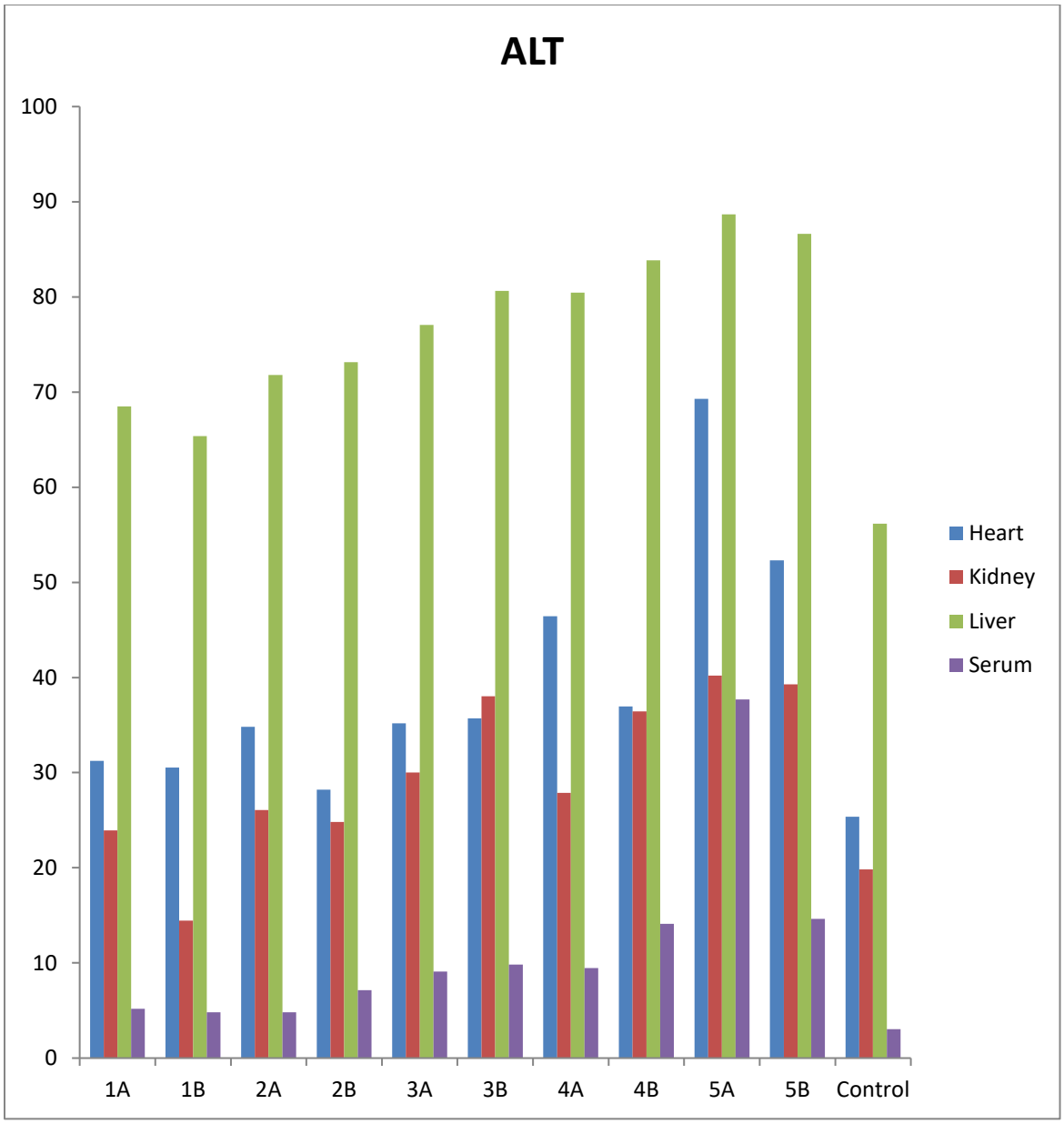

Fig 1: Biomarker of hepatic dysfunction ALT (alanine aminotransferase) in experimental rats

1A and B 200 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

2A and B 400 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

3A and B 600 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

4A and B 800 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

5A and B 1000 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

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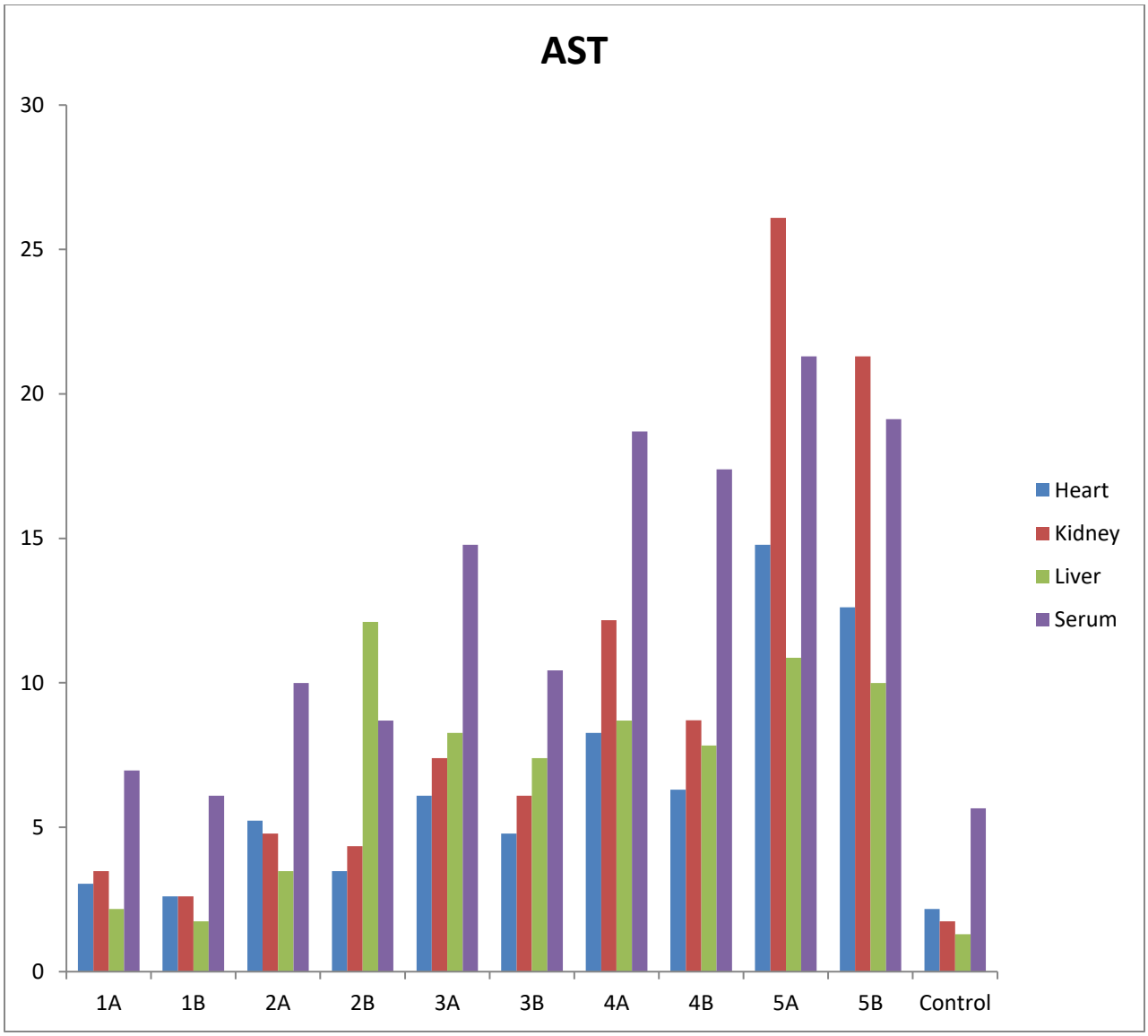

Fig 2: Biomarker of hepatic dysfunction AST (asparte aminotransferase) in experimental rats

1A and B 200 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

2A and B 400 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

3A and B 600 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

4A and B 800 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

5A and B 1000 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

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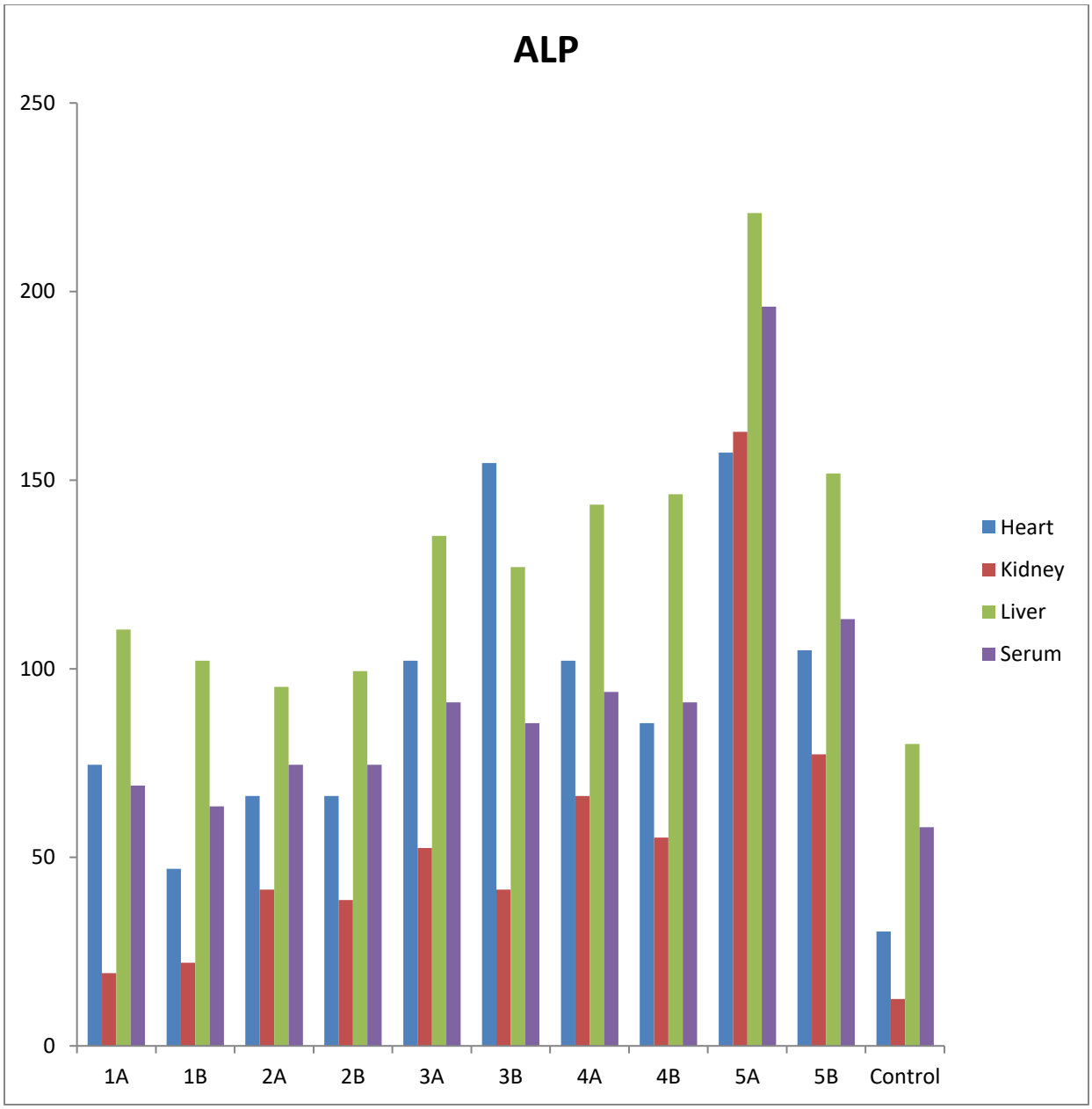

Fig 3: Biomarker of hepatic dysfunction ALP (alkaline phosphate) in experimental rats

1A and B 200 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

2A and B 400 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

3A and B 600 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

4A and B 800 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

5A and B 1000 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

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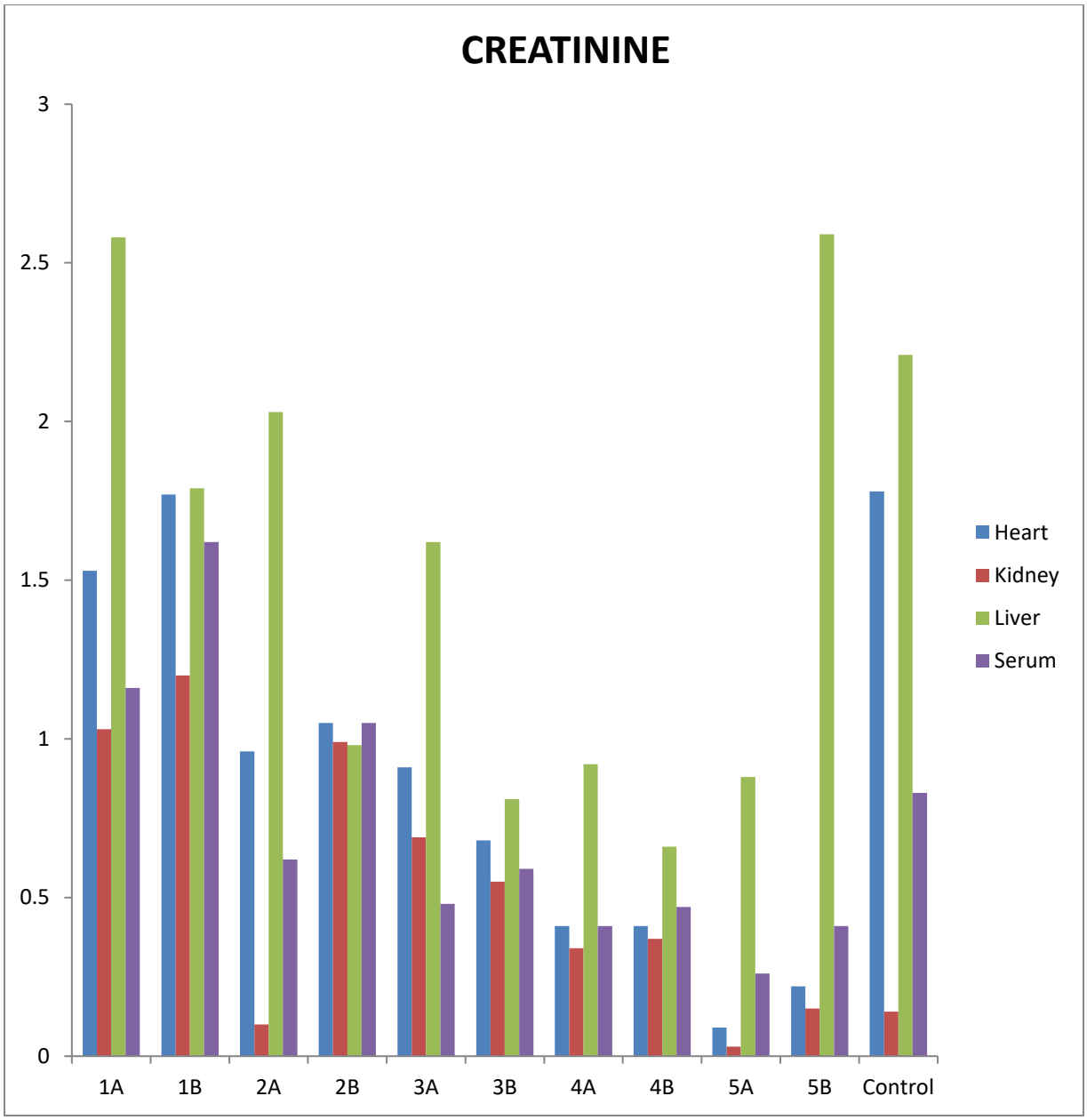

Fig 4: Biomarker of hepatic dysfunction Creatinine in experimental rats

1A and B 200 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

2A and B 400 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

3A and B 600 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

4A and B 800 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

5A and B 1000 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

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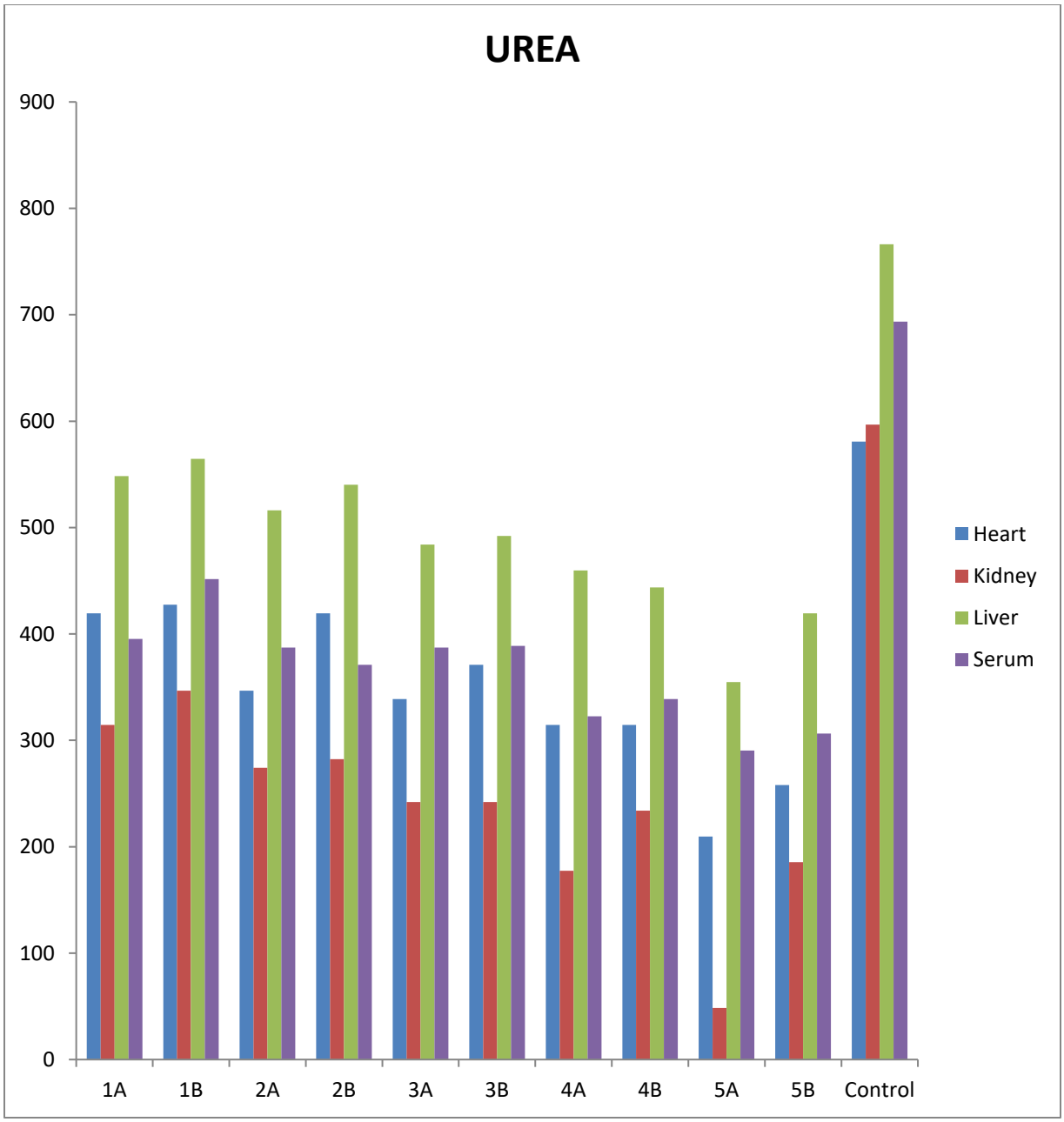

Fig 5: Biomarker of hepatic dysfunction Urea in experimental rats

1A and B 200 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

2A and B 400 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

3A and B 600 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

4A and B 800 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

5A and B 1000 mg kg^{-1}dose of aqeous FCF extract at 160 and 230 ^{o}C respectively

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Effects of extract on hematological parameter

Studies on hematological parameters can easily reveal abnormalities in body metabolic processes and the blood profile usually provides important information on the response of the body injury or lesion, deprivation and stress [18]. Red blood indices such as the mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) are the most useful indicators in the diagnosis of anemia in most animals [19]. The effects of FCF on hematological parameters of the test and control rats used in this study are as shown in Table 2.

It can be seen from Table 2 that, the aqueous extract of the FCF did not impact much on hematological parameters after 14 days administration compared to the control except in white blood count (WBC). A significant increase in the blood of test rats in groups 1A, 1B, 2A, 3A, 4A, 4B and 5B at the doses of 200, 400, 600, 800 and 1000 mg kg^{-1} was observed in comparison with the control group. In toxicity studies, increase in WBC may indicate the impact of extract in inducing the immune response of the treated animals [20]. On the other hand, significant decrease in the WBC of the blood indicates a decline in the production of leukocytes called leukopenia, meaning that the body is less able to fight off infections. However, in this study the estimated total WBC significantly changed in response to the administered aqueous FCF extract compared to the control. This result may indicate that aqueous FCF extract possesses chemicals capable of inducing leukocytosis which is abnormal high number of WBC in the blood circulation [19]. Also, there was a significant increase in the MCV of all the groups except 3B group which had a similar value as the control group. These observations demonstrated that the FCF extract in this study did not cause any significant toxic effect on the level of calculated red blood cell, even though the extract has the potential of causing problem if dosages continue for a longer period. This finding is in agreement with other findings in which the values of the various RBC parameters of extract treated rabbits were found to be comparable with those of the control group [21].

Table 2: Results of hematological study on both the treatment rats and the control

Sample	PVC %	Hb g/L	WBC 10^9 g/L	RBC 10^{12} g/L	MCV fL	MCH pg	MCHC g/dl

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1A	35	13.20	9.0	3.74	93.50	35.30	37.70
1B	34	11.20	7.9	3.75	90.60	29.90	32.90
2A	34	11.30	8.5	3.70	91.89	30.54	33.23
2B	31	10.30	4.4	3.40	91.17	30.29	33.22
3A	32	10.86	5.8	3.07	104.2	35.20	33.80
3B	32	10.80	4.2	3.92	81.63	27.70	33.93
4A	36	12.00	7.4	4.00	90.00	30.00	33.33
4B	34	11.42	6.2	3.84	88.54	29.73	33.58
5A	31	10.40	5.1	2.95	105.1	35.30	33.60
5B	33	11.20	10.3	3.71	88.90	30.20	33.90
Control	32	10.86	4.2	3.92	81.63	27.70	33.93

PVC = Pack volume cell Hb = Hemoglobine WBC = White blood cell

RBC = Red blood cell MCV = Mean corpuscular volume

MCH = Mean corpuscular hemoglobin MCHC = Mean corpuscular hemoglobine concentration

Histopathological examination

The results of the examination of tissue sections for histological changes supplemented and strengthened the evidence from biochemical and hematological parameters. Examination of kidney sections of the treated rats with repeated dosing of FCF extract indicated no structural difference compared to the control group. The architecture of sections of kidney test rats was similar to that of the control in which the renal corpuscles maintained their normal size of urinary space and normal tubular structures with no necrosis observed except in Plate 1b where there was the potential for glomerular tuft destruction and tubular necrosis in 3A, 4B and 5B.

Kidney is a sensitive organ, whose its functionality depends on so many factors. Kidney function test is a collective term for different types of tests and procedures that can be performed to determine how well the kidneys are functioning [22]. Accordingly, renal function can be assessed by measuring the levels of plasma createne, urea and uric acid concentration [22]. Assessment of possible damage due to the FCF extract in this present study was made by assaying plasma urea and createne levels [23]. Results showed that in all the treated rats there was no significant alteration in the blood urea and createne levels due to FCF treatment, although, there was a decrease in urea level and an increase in createne level from the control, but all the values were within the normal range [24].

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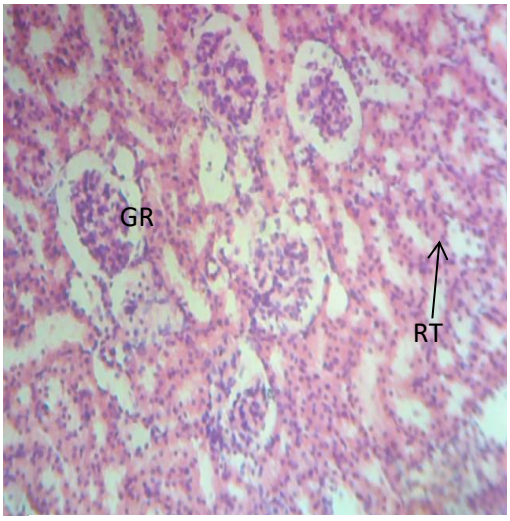
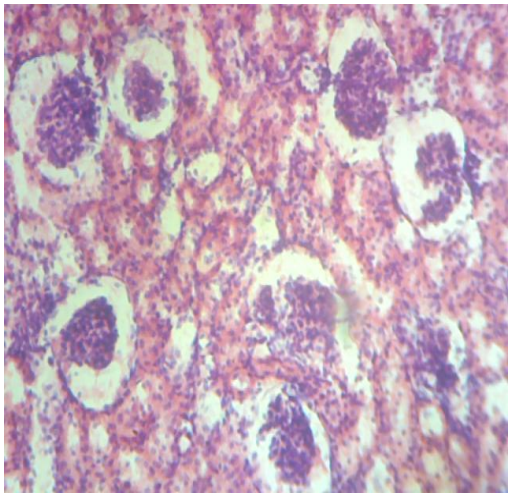
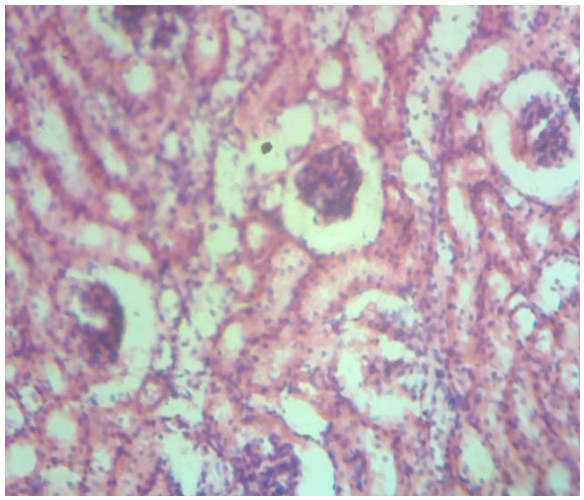


Control





1 B

1 A

Plate 1a: Histopathology defect on kidneys of rats exposed to FCF extract 1A, B 200 mg kg-1 and the control

Control: Normal kidney with almost near normal kidney histology

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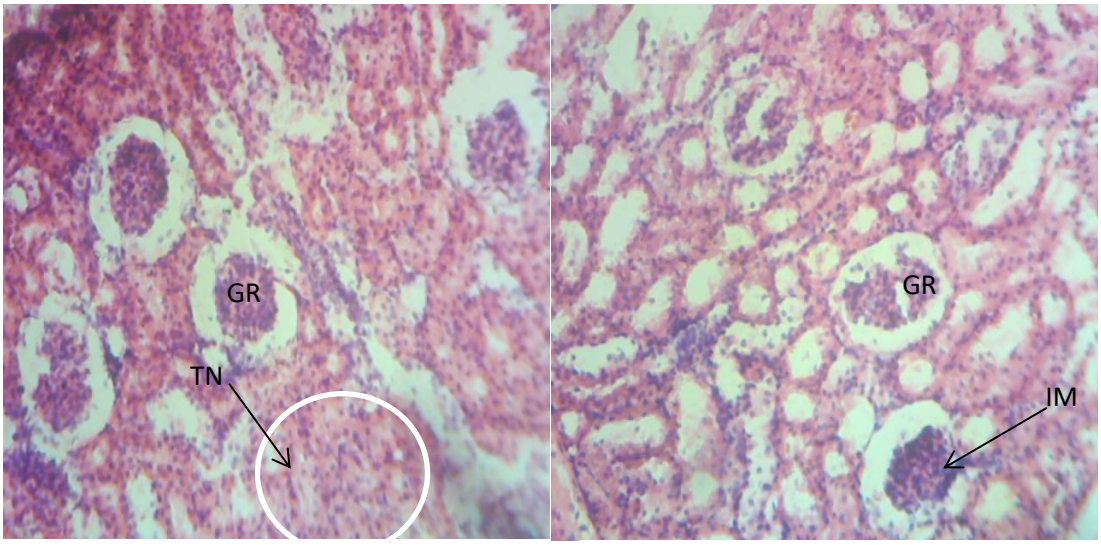
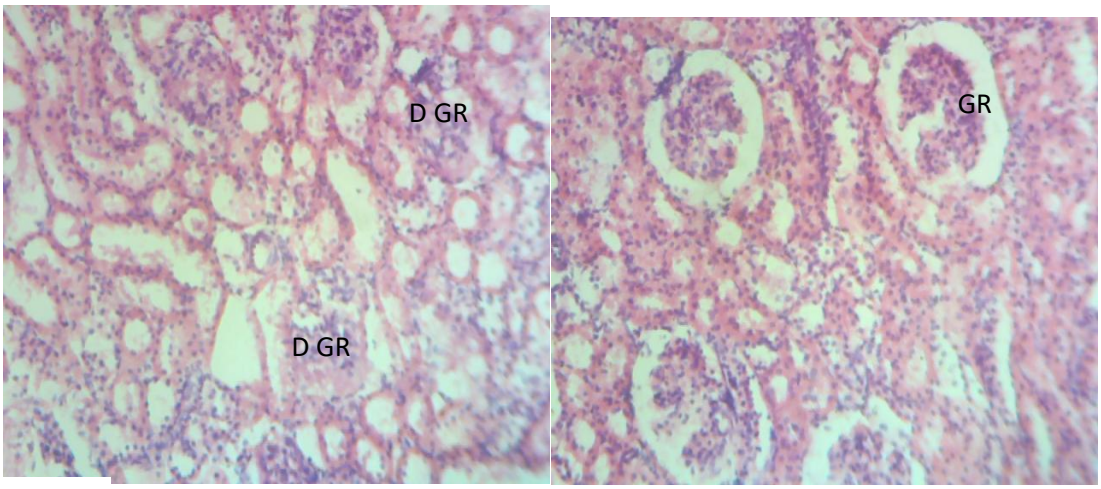
1 A: kidney with intact glomeruli room (GR) and possible deposition of immunological materials (IM) in the glomeruli basement

1 B: Kidney with intact glomeruli room (GR) and possible deposition of immunological materials (IM) in the glomeruli basement



2 A 2 B



3 A 3 B

Plate 1b: Histopathology defect on kidney of rats exposed to FCF extract 2 A, B 400 and 3 A, B 600 mg kg-1

2 A: Kidney with congregative acute tubular necrosis (TN) of the proximal convoluted tubule but with intact glomeruli room

2 B**:** Kidney with almost near architecture but with a possible deposition of immulogical materials (IM) in the glomeruli basement

3 A: kidney with destruction of the glomeruli turf and disappearance of the glomeruli room (GR)

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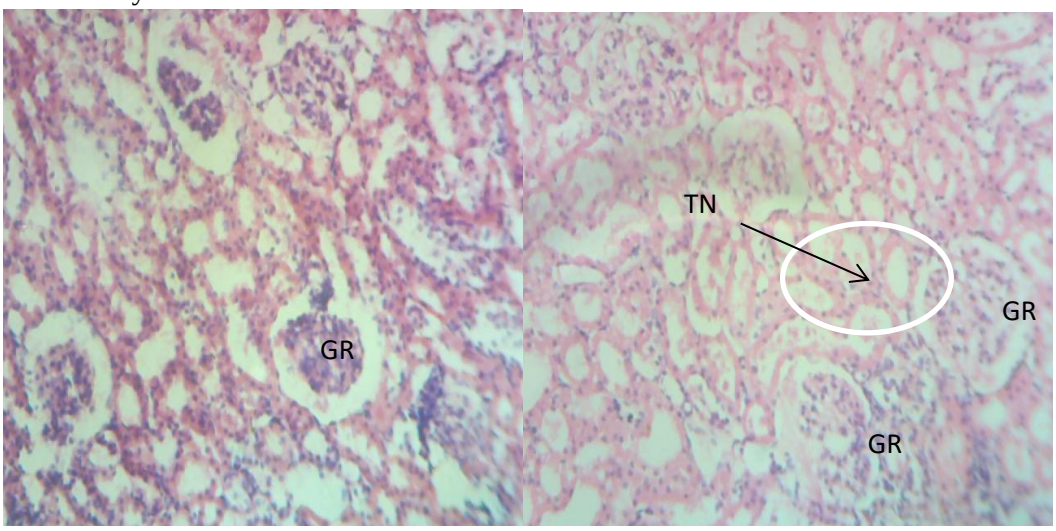
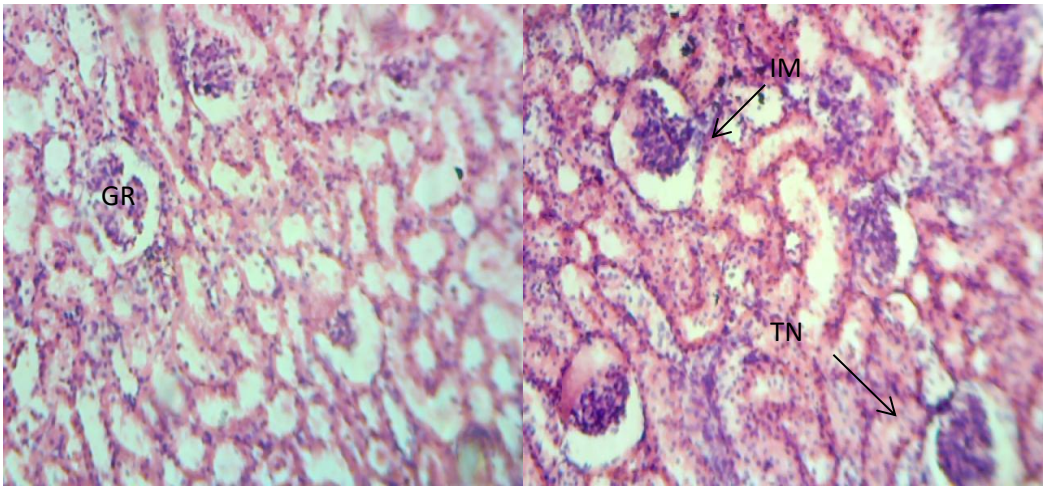
3 B: Kidney with almost near normal architecture



4 A 4 B



5 A 5 B

Plate 1c: Histopathology defect on kidneys of rats exposed to FCF extract 4 A, B 800 and 5 A, B 1000 mg kg-1

4 A: kidney with almost near normal architecture

4 B: Kidney with destruction of the glomerular tuft and congregative acute tubular necrosis

5 A: kidney with almost near normal architecture

5 B: Kidney with Tubular necrosis, and possible deposition of immulogical materials (IM) in the glomeruli basement

In this present study, the photomicrographs of liver sections of rat's Plate 2a to 2c showed the normal architecture of structural units of the liver, the hepatic lobules, formed by cords of hepatocytes separated by hepatic sinusoids. There were no significant histopathological

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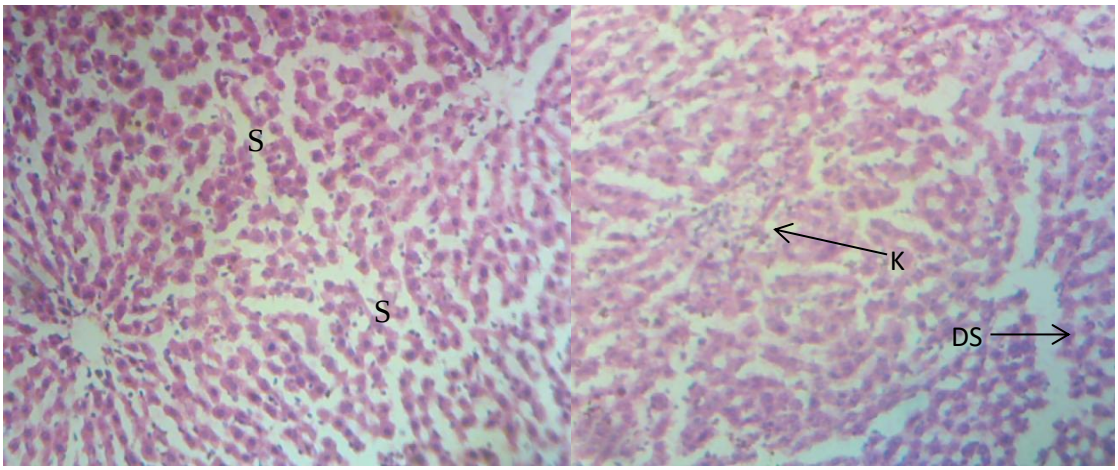
presentations observed in the groups treated and control. The liver appeared normal with preserved hepatic architecture, hepatocytes arranged as radial plates and having eosinophilic cytoplasm and basophilic central nuclei. No cytoplasmic inclusions were seen and no portal inflammation. Mild blood congestion was observed in the liver at doses of 600 and 800 mg kg-1 within the hepatic portal and central veins 3A, 3B, 4A and B.

Test for liver include several serum chemistries that reflect liver functions such as alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphate (ALP), Gammaglutamyl transpeptidase and albumin. The major intracellular enzymes of the liver are ALT and AST. However damages to liver allow the escape of these enzymes into the bloodstream and increase their levels [25]. Unlike AST, ALT is fairly specific because it is found largely in the liver and it is commonly used as biomarker for liver problems [26]. ALT is purely cytosolic and more for hepatocytes. Increment in serum levels for both AST and ALT can occur with states of altered hepatocellular membrane permeability. Because ALT is located only in the cytosol, serum levels tend to be relatively higher than AST. Since majority of AST is found within mitochondrial of cells, it is released slowly in comparison to ALT [27]. Therefore, serum transaminases, especially ALT, are the most important markers of hepatic injury [26].



1 A

1 B

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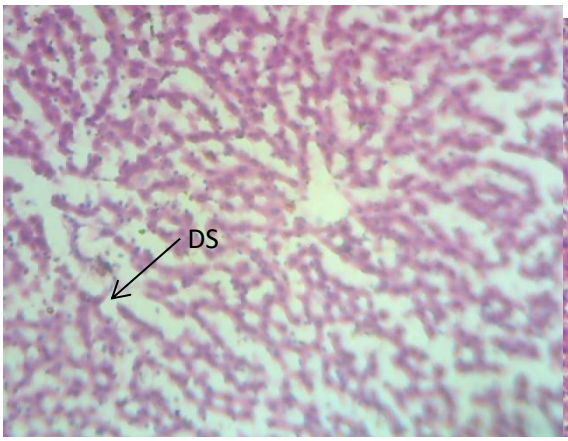
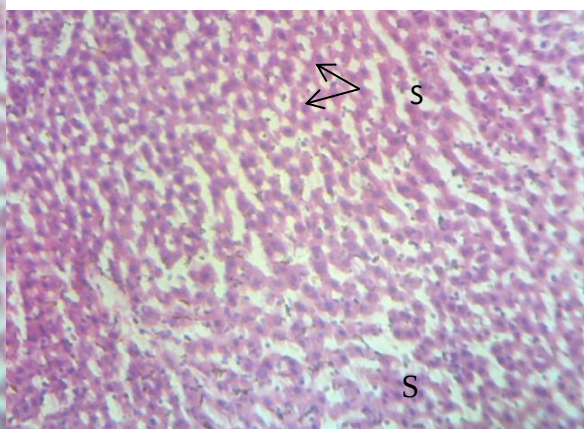




2 A 2 B

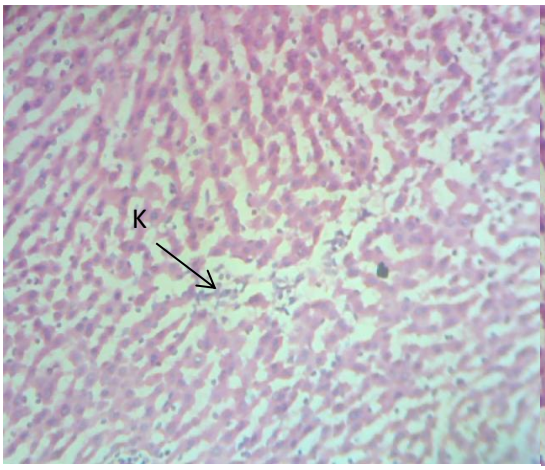
I ra A, B 400 mg kg-1

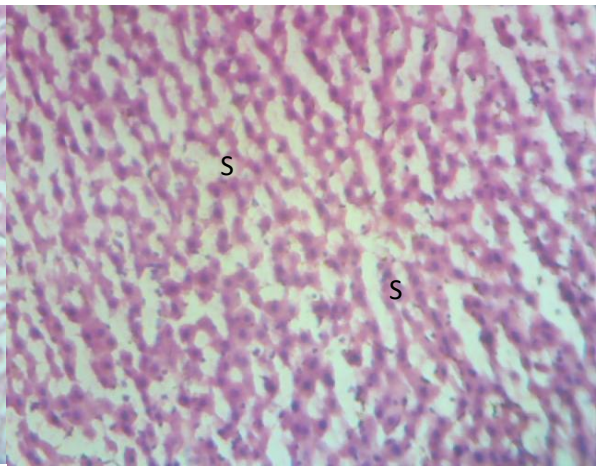
1 A: Normal liver structure with the hepatic cells in order and the hepatic sinusoids(S) separating the hepatic cord in place lined by Kupffer cell

1 **B:** Liver with karyolysis (K), dilation of sinusoid (DS) and having some cells with less prominent nucleus

2 **A:** Liver with dilation of sinusoid (DS

2 B: Liver having sinusoid (S) to have been greatly diffused.





3 A 3 B

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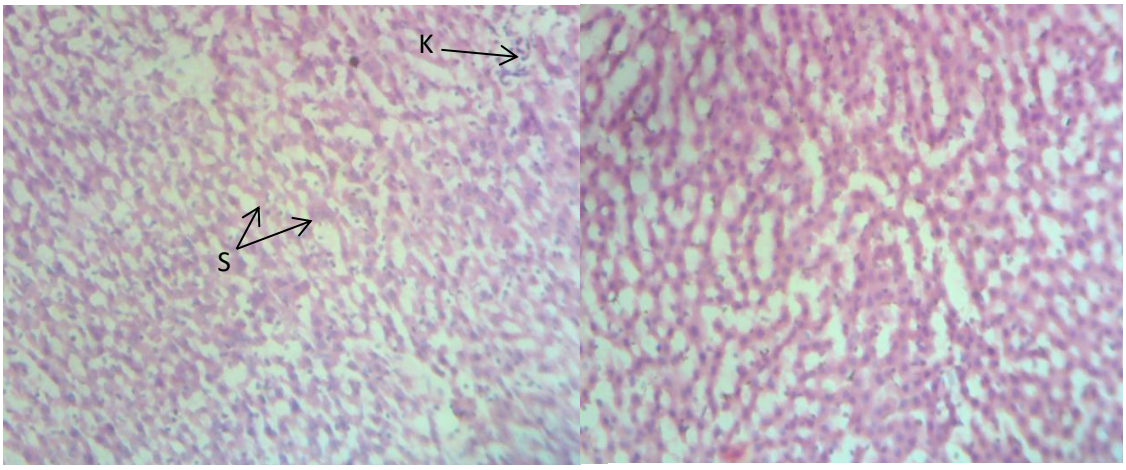


4 A 4 B

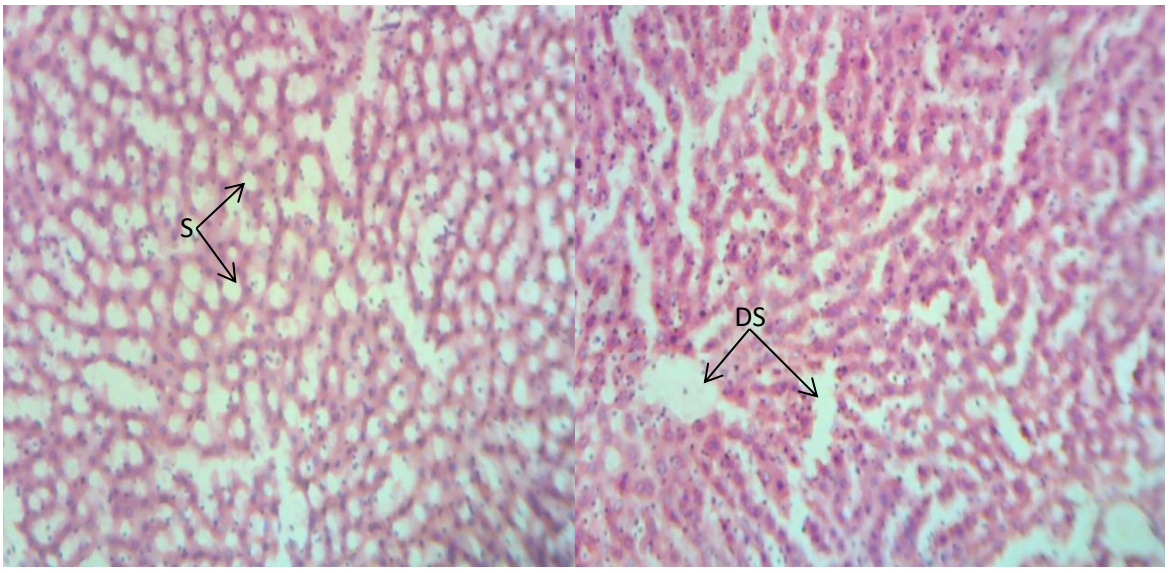
Plate 2b: Histopathology defect on Liver of rats exposed to FCF extract 3 A, B 600 and 4 A, B 800 mg kg-1

3 A: liver with karyolysis (K)

3 B**:** liver with almost near normal architecture

4 A**:** Liver with karyolysis (K) and sinusoid (S) to have been greatly diffused

4 B: liver with almost near normal architecture but with part of the sinusoid (S) to be diffused



5 A 5 B

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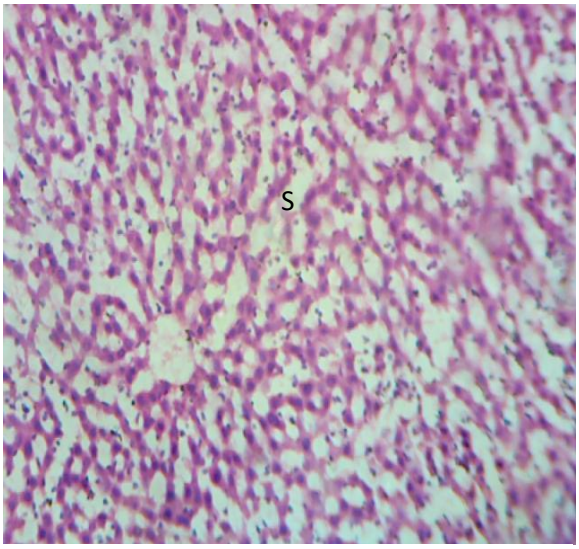

Control

Plate 2c: Histopathology defect on Liver of rats exposed to FCF extract 5 A, B 1000 mg kg-1 and the control

5 A: Liver section with diffused sinusoid (S)

5 B: Liver section with dilated sinusoid (DS)

Control:liver with almost near normal architecture

Karyolysis is the complete dissolution of the chromatin of a dying cell due to enzymatic degeneration. Hence, the whole cell will eventually stain uniformly with eosin. Cells with less prominent nucleus have their boundaries to be less distinct. Sinusoid are small vessels in the liver that serve as a location for the oxygen rich blood from the hepatic artery and the nutrient rich blood from the portal vein. Kupffer cells are macrophages that reside inside the sinusoids and can take up and destroy foreign material such as bacteria.

The histological assessment of the heart of rats in the control and treated groups is as shown in plate 3 - 3j. The profile alteration seen in the heart section of the treated rats as compared to the control includes defected tissue with severe enlargement / depletion of connective tissue and a tissue having enlarged muscle fibre. Even though, the alteration is not significant enough to cause damages to the heart tissues, it is a strong indication that consumption of fried cat fish should be done with caution, as higher dose may trigger on an unexpected reaction. Rate of reactions are known to be dependent of the concentration of the reactant(s).

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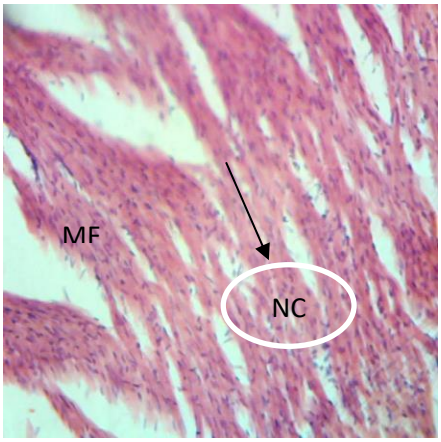
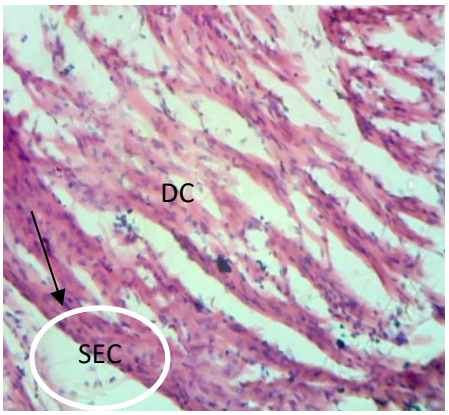


Plate 3: Control

(Hematocylin and eosin stain, Mag. X400)

Normal heart architecture with a regular cardiomyocyte arrangement and normal connective tissue (NCT) and muscle fibre (MF)



1 A

Plate 3a: Histopathology defect on heart of rats exposed to FCF extract 1A 200 mg kg^{-1}

(Hematocylin and eosin stain, Mag. X400)

Defected heart tissue with severe enlargement of connective tissue (SEC) or vaculations and depleting cardiomyocytes.

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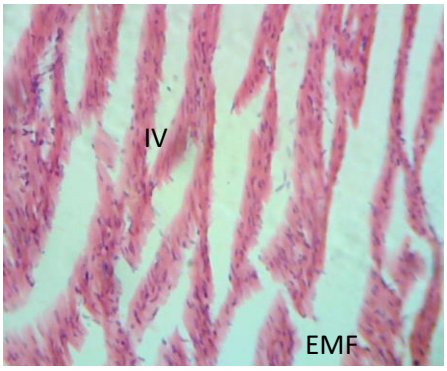
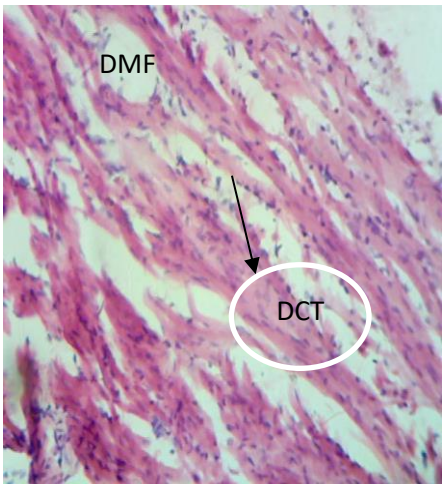


1 B

Plate 3b: Histopathology defect on heart of rats exposed to FCF extract 1B 200 mg kg^{-1}

(Hematocylin and eosin stain, Mag. X400)

Defected heart tissue having irregular vaculations and enlargement of muscle fibre.



2 A

Plate 3c: Histopathology defect on heart of rats exposed to FCF extract 2A 400 mg kg^{-1}

(Hematocylin and eosin stain, Mag. X400)

Defected heart tissue having depleting connective tissue (DCT) and diffuse muscle fibre (DMF).

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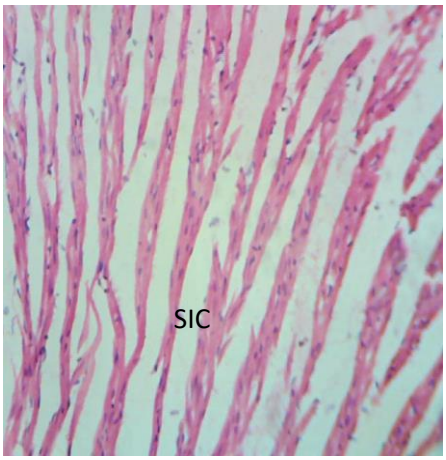
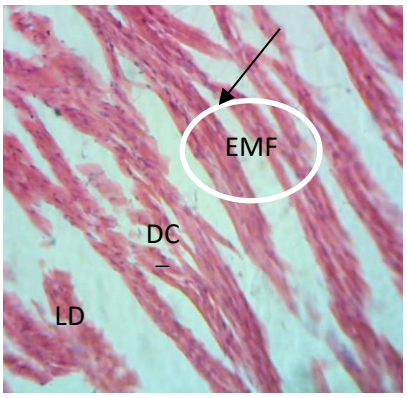


2 B

Plate 3d: Histopathology defect on heart of rats exposed to FCF extract 2B 400 mg kg^{-1}

(Hematocylin and eosin stain, Mag. X400)

Defected heart tissue having severe irregular connective tissue drainage (SIC) or vaculations.



3 A

Plate 3e: Histopathology defect on heart of rats exposed to FCF extract 3A 600 mg kg^{-1}

(Hematocylin and eosin stain, Mag. X400)

Defected heart tissue having enlarged muscle fibre (EMF) and depleting connective tissue (DCT) with lipid deposits (LD).

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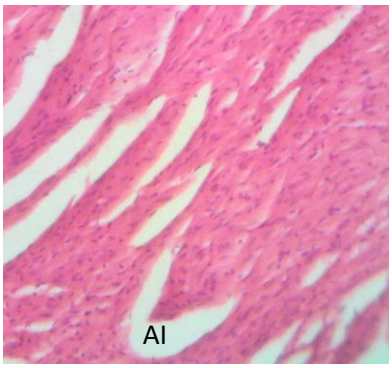
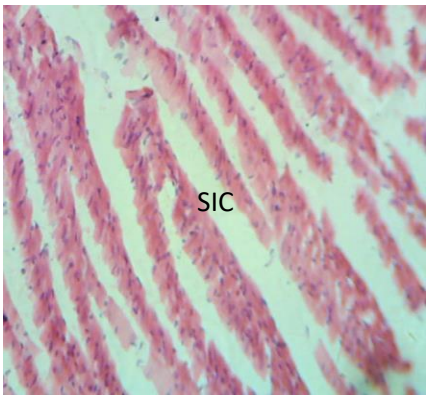


3 B

Plate 3f: Histopathology defect on heart of rats exposed to FCF extract 3B 600 mg kg^{-1}

(Hematocylin and eosin stain, Mag. X400)

Defected heart tissue having slight acute interstitial drainage of cardiomyocytes (AID)



4 A

Plate 3g: Histopathology defect on heart of rats exposed to FCF extract 4A 800 mg kg^{-1}

(Hematocylin and eosin stain, Mag. X400)

Defected heart tissue having severe irregular connective tissue drainage (SIC) or vaculations.

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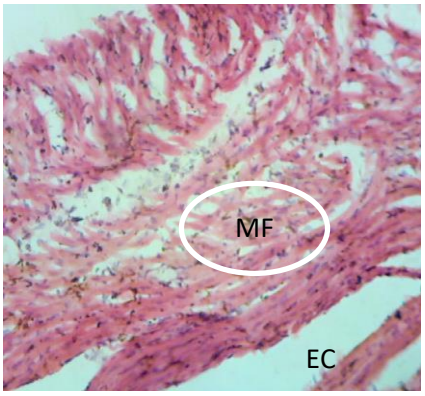
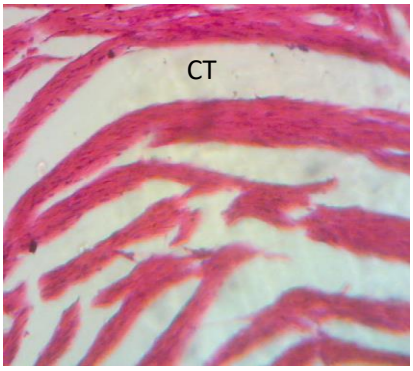


4 B

Plate 3h: Histopathology defect on heart of rats exposed to FCF extract 4B 800 mg kg^{-1}

(Hematocylin and eosin stain, Mag. X400)

Defected heart tissue having normal muscle fibre (MF) and enlarge connective tissue (ECT).



5 A

Plate 3i: Histopathology defect on heart of rats exposed to FCF extract 5A 1000 mg kg^{-1}

(Hematocylin and eosin stain, Mag. X400)

Defected heart tissue having severe depletion of interstitial layer/connective tissue (CT)

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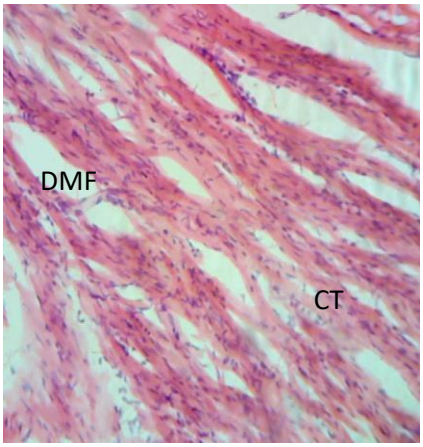

5 B

Plate 3j: Histopathology defect on heart of rats exposed to FCF extract 5B 1000 mg kg^{-1}

(Hematocylin and eosin stain, Mag. X400)

Defected heart tissue having normal connective tissue with diffuse muscle fibre.

CONCLUSION

Acute toxicity test to determine the effects of aqueous extract of fried cat fish was carried out The acute toxicity test carried out showed that the oral dosage administered from 200 mg kg^{-1} to 1000 mg kg^{-1} were not toxic to the Wistar rats when administered orally. The biochemical studies showed that serum, ALT, ALP, AST as well as urea and creatinine concentration were affected across the treatment groups of animals which were indications for potential hepatorenal damage. The WBC and MCV showed significant changes in response to the administered dosage, while the liver, kidney and the heart showed no significant histopathological presentations observed in the groups treated and control. The study revealed that there can be potential toxicity if there is repeated indulgence to fried cat fish at the temperatures between 160 – 230 ^{o}C.

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