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Research Article

Histopathological Effects of Three Types of Potash on the Hearts of Wistar Rats

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Abstract

Potash (kanwa) is widely consumed in many African countries as a food additive despite concerns regarding its possible toxic effects on body organs. This study investigated the histopathological effects of three types of potash on the hearts of Wistar rats. The study was carried out in Ekpoma, Edo State, Nigeria using sixty (60) adult albino Wistar rats randomly assigned into control and experimental groups. The experimental groups received graded doses of potash (0.4g/ml, 0.6g/ml, and 0.8g/ml orally) for four weeks, while the control group received normal feed and water only. At the end of the experiment, the animals were sacrificed under chloroform anaesthesia and heart tissues were harvested for histological examination using Haematoxylin and Eosin staining techniques. The findings revealed varying degrees of histopathological alterations in the treated groups compared with the control group which showed normal cardiac architecture. The observed lesions included cellular necrosis, haemorrhage, oedema, vacuolations, vascular carbon deposits, and cyto-architectural distortion of the cardiac tissues. The severity of tissue damage increased with higher concentrations of potash administration, with the highest dose producing the most pronounced pathological changes. These findings suggest that prolonged consumption of potash may exert toxic and degenerative effects on cardiac tissues. The study concludes that excessive intake of potash could be harmful to the heart and may predispose individuals to cardiovascular abnormalities. Therefore, moderation in potash consumption is strongly recommended, while further studies are needed to evaluate its long-term functional effects on vital organs.

1. Introduction

Potash is any of the various mined and manufactured salts that contains potassium (potassium sulfates, hydroxides, chlorates, nitrates and carbonates) in water-soluble form [1]. The name is derived from "pot ash", which refers to plant ashes soaked in water in a pot, the primary means of manufacturing the product before the industrial era. The word "potassium" is derived from potash [2]. Potash refers to one of the salts that been mined; which is made up of potassium that can easily dissolve in water, of which the name pot ash was obtained from the plant burn to ashes which is then dissolved in water housed by a pot, and this serves as the major production method on before the use of technology [2]. Potash ore deposits are also found below the earth's surface as potash salts in evaporite sequences in pre-quaternary sedimentary basins formed in arid climates with restricted seawater [3]. They are obtained by shaft or dissolution mining (Khan, 2014). They typically exist as white or a mixture of grey and reddish brown depending on the presence of other trace elements like Iron and Manganese [4].

Kanwa (potash), a sesquicarbonate or hydrated carbonate of sodium is a product of the salt industry in many parts of northern Africa [5]. In Nigeria, potash is traditionally called kaun, kanwa and akanwu by the Yoruba, Hausa and the Igbos respectively while the Igalas and the Egbira refer to them as okanwa and ikoro respectively. Huge deposits are found in northern Nigeria like Kano and Maiduguri and other border countries like Niger and Chad [4, 6]. Though its actual name is trona or natron salt because it contains high amount of sodium compared to potassium. It occurs as a mixture of different substances with sodium constituting about 30% and other minerals such as potassium, iron, zinc in varying proportions. There are different varieties and its composition varies with respect to their locations [7, 8].

Potash has several uses; it is used in cooking as a food tenderizer especially in pulses [8, 9], to curdle milk, in the tanning industry and in the preparation and enhancement of flavour of local beverages and snuff. It is widely used in the local production of fertilizers, soap, fire extinguishers and bleaching textiles with varying uses for culinary and medicinal purposes. They are traditionally used as a tenderizing agent in cooking beans, breadfruit (*ukwa*), black Mexican beans (*akidi*), cowpea beans (*fiofio*) and cowhide (*ponmo*). Also employed to retain and enhance the green colour of jute leaves (*ewedu*) and okra vegetables and to emulsify oil and water in the preparation of local delicacies like *nkwobi ugba* and *abacha*. In folk medicinal applications, they are used in the treatment of stomach ache, toothache, cough and constipation. They have also found use among nursing mothers to enhance lactation [10].

The heart is described as a hollow muscular organ responsible for pumping blood from the blood vessels to various parts of the body using repeated, rhythmic contractions [11]. All animals including vertebrates with a circulatory system possess a heart. The vertebrate heart is principally composed of cardiac muscle and connective tissue. Cardiac muscle is an involuntary striated muscle tissue specific to the heart and is responsible for the heart's ability to pump blood [11]. Cardiovascular diseases have been reported in several literatures which includes of stroke and hypertension attributed to the increased levels of plasma lipids [12, 13]. Heart diseases as a result of increased lipid are largely due to unhealthy nutritional lifestyle. These unhealthy lifestyles could result in the accumulation of lipids on the arterial walls to form plagues and blocks the vascular lumen and subsequently inhibits the flow of blood the brain, liver, heart and kidney and this results to coronary heart diseases, hypertension or stroke [14]. Heart diseases such atherosclerosis and myocardial infarction as has also been reported to be responsible for increased morbidity and mortality [15, 16]. Considering the role of the heart as the pumping organ of the body [11]; the heart might be at risk since all ingested substances must be circulated in the body via the blood. Similarly, cases of death have been attributed to the ingestion of toxic substances [17]. Nitrate poisoning of which can occur virtually in all animals has also been linked to potash consumption [18]. Also, several acute and chronic health hazards have been reported to be caused by potash when it is in nitrate form. It is a known fact that the consumption of potash is quite high and also limited literature exists as regards the effect of potash on various body organs. The increased use of potash has been reported to affect the heart [19]. Although potash and related compounds such as potassium bromate are used for certain food preparations however, studies by Bankole [20] and Kurokawa [21] have revealed that it is not suitable for our health thus curtailing the level of consumption is highly advisable. Therefore, the study determined the histopathological effects of potash on the hear of treated animals.

2. Methods

2.1. Area of Study

This study was carried out in Ekpoma. Ekpoma is the capital of Esan West Local Government Area in Edo State which falls within the rain forest/savannah transitional zone of south western Nigeria. The area lies between latitudes $6°43'$ and $6°45'$ North of the equator and longitudes $6°5'$ and $6°8'$ East of the Greenwich Median. Ekpoma has a land area of 923 square kilometers with a population of 170, 123 people as at the 2006 census [22]. The town has an official post office and it is the home of Ambrose Alli University. Majority of people in Ekpoma are students, Lecturers/Teachers, civil servants, farmers, traders, business men/women, doctors, lawyers and self-employed. Ekpoma is made up of many communities, including Eguare, Iruekpen, Emaudo, Ujoelen, Ihumudumu, Illeh, Uke, Uhiele, Ujemen, Ukpenu, Egoro, Emuhi, Igor and Idumebo. The inhabitants are mainly Christians, few Muslims and pagans scattered within the area. The samples were examined in the Research Diagnostic Laboratory, of the Department of Histopathology, Faculty of Medical Laboratory Science, College of Medicine, Ambrose Alli University, Ekpoma.

2.2. Research Design

A total of sixty (60) albino-Wister rats with twenty (20) rats used in each kanwa tested were used for this experiment which were randomly assigned three test groups of 5 rats in each group and 5 as control group. They were sacrificed after 4 weeks of kanwa administration. After the administration, the rats were put under light chloroform anaesthesia and the heart were harvested for histological processing. Heart sections were obtained from the adult albino wistar rat and fixed in 10% formal saline for 2 weeks. Samples were cut with thickness of 3mm in the cutting- up room. The selected tissues were placed in tissue baskets carefully labeled and processed histologically. All steps of this study were conducted according to the National Institute of Health (NIH) guidelines, as well as ethical guidelines for investigation of animals, which was approved by the Health Research Ethics Committee of Ambrose Alli University, Ekpoma, Edo State, Nigeria. The preliminary studies, animal acclimatization, ingredient procurement (kanwa preparation and production), actual animal experiment and evaluation of results and biochemical assessment lasted for a period of three months.

2.3. Ethical Approval

Approval for the study was obtained from the Health Research Ethics Committee of Ambrose Alli University, Ekpoma, Edo State, Nigeria and was carried out in strict accordance with the guideline for the care and use of animals for research committee which is in line with that set by WHO.

2.4. Experimental Animals/Housing Condition

Sixty (60) adult albino Wistar rats having a mean weight of 180-190 g were procured from the animal farm, College of Medical Sciences, Ambrose Alli University Ekpoma and transferred to the experimental Laboratory at the Histology laboratory of the Faculty of Medical Laboratory Science, College of Medical Sciences, Ambrose Alli University Ekpoma, Edo State, where they were allowed one (1) week of acclimatization. The animals were maintained under conditions of controlled temperature and 12-hour light and dark cycle; they were given access to food and water. They were kept in wire mesh cages with tripod that separates the animal from its faeces to prevent contamination. During this period of acclimatization, the rats were fed with growers' mash obtained from Edo Feeds and Flour Mill Limited, Ewu, Edo State, Nigeria and water under strict hygienic conditions. The animals were maintained and utilized in accordance with the standard guide for the care and use of Laboratory animals.

2.5. Animal Grouping

The experimental animals were separated into four groups (A – D) for each kanwa. Each group contained five rats each (n = 5) using 4 big cages to house them. Group A served as the control, while groups B - D served as the test groups. Group B – D received graded doses of Kanwa prepared accordingly and weighed to determine the quantity to be administered. Group A received only the normal feed (grower's mash) and water/saline with no administration of Kanwa.

2.6. Fundamental Animal Housing Condition

Sixty (60) male albino Wistar rats of comparable sizes and weights ranging from 180-190g were procured from the Animal Farm, College of Medical Sciences, Ambrose Alli University Ekpoma, Edo State and transferred to the Histology Laboratory where they were allowed two (2) weeks of acclimatization. They were kept in wire mesh cages with tripod that separates the animal from its faeces to prevent contamination. During this period of acclimatization, the rats were maintained in accordance with the standard guide for the care and use of Laboratory animals.

2.7. Substance Preparation

The crystals of the different Kanwa procured from general Ekpoma market was grounded into powdery form using mortar and pestle. The potash powder was mixed with the grower's mash used in feeding the Wistar rats except the animals serving as controls that were fed with only the grower's mash and water liberally. Wistar rats of (180-190g) in size were used throughout the experiments. Each sample was visually observed to establish its physical properties. The samples were then carefully ground with mortar and pestle and sieved through a 9µ-pore size sieve. Sample specimens were weighed on a Mettler AE 160 model weighing balance, mixed thoroughly and stored in a stoppered bottle under room temperature until used.

2.8. Substance Administration

The rats were weighed before the administration of Kanwa and before they were sacrificed. Graded doses of Kanwa (3g, 6g and 9g) mixed with 50g of grower's mash respectively was administered. The feeds were always there for the animals, but usually changed daily for a fresh one after cleaning the container and the animal cages. The administration of potash was primarily done by the following;

Group A (Control): 5 Wistar rats (control) were fed with grower's mash 50grams/190grams body weight. Feeding was performed daily for four weeks

Group B (Test): 5 Wistar rats were fed with 50 grams of grower's mash and given 0.4grams/ml of grounded Kanwa/190grams body weight. The feeding was performed daily for four (4) weeks.

Group C (Test): 5 Wistar rats were fed with 50 grams of grower's mash and given 0.6grams/ml of grounded Kanwa/190grams body weight. The feeding was performed daily for four (4) weeks.

Group D (Test): 5 Wistar rats (test) were fed with 50 grams of grower's mash and given 0,8grams/ml of grounded potash/190grams body weight. The feeding was performed daily for four (4) weeks.

2.9. Sample Collection for Histological Study

At the end of the third week, all the animals were euthanized after been anaesthetized using chloroform as sedative. Tissue samples were thereafter collected from the kidneys. These tissue samples were preserved in 10% neutral buffered formalin and were later processed using standard histopathology techniques; 70% alcohol (2hrs), 80% alcohol (2hrs), 90% alcohol (2hrs), 90% alcohol (2hrs), 95% alcohol, (2hrs), Absolute (2hrs), Xylene 1 (2hrs), Xylene II (2hrs), Molten paraffin wax 1 (2hrs) and Molten paraffin Wax II (2hrs), as used in AAU, Ekpoma. After the last timing, the tissues were removed from their plastic cassettes and placed at the centre of the metallic tissue mould and then filled with molten paraffin wax. They were also left to solidify after which they were now placed in the refrigerator at 5°C for 15 minutes. After the blocks were cool in the refrigerator for the time stated above (15 minutes), the blocks were then removed from the metallic case using a knife and after which the paraffin wax at the side of the blocks were removed.

The blocks were then trimmed and cut serially at 3µm on a rotary microtome. The sections were floated in water bath at 55°C and picked up by the use of a clean frosted end slides. The frosted end slides were now placed on the hot plate for 40 minutes for adequate attachment of the sections on the slides after which the sections were de-waxed, hydrated, air dried and stored in a slide box ready for staining process.

2.10. Staining Procedure (Haematoxylin and Eosin staining technique)

Sections for general tissue structure were stained by Haematoxylin and Eosin technique as follows: the sections were dewaxed in 3 changes of xylene (5 minutes). The sections were hydrated through descending grades of alcohol (absolute, 95%, 80% and 70%). The sections were stained in Harris haematoxylin (5 minutes), the sections were rinsed in running tap-water to remove excess stain. The sections were differentiated in 1% acid alcohol (3 seconds) and were blued in running tap water (10 minutes). The sections were counterstained with 1% eosin (1 minute) and finally rinsed in water, dehydrated in ascending grades of alcohol (70%, 80, 95% and absolute). The sections were cleared in xylene, air-dried and mounted with dibuthylphthalate propylene xylene (DPX). The slides were examined under a light microscope and photomicrographs were taken (The staining procedures are as used in AAU, Ekpoma, Edo State). All the necessary clinical quality control were ensured with the standard precautions in terms of proper timing, pH test of the solution's accurate measurement of reagents and prevention of particulate contaminants.

2.11. Photomicrography

The sections were examined under a light microscope and photomicrographs of each group was taken. The photomicrographs were used to interpret the results of all the groups.

3. Result

All the samples were crystalline in texture and the sample colours varied. One was light coloured while others were dark coloured brown.

Table 1: Physical Properties of the Analysed Samples of Kanwa

Kanwa samples	Appearance	Colour
Ja kanwa(A)	Crystal aggregates	Light pink
Balma (B)	Crystal aggregates	Light grey
Manda (C)	Crystalline	White

3.1. Results from Ja kanwa (Potash)

Histological Observations of the Heart (Ja kanwa)

Heart sections (H & E) × 400) showing (Group A) normal histological architecture i.e., the cellular integrity was well maintained.

Group B: The pre-acute phase of the treated group B showed varied degree of pathologic conditions which are vacuolations, cellular necrosis, haemorrhage as well as mild Oedema. Whereas, in the acute phase, the treated group B showed cellular necrosis, vacuolations, vascular carbon deposit, haemorrhage and Oedema.

Group C: Treated group C in the pre-acute phase showed severe vacuolations, cellular necrosis, Oedema and wavy cardiac myofibrils while the acute phase showed necrosis, vacuolations, Oedema, wavy cardiac myofibrils and Oedema with mild fibrolysis.

Group D: Treated group D, three out of six rats in this group died before the end of pre-acute phase. However, the histological result of the remaining three showed severe architectural distortion with numerous carbon deposit as well as cardiac infarction with deposit of carbon.

Table 2: Histopathological Observations of the Effect of Potash on the Heart of Wister Rats (Pre-Acute Phase)

Histopathological	Control Group A			Test group B			Test group C			Test group D		
Observations	1	2	3	1	2	3	1	2	3	1	2	3
Vacoulations	-	-	-	+	+	+	+++	+++	+++	-	-	-
Cellular necrosis	-	-	-	+	+	+	+	+	+	-	-	-
Haemorrhage	-	-	-	+	+	+	+	+	+	-	-	-
Mild Oedema	-	-	-	+	+	+	+	+	+	-	-	-
Wavy cardiac myofibrils	-	-	-	-	-	-	+	+	+	-	-	-
Cardiac infarction	-	-	-	-	-	-	-	-	-	+	+	+
Carbon deposit	-	-	-	-	-	-	-	-	-	+++	+++	+++

Table 3: Histopathological Observations of the Effect of Potash on the Heart of Wister Rats (Acute Phase)

Histopathological Observations	Control group A 4	5	6	Test group B 4	5	6	Test group C 4	5	6	Test group D 4	5	6
Vacoulations	-	-	-	+	+	+	+	+	+	-	-	-
Cellular necrosis	-	-	-	+	+	+	+	+	+	-	-	-
Haemorrhage	-	-	-	+	+	+	+	+	+	-	-	-
Mild Oedema	-	-	-	+	+	+	+	+	+	-	-	-
Ooedema with mild fibrolysis	-	-	-	-	-	-	+	+	+	-	-	-
Wavy cardiac myofibrils	-	-	-	-	-	-	+	+	+	-	-	-
Vascular carbon deposit	-	-	-	+	+	+	-	-	-	-	-	-

KEY; - = Negative; + = Mild/severe positive

Photomicrographs (Histopathological Observations) (Balma)

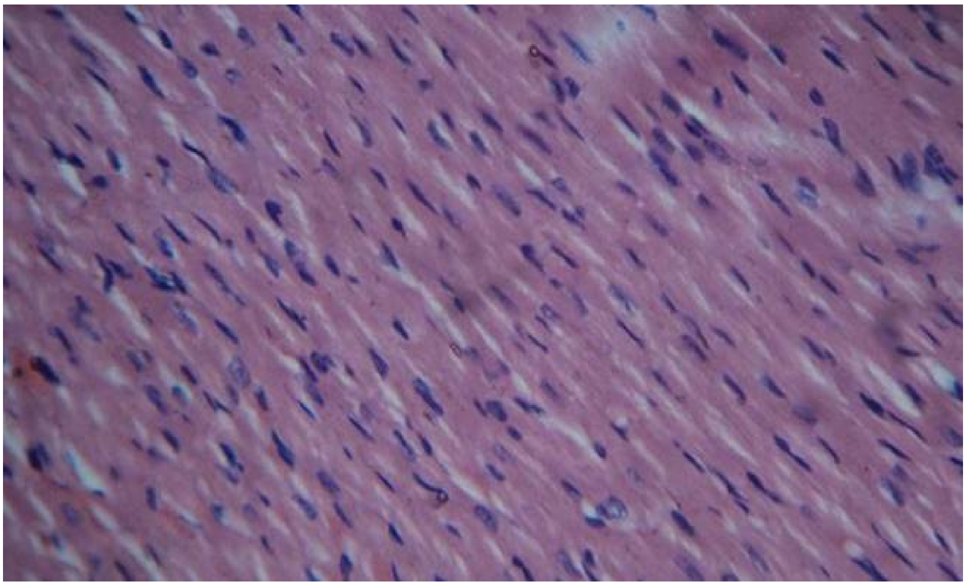

Figure 1: (H & E × 400) Control Group A heart section (A1) showing normal myocardial cells and intact architectural integrity.

Pre-Acute Phase

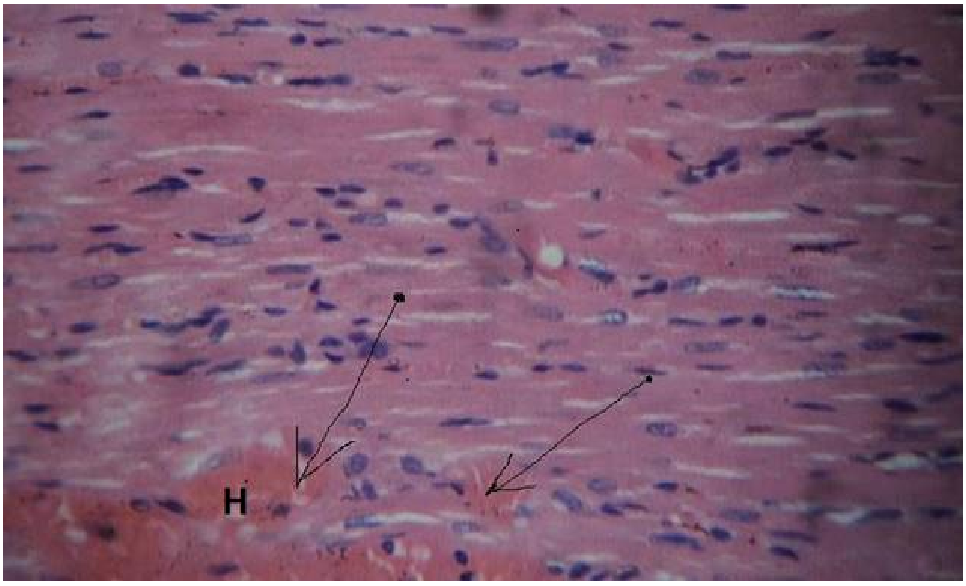

Figure 2: (H & E × 400) Test Group B (pre-acute phase): heart section showing haemorrhage (see arrow)

Figure 3: (H & E × 400) Test Group C (pre-acute phase): heart section showing wavy cardiac myofibrils (see arrow)

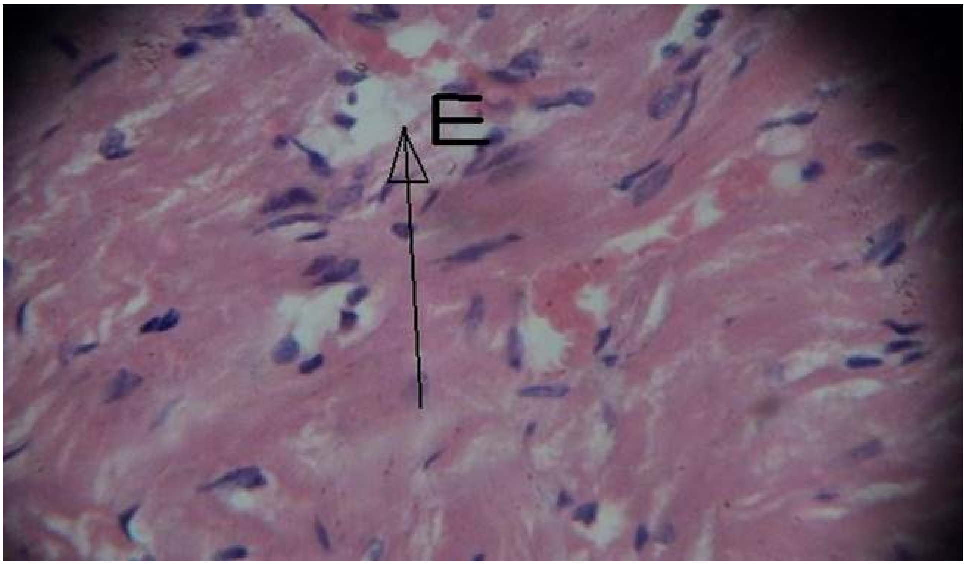

Figure 4: (H & E × 400) Test Group C (pre-acute phase): heart section showing oedema (see arrow)

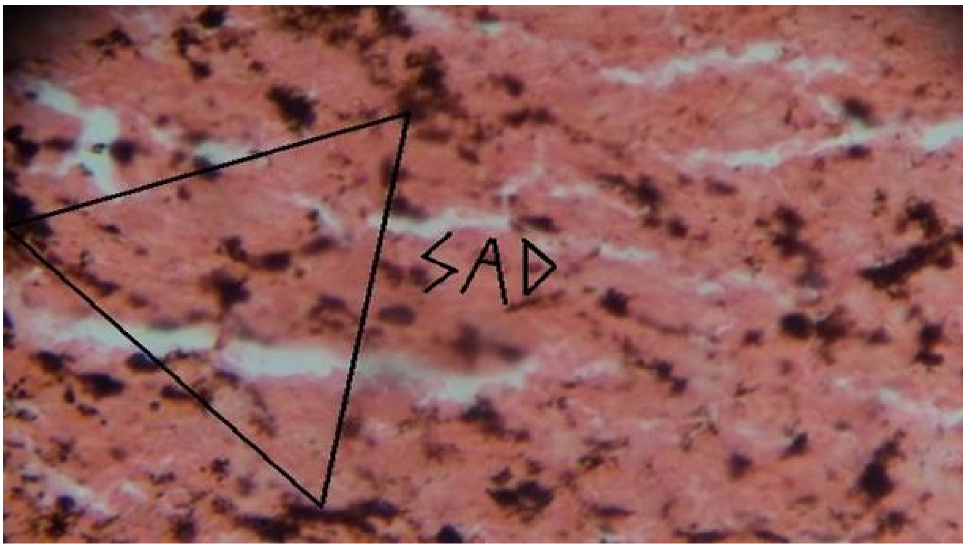

Figure 5: (H & E × 400) Test Group D: heart section showing severe architectural distortion with numerous carbon deposits

Acute Phase

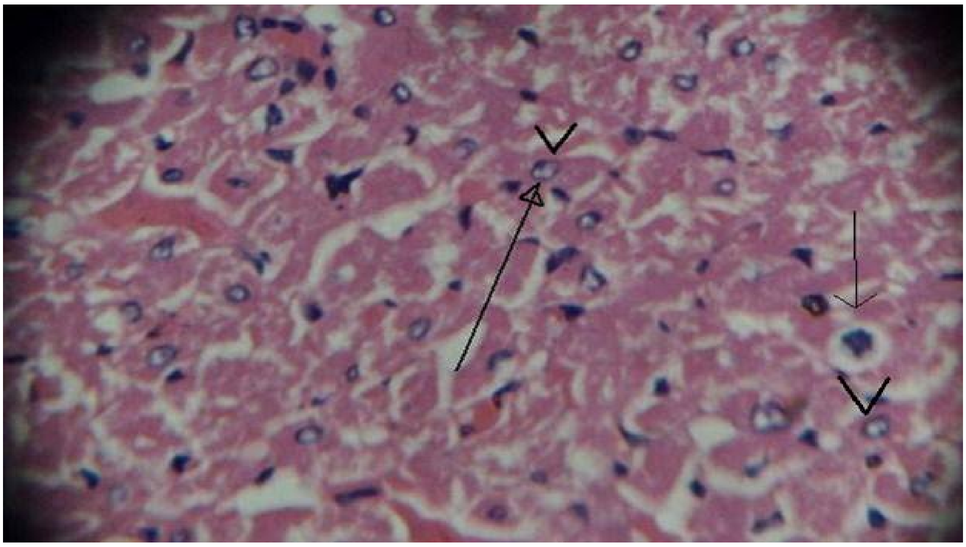

Figure 6: (H & E × 400) Test Group B (acute phase) heart section showing vacuolations (see arrows)

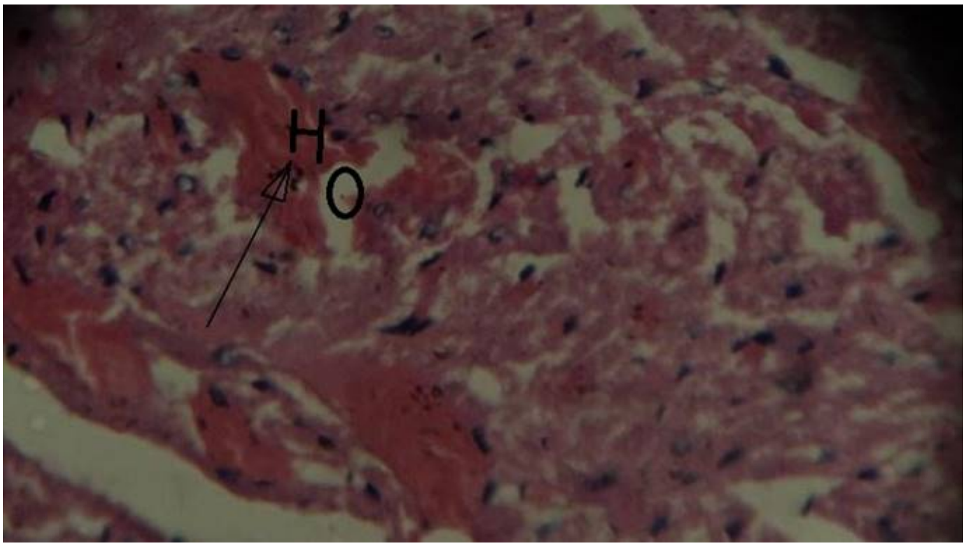

Figure 7: (H & E × 400) Test Group C (acute phase): heart section showing haemorrhage and oedema (see arrow)

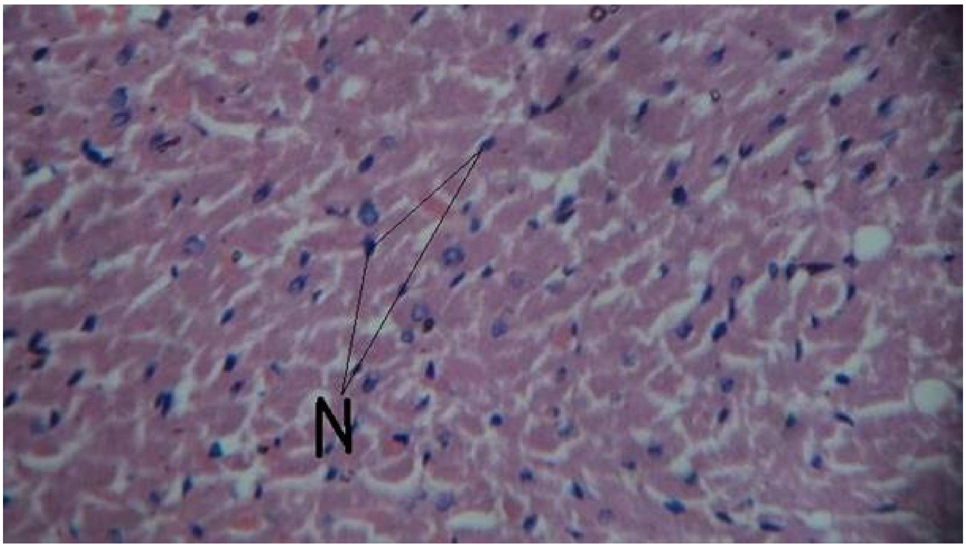

Figure 8: (H & E × 400) Test Group C (acute phase) heart section showing necrosis

3.2. Results from Balma (Potash)

Table 4: Physical Observation of Control and Experimental Rats During Treatment with Balma

Physical observation	Experimental groups			
	Group A	**Group B**	**Group C**	**Group D**
Skin Changes	-	-	-	-
Fur Changes	-	-	-	+
Tail Changes	-	-	-	+
Eyes	-	-	-	-
Behaviour Pattern	-	+	+	+
Tremors	-	-	-	-
Diarrhea	-	-	-	-
Death	-	-	-	1

KEY: - = Absent; + = Change in behaviour; + = The tail was sloughing off

Histological Observations of the Heart (Balma)

Heart sections (H & E) × 400) showing (Group A) normal histological architecture i.e., the cellular integrity was well maintained.

Group B: The pre-acute phase of the treated group B showed varied degree of pathologic conditions which are vacuolations, cellular necrosis, haemorrhage as well as mild Oedema. Whereas, in the acute phase, the treated group B showed cellular necrosis, vacuolations, vascular carbon deposit, haemorrhage and Oedema.

Group C: Treated group C in the pre-acute phase showed severe vacuolations, cellular necrosis, Oedema and wavy cardiac myofibrils while the acute phase showed necrosis, vacuolations, Oedema, wavy cardiac myofibrils and Oedema with mild fibrolysis.

Group D: Treated group D, three out of six rats in this group died before the end of pre-acute phase. However, the histological result of the remaining three showed severe architectural distortion with numerous carbon deposit as well as cardiac infarction with deposit of carbon.

Table 5: Histopathological Observations of the Effect of Potash on the Heart of Wister Rats (Pre-Acute Phase)

Histopathological Observations	Control group A			Test group B			Test group C			Test group D		
	1	**2**	**3**	**1**	**2**	**3**	**1**	**2**	**3**	**1**	**2**	**3**
Vacuolations	-	-	-	+	+	+	+++	+++	+++	-	-	-
Cellular necrosis	-	-	-	+	+	+	+	+	+	-	-	-
Haemorrhage	-	-	-	+	+	+	+	+	+	-	-	-
Mild Oedema	-	-	-	+	+	+	+	+	+	-	-	-
Wavy cardiac myofibrils	-	-	-	-	-	-	+	+	+	-	-	-
Cardiac infarction	-	-	-	-	-	-	-	-	-	+	+	+
Carbon deposit	-	-	-	-	-	-	-	-	-	+++	+++	+++

Table 6: Histopathological Observations of the Effect of Potash on the Heart of Wister Rats (Acute Phase)

Histopathological Observations	Control group A			Test group B			Test group C			Test group D		
	4	**5**	**6**	**4**	**5**	**6**	**4**	**5**	**6**	**4**	**5**	**6**
Vacoulations	-	-	-	+	+	+	+	+	+	-	-	-
Cellular necrosis	-	-	-	+	+	+	+	+	+	-	-	-
Haemorrhage	-	-	-	+	+	+	+	+	+	-	-	-
Mild Oedema	-	-	-	+	+	+	+	+	+	-	-	-
Ooedema with mild fibrolysis	-	-	-	-	-	-	+	+	+	-	-	-
Wavy cardiac myofibrils	-	-	-	-	-	-	+	+	+	-	-	-
Vascular carbon deposit	-	-	-	+	+	+	-	-	-	-	-	-

KEY: - = Negative; + = Mild positive; + = Severe positive

Photomicrographs (Histopathological Observations) (Balma)

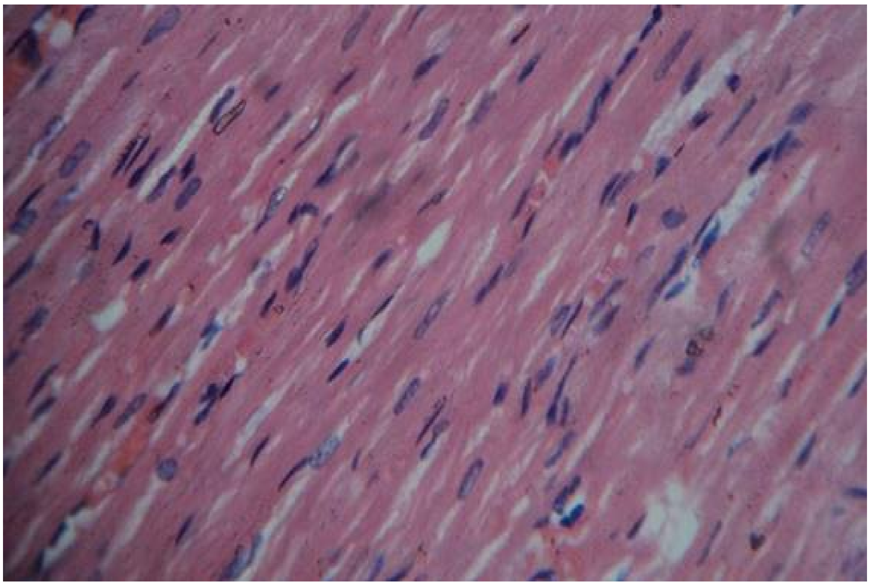

Figure 9: (H & E × 400) Control group A heart section (A5) showing normal myocardial cells and intact architectural integrity

Pre-Acute Phase

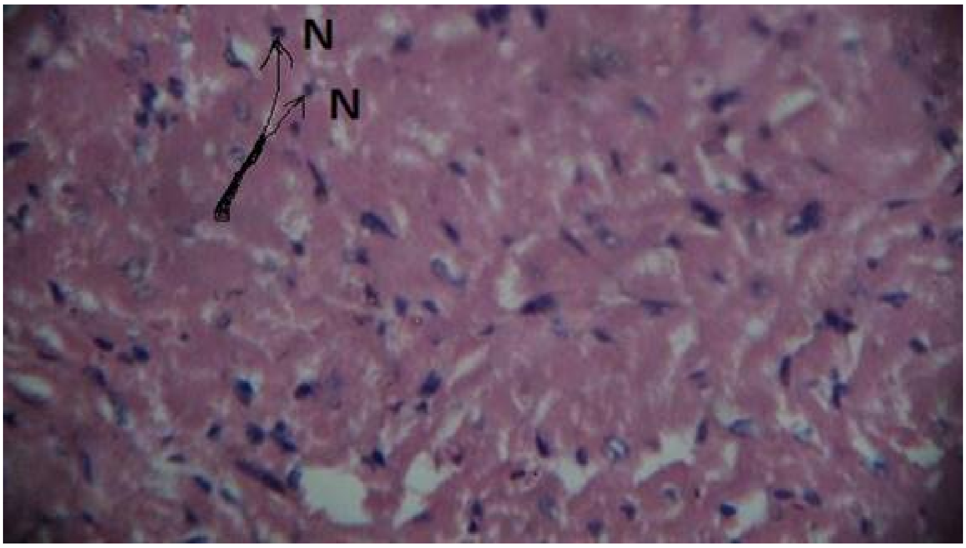

Figure 10: (H & E × 400) Test Group B (pre-acute phase) heart section showing necrosis (see arrow)

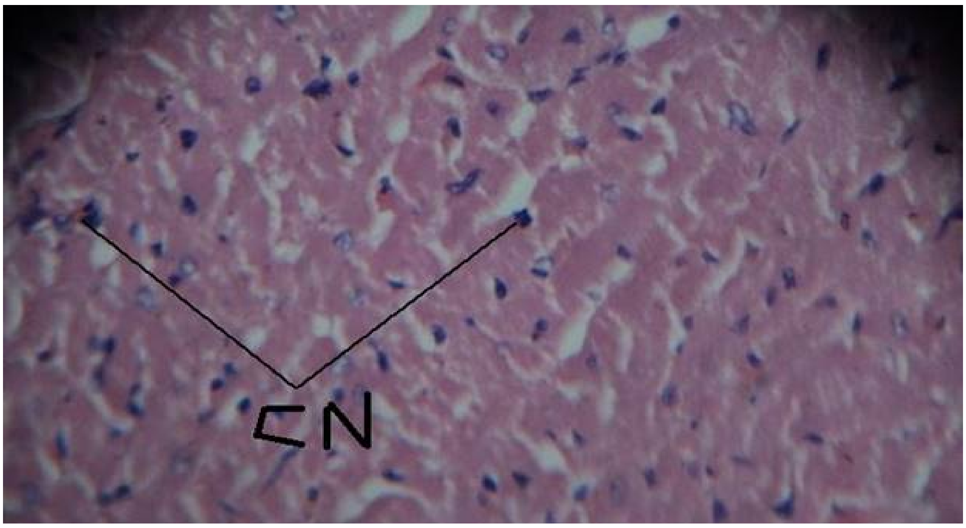

Figure 11: (H & E × 400) Test Group C: heart section showing cellular necrosis (see arrow)

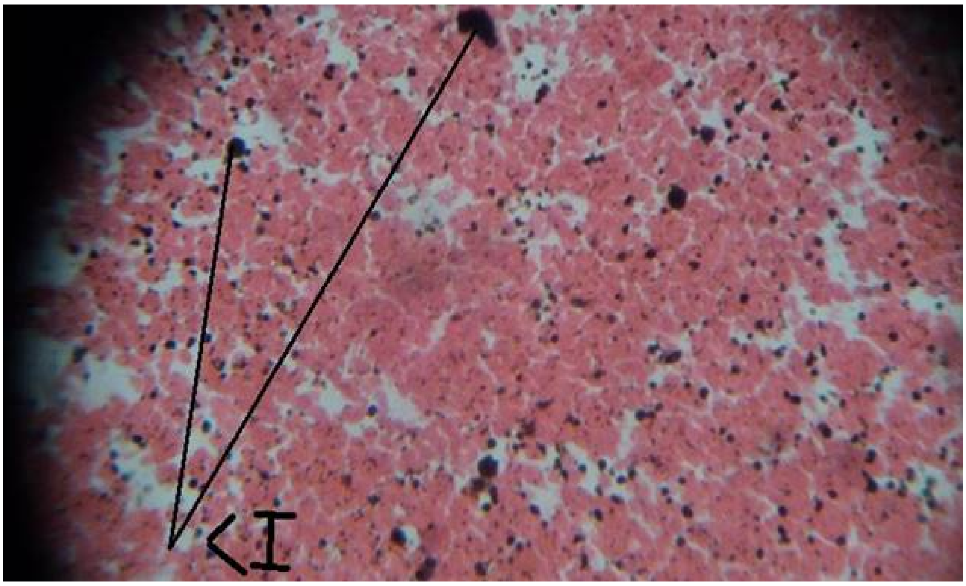

Figure 12: (H & E × 400) Test Group D: heart section showing cardiac infarction with deposit of carbon

Acute Phase

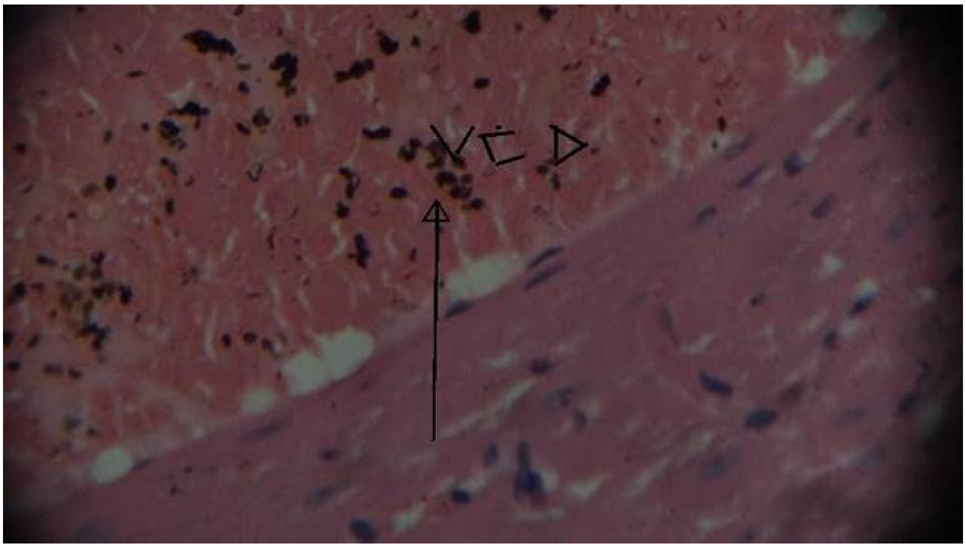

Figure 13: (H & E × 400) Test Group B (acute phase): heart section showing vascular carbon deposit (see arrow)

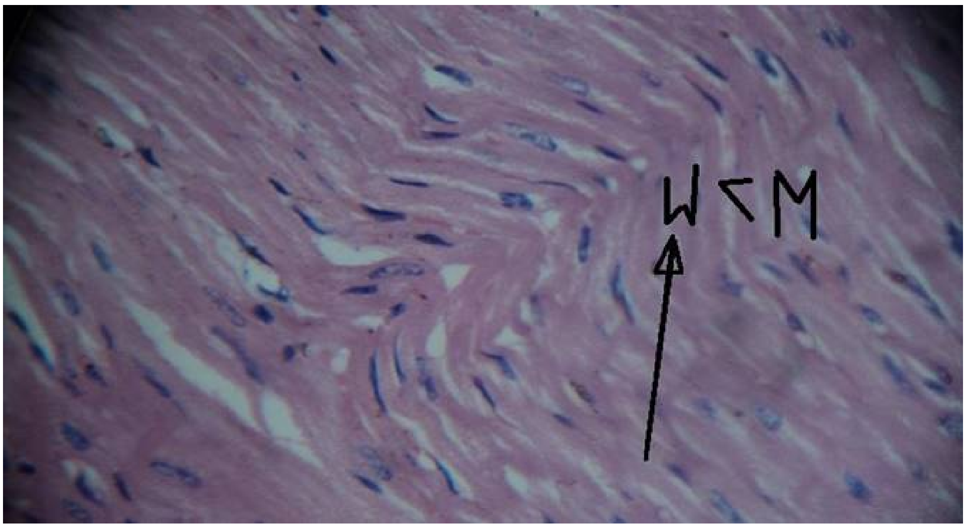

Figure 14: (H & E × 400) Test Group C (acute phase) heart section showing wavy cardiac myofibrils (see arrow)

3.3. Results from Manda (Potash)

Preliminary Observations

Table 7: Physical Observation of Control and Experimental Rats During Treatment with Manda kanwa

Physical observation	Experimental groups			
	Group A (Control)	Group B	Group C	Group D
Skin Changes	-	-	-	-
Fur Changes	-	-	-	+
Tail Changes	-	-	-	+
Eyes	-	-	-	-
Behaviour Pattern	-	+	+	+
Tremors	-	-	-	-
Diarrhea	-	-	-	-
Death	-	-	-	1

KEY: - = Absent; + = Change in behaviour; + = The tail was sloughing off

Histological Observations of the Heart (Manda kanwa)

Heart sections (H & E) $\times$ 400) showing (Group A) normal histological architecture i.e., the cellular integrity was well maintained.

Group B: The pre-acute phase of the treated group B showed varied degree of pathologic conditions which are vacuolations, cellular necrosis, haemorrhage as well as mild Oedema. Whereas, in the acute phase, the treated group B showed cellular necrosis, vacuolations, vascular carbon deposit, haemorrhage and Oedema.

Group C: Treated group C in the pre-acute phase showed severe vacuolations, cellular necrosis, Oedema and wavy cardiac myofibrils while the acute phase showed necrosis, vacuolations, Oedema, wavy cardiac myofibrils and Oedema with mild fibrolysis.

Group D: Treated group D, three out of six rats in this group died before the end of pre-acute phase. However, the histological result of the remaining three showed severe architectural distortion with numerous carbon deposit as well as cardiac infarction with deposit of carbon.

Table 8: Histopathological Observations of the Effect of Manda kanwa on the Heart of Wister Rats (Pre-Acute Phase)

Histopathological Observations	Control group A			Test group B			Test group C			Test group D		
	1	2	3	1	2	3	1	2	3	1	2	3
Vacoulations	-	-	-	+	+	+	+++	+++	+++	-	-	-
Cellular necrosis	-	-	-	+	+	+	+	+	+	-	-	-
Haemorrhage	-	-	-	+	+	+	+	+	+	-	-	-
Mild Oedema	-	-	-	+	+	+	+	+	+	-	-	-
Wavy cardiac myofibrils	-	-	-	-	-	-	+	+	+	-	-	-
Cardiac infarction	-	-	-	-	-	-	-	-	-	+	+	+
Carbon deposit	-	-	-	-	-	-	-	-	-	+++	+++	+++

Table 9: Histopathological Observations of the Effect of Manda kanwa on the Heart of Wister Rats (Acute Phase)

Histopathological Observations	Control group A			Test group B			Test group C			Test group D		
	4	5	6	4	5	6	4	5	6	4	5	6
Vacoulations	-	-	-	+	+	+	+	+	+	-	-	-
Cellular necrosis	-	-	-	+	+	+	+	+	+	-	-	-
Haemorrhage	-	-	-	+	+	+	+	+	+	-	-	-
Mild Oedema	-	-	-	+	+	+	+	+	+	-	-	-
Ooedema with mild fibrolysis	-	-	-	-	-	-	+	+	+	-	-	-
Wavy cardiac myofibrils	-	-	-	-	-	-	+	+	+	-	-	-
Vascular carbon deposit	-	-	-	+	+	+	-	-	-	-	-	-

KEY: - = Negative; + = Mild positive; + =Severe positive

Heart Photomicrographs (Manda kanwa)

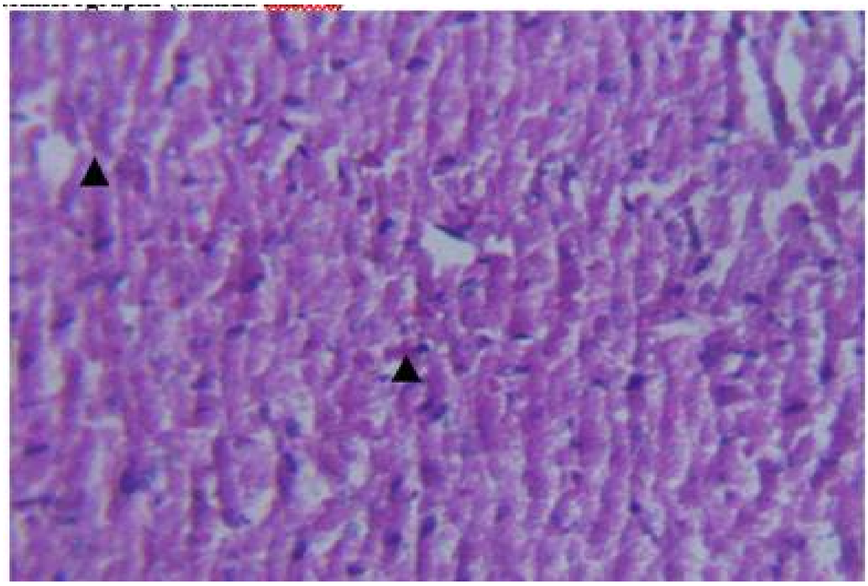

Figure 15: Heart control (GROUP A) shows normal myocardium at higher magnification (H/E *times* 400 mg)

Pre-Acute Phase

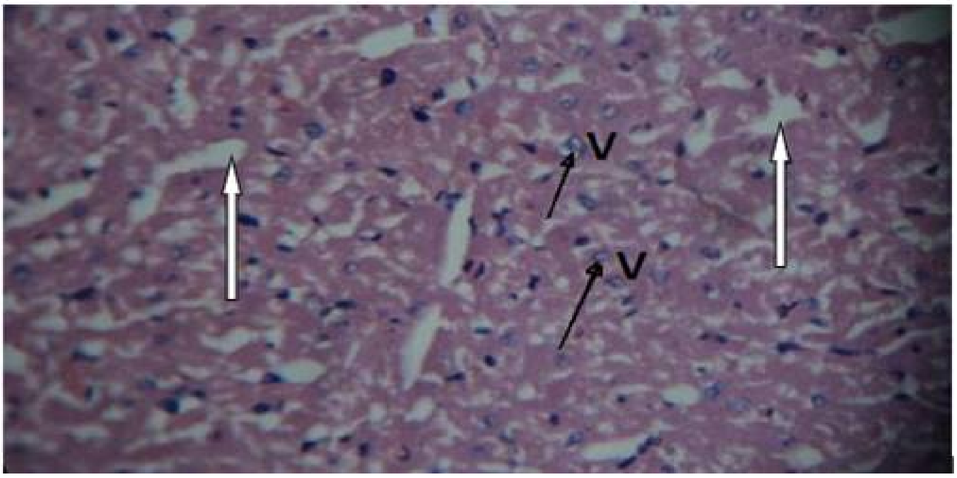

Figure 16: (H & E × 400) Test Group B (pre-acute phase): heart section showing vacuolations and oedema (black and white arrows respectively)

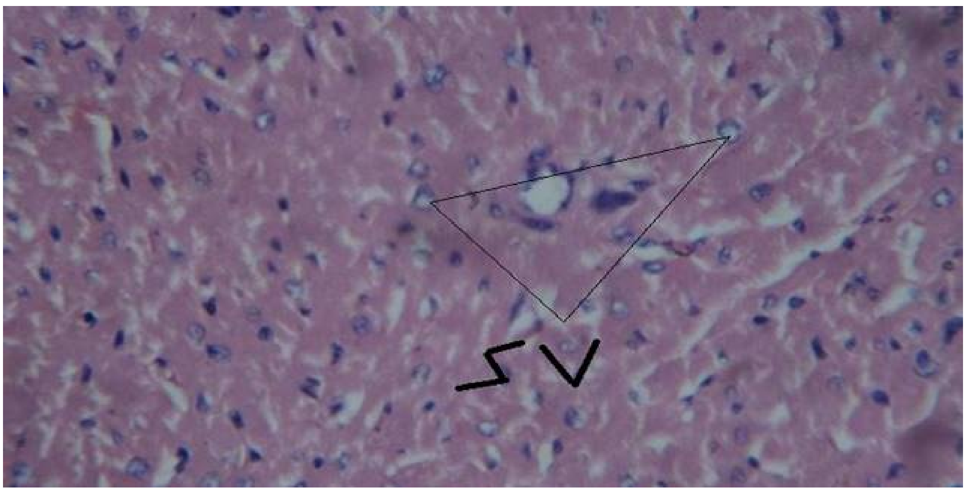

Figure 17: (H & E × 400)Test Group C (pre-acute phase) heart section showing severe vacuolations (see arrow)

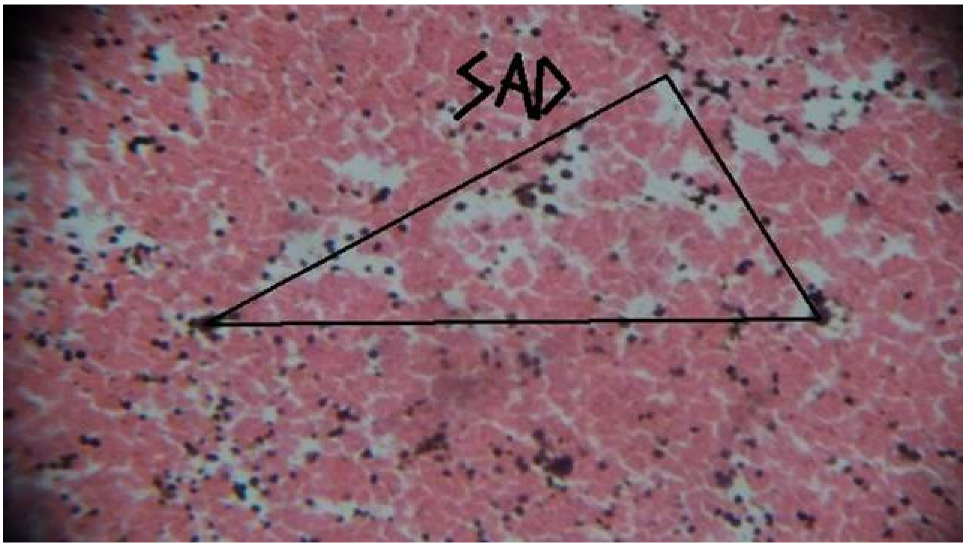

Figure 18: (H & E × 400)Test Group D: heart section showing severe architectural distortion with numerous carbon deposits (see arrow)

Acute Phase

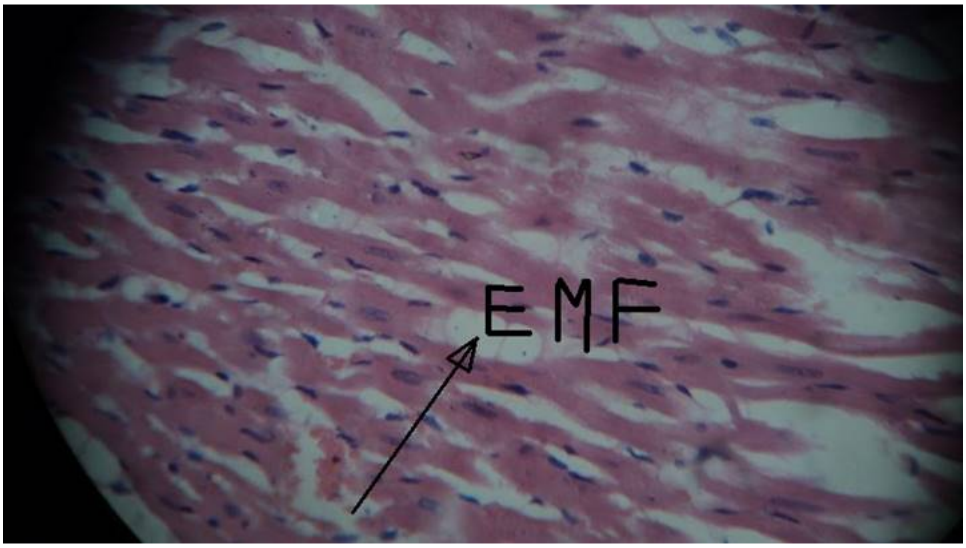

Figure 19: (H & E × 400)Test Group B (acute phase): heart section showing oedema with mild fibrolysis (see arrow)

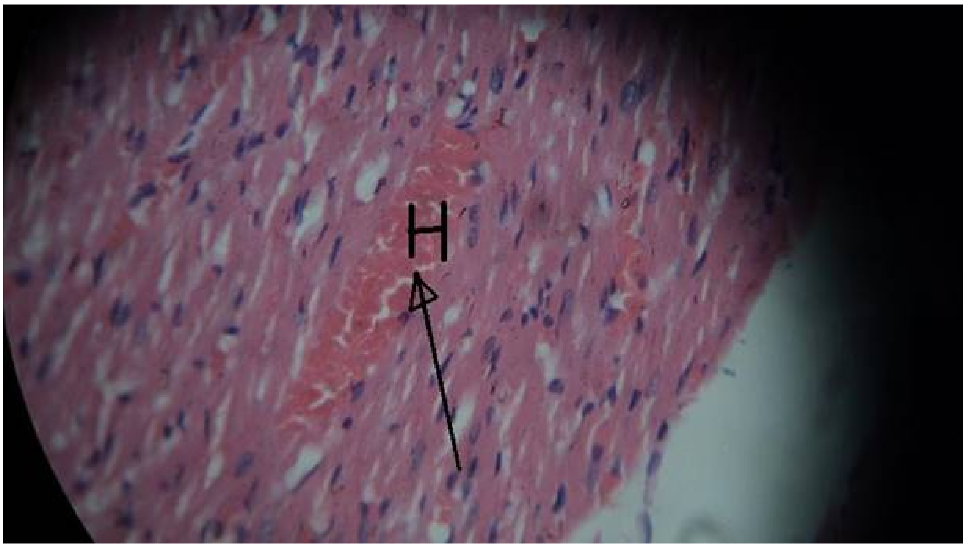

Figure 20: (H & E × 400) Test Group C (acute phase) heart section showing haemorrhage (see arrow)

4. Discussion

The results of the Haematoxylin and Eosin staining (H & E) reactions showed that kanwa exposure induced marked abnormalities in the heart initiated with disruption as seen in the cyto-architecture of each organ in the treated groups (B, C and D) which was at variance with that of the control group A. This could be as a result of the fact that experimental animals in this group were fed with normal feed as such no observable effects were seen. The micrograph of the heart sections in control group A showed normal histological features. The heart sections of the control group A showed normal myocardial cells and intact architectural integrity. But the results of group B in the pre-acute phase showed varying degree of distortions including vacuolations, cellular necrosis, haemorrhage and mild oedema. The result of the acute phase showed vascular carbon deposit in addition to the pathological distortions.

The kanwa administered to the experimental animals resulted in progressive cellular alterations as seen in the cytostructural distortion in the treated groups B, C and D which was at variance with that of the control group A. In test group D, only infarction with numerous carbon deposit were observed. There were no vacuolations, cellular necrosis, haemorrhage or oedema as present in groups B and C. It may be due to the fact that group D rats were not feeding well during the administration of the potash as a result of high concentration that may have greatly altered the taste of animal feed. The high concentration could possibly be the cause of death of three rats in test group D before the end of pre-acute phase. But there is no literature found at the time of this study to back my findings at the time of this experiment.

The kanwa administered to the experimental animals resulted in progressive cytoarchitectural distortion of the stomach in the treated groups (B, C and D) which was at variance with that of the control group A. The sections of group B animals fed with 3g of kanwa showed mild epithelia distortion and hyperplasia of neck mucus secreting cells. Slides from group C experimental animals fed with 6g of kanwa revealed similar distortion as in group B although more intense. The more severe effects observed in these slides are suggestive that as the concentration of kanwa increased, the defects induced became more severe. Furthermore, slide from the tissues of the experimental animals in group D revealed severe epithelial distortion and hyperplasia of neck mucus secreting cells. The results are further in line with the observed effects noticed in group C that indicated that destruction process is a function of dosage concentration. From these observations, kanwa could be referred to as been cytotoxic to the stomach tissues. However, at the time of this research, there was no documented report found to support these findings.

According to Rani, (2023), the extent of severity of tissue damage of a particular compound as toxicant depends on its toxic potentiality on the tissues of organisms. Also, susceptibility to chemical injury varies greatly in the tissues and cells of the same animal [23]. It is even greater in different animal groups. However, the location of the major damage may be determined by the mode of action of the chemical [24]. The mode of action of each poison and the pattern of tissue vulnerability has been well defined and the toxic level of each agent at which a fairly standard distinctive pattern of tissue damage has been studied. The level of potash consumed in various homes may not be toxic if not taken continuously or repeatedly as the results indicated that these effects are dosage dependent. It can therefore be inferred generally that more or less similar pathological changes are induced in the organs of the experimental rats but the extent of damages varies depending on the concentration of potash administered.

From the histopathologic distortion observed, it could be inferred from the results of this study that potash administration caused degenerative and destructive damages observed in the liver tissue sections. In view of the above observations, it is clear in this study that in every concentration of the administered potash caused considerable histological damages to the liver. It could, therefore, be deduced that the repeated consumption of potash by the experimental animals lead to serious toxicological implications on liver tissues and the extent of damages varies with potash concentration. The observed effects from this present study dispute the common believe that potash has no health implications [25].

Toxic necrosis occurs when cells are exposed to toxic substances while ischemic necrosis is as a result of oxygen deficiency as they are sensitive and susceptible to it due to their high metabolism [26]. Pathological cell death on the other hand has been described as apoptotic death and is an organized programmed cell death mediated by an active and intrinsic mechanism.

According to Xu et al., [27], the extent of severity of tissue damage of a particular compound as toxicant depends on its toxic potentiality on the tissues of organisms. Also, susceptibility to chemical injury varies greatly in the tissues and cells of the same animal [28]. It is even greater in different animal groups. However, the location of the major damage may be determined by the mode of action of the chemical [29]. The mode of action of each poison and the pattern of tissue vulnerability has been well defined and the toxic level of each agent at which a fairly standard distinctive pattern of tissue damage has been studied.

Histopathology is mainly directed to study the effect of chemicals on the structural components of the living system and the ways in which cells and tissues respond to injury. A chemical or a derivative acting directly on the cell or most frequently causes chemical cytotoxicity by altering its environment [28]. The cells in turn respond histopathologically by degeneration, proliferation, inflammation and repair.

This study revealed that excessive consumption of potash could result in the distortion and disruption of the microanatomy of the kidney and hence detrimental to human health. It is probable that the function of the kidney may be adversely affected. However, the results showed that the substance of study is cytotoxic to the heart tissue and degenerative changes were visible even at the lowest dosage of 3g. The report of this study is limited simply to a general statement about the overall architecture of the tissue. What this report adds to the literature is the perspective of the histological or microanatomy of the heart. The hypothesis of possible functional changes that may be detrimental to the health status of the animals would require further studies, including quantification of the structural changes.

5. Conclusion

The aim of using Kanwa on experimental rats was to determine the effects of its repeated use on the tissues of the heart and also to project a possible analogy with humans as it has become a major food supplement in most rural areas in Nigeria, Ghana, Cameroon and other West African countries. In view of the above observations in the results, it is clear in this study that in every concentration of the administered kanwa caused considerable histological abnormalities and cyto-architectural distortion of the visceral structures, which may be attributed to the cytotoxic effects of kanwa on the organs. It may be inferred from the present study that higher doses of kanwa resulted in increased toxic effect on the organs. The organs sections from group D tissues treated with higher doses of kanwa were most severely affected in this experiment.

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