African Scientist Vol. 24, No. 4 December 31, 2023
Printed in Nigeria

1595-6881/2023 $80.00 + 0.00

https://africansciientistjournal.org

AFS2023058/24405

Histological Changes in the Heart and Aorta of the Adult Wistar Rat Following Exposure to Cement Dust

Ehi-Omosun, Mabel Bilu and Olise, Nkechi Augustina

Department of Anatomy and Medical Laboratory Science, School of Basic Medical Sciences, College of Medical Sciences, University of Benin, Nigeria.

*Corresponding author Email: mabel.ehi-omosun@uniben.edu, Tel: +234 (0) 704 994 8914

(Received December 2, 2023; Accepted in revised form December 15, 2023)

ABSTRACT: The heart is a vital organ of the body that pumps blood through the aorta and its branches which provide blood flow to all organs and tissues of the body. Any injury causes distortion of the smooth functioning of the cardiovascular system. This study investigated the histological changes in the heart and aorta of Wistar rats following exposure to cement dust. Twenty-four (24) Wistar rats weighing between 250 g and 280 g were divided into 4 groups of 6 rats per group. Group A rats were placed in a cement dust free chamber while Group B - D rats were exposed to various concentrations of cement dust dispersed from 5 g, 10 g and 20 g of cement, respectively. The weights of the rats were taken weekly and the difference between them and previous weights were noted. At the end of the 30th day of exposure, the animals were euthanized under chloroform anaesthesia and the heart and aorta were harvested and processed for histological examination. The histological sections of the heart and aorta of rats in Group A revealed normal myocardial and vascular architecture. There were observable histological variations in the myocardial and vascular architecture of the exposed rats (Groups B-D) which include haemorrhages in the myocardial and aortic wall, vascular congestion as well as inflammation of the myocardial and aortic wall. These injuries are consistent with usual histological findings in myocarditis and aortitis. It was concluded that cement dust had histomorphological effects on the muscles of the heart and aorta which are capable of compromising the health of the research animals.

Keywords: Cement dust; Heart; Histoarchitecture; Myocarditis, Aortitis

Introduction

The heart is a fist-size organ that pumps blood throughout the body (Moore *et al.,* 2016). It's the primary organ of the circulatory system. The aorta is the largest artery of the body and it has branches that provide blood flow to all organs and tissues of the body (Moore *et al.,* 2016). Any injury to the heart and/or aorta causes distortion of the smooth functioning of the cardiovascular system. About 20,000 deaths occur every year in United States due to cardiovascular diseases (Wolf, 2012). Prolonged exposure to cement dust may cause cardiovascular damage because cement dust contains many hazardous chemicals, and if not corrected can lead to heart failure (Mark *et al.,* 2014).

Cement dust is an atmospheric pollutant which poses a significant threat to the environment and humans. It is emitted during the manufacturing and processing of cement, transportation, bag dumping and emptying, storage, usage and concrete cutting (Zahra and Samaneh, 2012; Chukwu and Ubosi, 2016). The basic components of cement dust include calcium, silicon, aluminium, manganese, iron and zinc (Alakija *et al.,* 2017; Yahaya *et al.,* 2011; Omigie *et al.,* 2020). Many of the chemical elements of cement dust are toxic or mutagenic to both animals and humans (Akinola *et al.,* 2019; Zeleke *et al.,* 2011; Nwafor *et al*., 2019).

Previous studies have shown that prolonged inhalation of cement dust can damage the heart, aorta, lungs, respiratory tract, stomach, liver, kidneys and blood/blood-forming organs (Saji *et al.,* 2018; Mark *et al.,* 2014; Christopher *et al.,* 2018). Signs and symptoms of cement dust related cardiovascular disorders include

substernal chest pain, chronic fatigue, palpitations, shortness of breath, dizziness, nausea and vomiting, (Mark *et al.*, 2014; Saji *et al*., 2018). Hence, the objective of this paper was to investigate the effects of cement dust on the heart and aorta of adult Wistar rats.

Materials and methods

Experimental animals: Twenty-four (24) adult Wistar rats, weighing between 250 g and 280 g purchased from the Animal house, Department of Anatomy, University of Benin, Nigeria, were utilized for this study. The animals were left to acclimatize for two (2) weeks before commencement of the experiment. During this period, they were allowed access to standard animal feed and water *ad libitum.*

Ethical considerations: Each animal procedure was carried out in accordance with approved protocols and in compliance with the recommendations for the proper management and utilization of laboratory animals used for research (Buzek and Chastel, 2010).

Experimental design: In this study, 24 animals were divided into 4 groups comprising of 6 rats per-group. Group A rats, which served as control, were placed in a cement dust-free dust distributor glass chamber (DDGC). Group B rats were exposed to cement dust dispersed from 5 g of cement via DDGC for 1 hour daily for 30 days. Group C rats were exposed to cement dust dispersed from 10 g of cement via DDGC for 1 hour daily for 30 days. Group D rats were exposed to cement dust dispersed from 20 g of cement via DDGC for 1 hour daily for 30 days. The weights of the animals in each group were taken and recorded weekly and the difference noted.

At the end of 30th day exposure, the animals were weighed, euthanized under chloroform anaesthesia and a midline incision was made through the ventral wall of the thorax of the rats to access the heart and aorta. The harvested organs were immediately fixed in 10% formal saline for 24 hours before the histological procedures. The tissues were trimmed to about 3-5 mm thick sections and processed according to the method of Drury and Wallington (2014). The trimmed tissues were histologically processed using the methods of fixation, embedding and tissue staining for microscopy. Histological sections were examined under a Leica DM750 research microscope with a digital camera (Leica ICC50) attached. Photomicrographs of the tissue sections were taken at magnification of x40 and x100.

Statistical analysis: Statistical analysis was carried out with Statistical Software Package, Microsoft Excel, (2010) and Statistical Package for Social Sciences (S.P.S.S.) version 20. Results were presented as Mean (X) ± Standard error of mean (SEM). The one-way Analysis of Variance (ANOVA) was used to determine the significance of the difference in means at 95% confidence interval. $P \leq 0.05$ was considered significant.

Results and Discussion

Changes in body weights of the animals in all the experimental groups are presented in Table 1. It was observed that there was a significant increase in body weights of the rats in Group A (Control Group) while Group B, C and D (Groups that were exposed to cement dust) showed a significant decrease in body weight.

Table1: Changes in body weights of the rats in all the experimental groups

Period of exposure	Group A	Group B	Group C	Group D	
Ist week	5.60 ± 0.68	0.60 ± 0.19*	0.42 ± 0.16*	0.38 ±0.16*	0.000
2nd week	6.70 ± 0.93	0.30 ± 0.05*	0.30 ± 0.09*	0.20± 0.14*	0.000
3rdweek	7.40 ± 1.24	0.06 ± 0.17*	0.20 ± 0.05*	0.16± 0.07*	0.000
4thweek	7.74 + 0.60	0.18 ± 0.09*	0.36 ± 0.10*	0.04± 0.08*	0.000

n=6; Values are Mean ± S.E.M

Cement dust inhalation has been implicated in a variety of maladies including systemic lupus erythematosus, restrictive lung disease, occupational asthma, and other cement factory lung diseases both in animals and humans. However, due to the unavailability of appropriate chamber for laboratory use, investigations to determine the impact of cement dust on the organs of cardiovascular system in Wistar rats and the extent of the associated toxicity have been limited (Nwafor *et al.,* 2019. Against this background, this study was conducted to evaluate the effects of cement dust inhalation in the heart and aorta of Wistar rats.

As shown in Table 1, there was a significant increase in body weight of rats in the control group (Group A). A significant decrease was observed in the body weight of rats in Group B, C and D that were exposed to cement

dust which was what we actually expected because cement dust is toxic and so, it's expected to cause a decrease in body weight and this concurs with previous work (Yahaya *et al.,* 2011; Nwafor *et al.,* 2019). The weight loss could be attributable to dysgeusia (El-Hilaly *et al.,* 2016), anorexia (Akinola *et al.,* 2019) or toxicity of the basic constituent chemical elements of cement dust (Hahaya *et al.,* 2011).

Figures 1 and 2 are micrographs of a section of rat's heart in the control group (Group A), (H&E at x 40 and x 100) magnifications respectively showing normal architecture of A, cardiac muscle and B, blood vessels. Figures 3 and 4 are photomicrographs of a section of rat's heart exposed to cement dust dispersed from 5 g of cement. (Group B), H&E at x 40 and x 100 magnifications respectively showing A, haemorrhage in the myocardial wall (intramyocardial haemorrhage), B, myocardium. Figures 5 and 6 are photomicrographs of a section of rat's heart exposed to cement dust dispersed from 10 g of cement. (Group C), H&E at x 40 and x 100 magnifications respectively showing A, myocardium, B, haemorrhage in the myocardial wall (intramyocardial haemorrhage). Figures 7 and 8 are photomicrographs of a section of rat's heart exposed to cement dust dispersed from 20 g of cement. (Group D), H&E at x 40 and x 100 magnifications respectively showing A, myocardium, B, haemorrhage in the myocardial wall (intramyocardialhaemorrhage) and C, vascular congestion.

Figures 9 and 10 are photomicrographs of a section of rat's aorta in the control group (Group A), H&E at x 40 and x 100 magnifications respectively showing normal architecture of A, lumen, B, aortic wall and C, adventitia. Figures 11 and 12 are photomicrographs of a section of rat's aorta exposed to cement dust dispersed from 5 g of cement. (Group B), H&E at x 40 and x 100 magnifications respectively showing A, lumen containing blood, B, aortic wall (tunica media), C, fibro-fatty tunica adventitial tissue and D, heavy lymphocytic infiltrates of inflammatory cells. Figures 13 and 14 are photomicrographs of a section of rat's aorta exposed to cement dust dispersed from 10 g of cement (Group C). Figure 13 shows A, aortic lumen containing blood, B, aortic wall, C, lymphocytic infiltrates of inflammatory cells surrounding the vessel (H&E at x 40) while Figure 14 shows A, aortic wall, B, lymphocytic infiltrates of inflammatory cells and C, haemorrhage in the aortic lumen (H&E at x 100). Figures 15 and 16 are micrographs of a section of rat's aorta exposed to cement dust dispersed from 20 g of cement. (Group D). Figure 15 shows A, lymphocytic infiltrates of inflammatory cells surrounding the vessel B, blood in the aortic lumen and C, the aortic wall (H&E at x 40). Figure 16 shows A, the aortic wall, B, blood in the aortic lumen and C, heavy lymphocytic infiltrates of inflammatory cells surrounding the vessel (H&E at x 100).

The histological sections of the heart and aorta of the control group (Group A) show normal histoarchitecture of the myocardium and aortic wall. There were observable histological variations in the heart and aorta histoarchitecture of the rats exposed to cement dust dispersed from 5g (Group B), 10g (Group C) and 20g (Group D) of cement respectively. The histological findings include inflammation of the myocardial and aortic tissue, haemorrhages in the myocardial and aortic wall, vascular congestion and activation of myocardial and aortic lymphoid aggregates. These findings agree with a similar work done by Poinen-Rughooputh *et al.* (2016) where they used silica dust to induce coronary heart disease.

The histomorphological changes indicate diseases and pathological symptoms of a variety of maladies including aortitis and myocarditis It was concluded that cement dust had histomorphologic effects on the tissues of the heart and aorta which are capable of compromising the health of the research animals.

The histological sections of the heart and aorta of the control group (**Group A**) show normal histoarchitecture of the myocardium and aortic wall. There were observable histological variations in the heart and aorta histoarchitecture of the rats exposed to cement dust dispersed from 5g (**Group B**), 10g (**Group C**) and 20g (**Group D**) of cement respectively. The histological findings include inflammation of the myocardial and aortic tissue, haemorrhages in the myocardial and aortic wall, vascular congestion and activation of myocardial and aortic lymphoid aggregates. These findings agree with a similar work done by Poinen-Rughooputh *et al.,* (2016) where they used silica dust to induce coronary heart disease.

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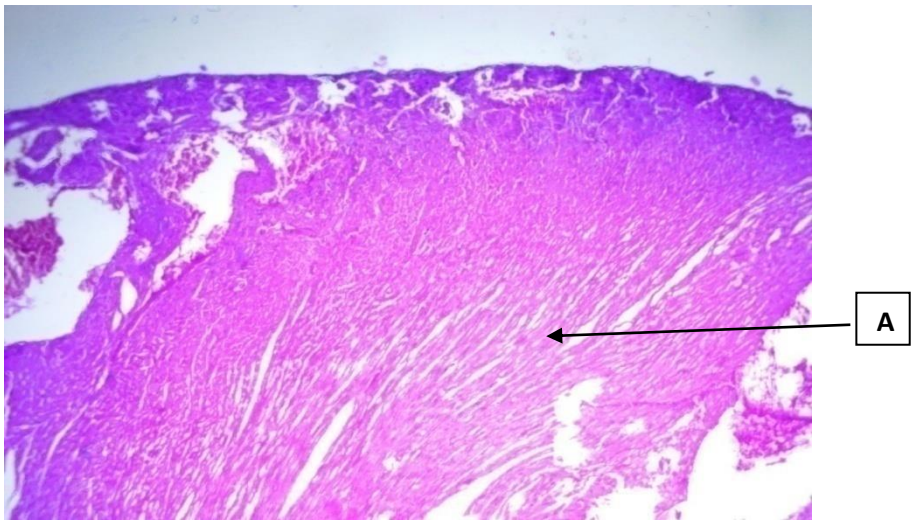

Figure 1: Photomicrograph of heart of rats in the control group (Group A) showing: A, bundles of myocardial fibres (H&E x 40)

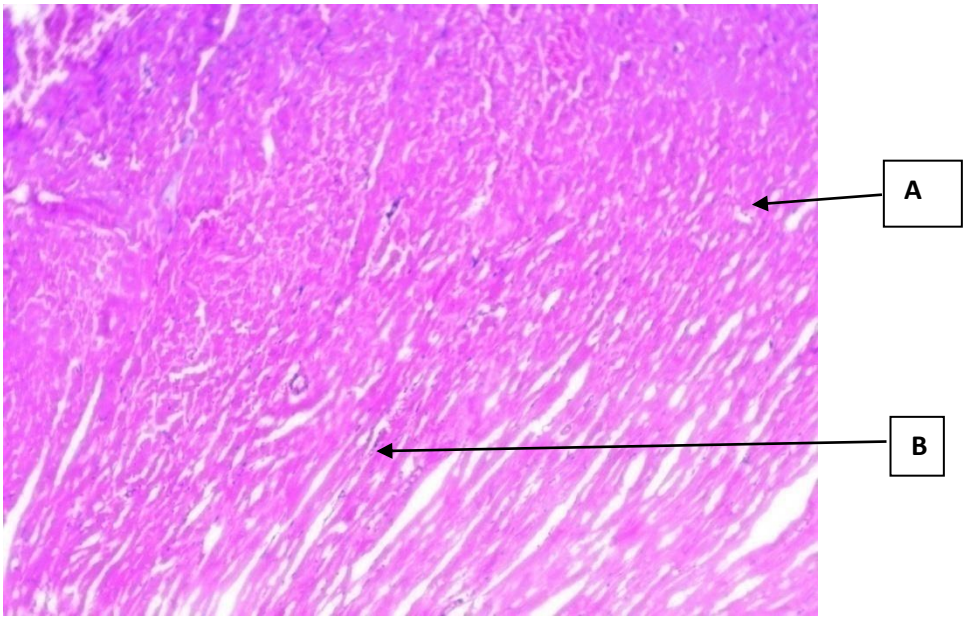

Figure 2: Photomicrograph of heart of rats in the control group (Group A) showing: A, bundles of cardiac muscle, B, blood vessel (H&E x100)

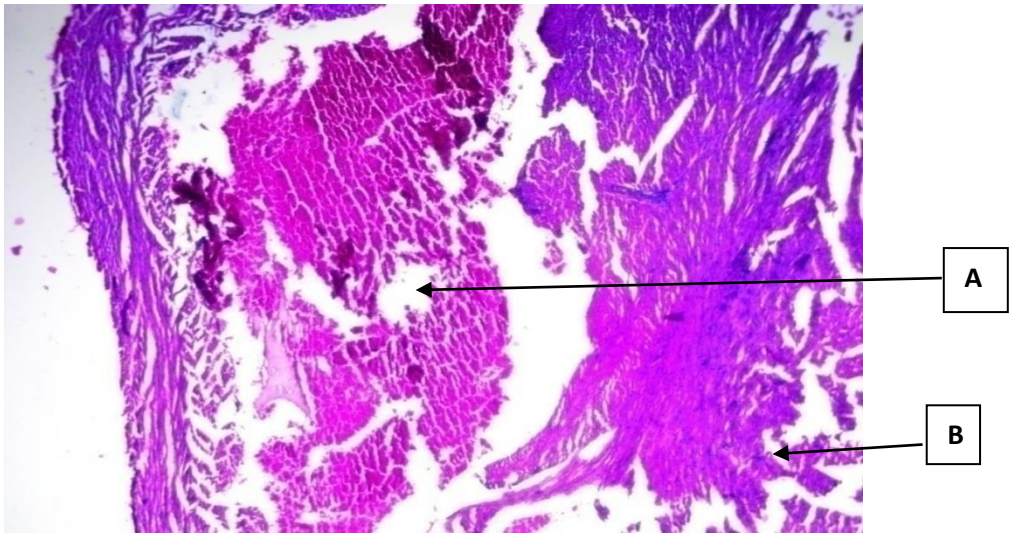

Figure 3: Photomicrograph of rat heart exposed to 5 g cement dust (Group B) showing: A, haemorrhage in the myocardial wall, B, cardiac muscle fibres (H&E x 40)

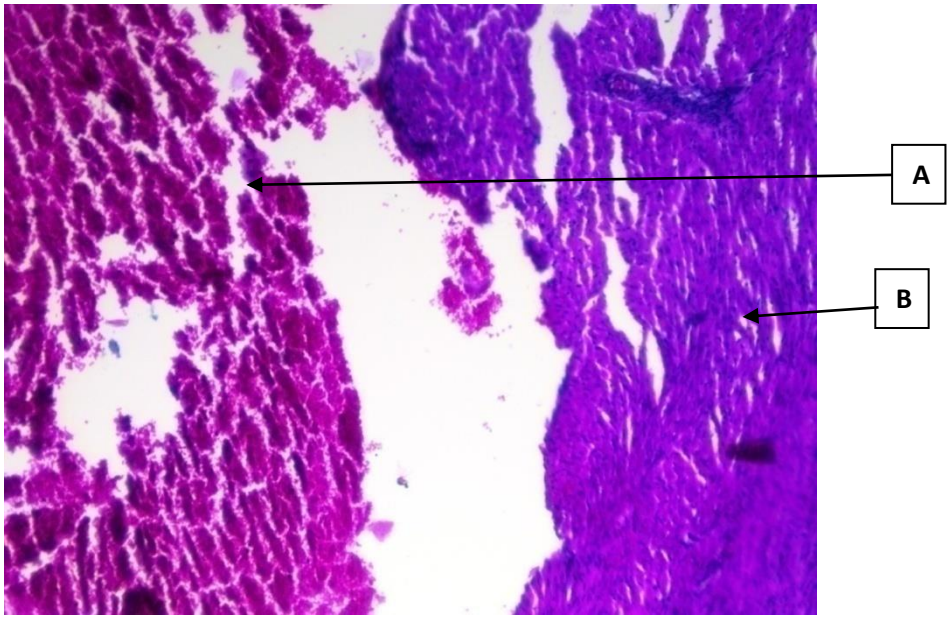

Figure 4: Photomicrograph of rat heart exposed to 5 g cement dust (Group B) showing: A, haemorrhage in the myocardium (intramyocardial haemorrhage, B, cardiac muscle fibres (H&E x 100)

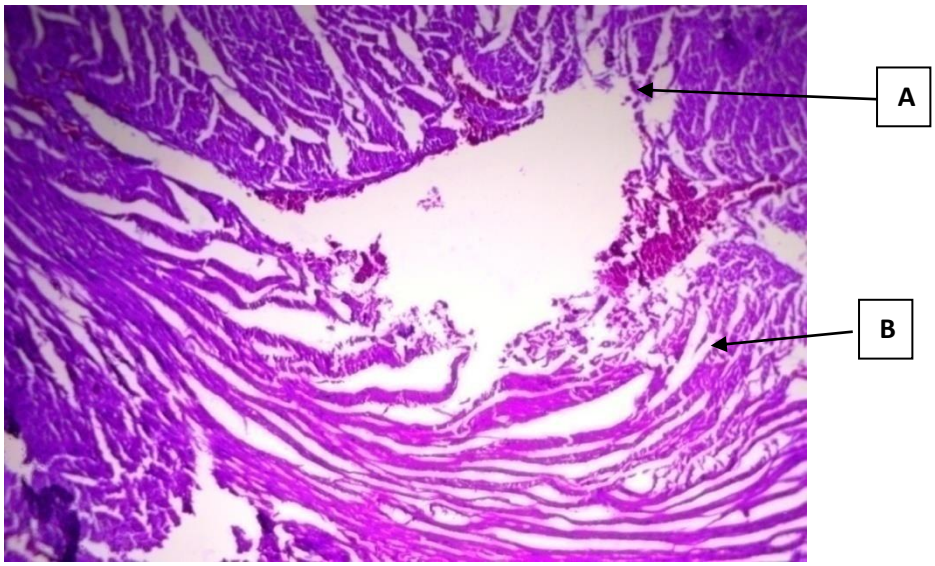

Figure 5: Photomicrograph of rat heart exposed to 10 g cement dust (Group C) showing: A, myocardium and B, intramyocardial haemorrhage (H&E x 40)

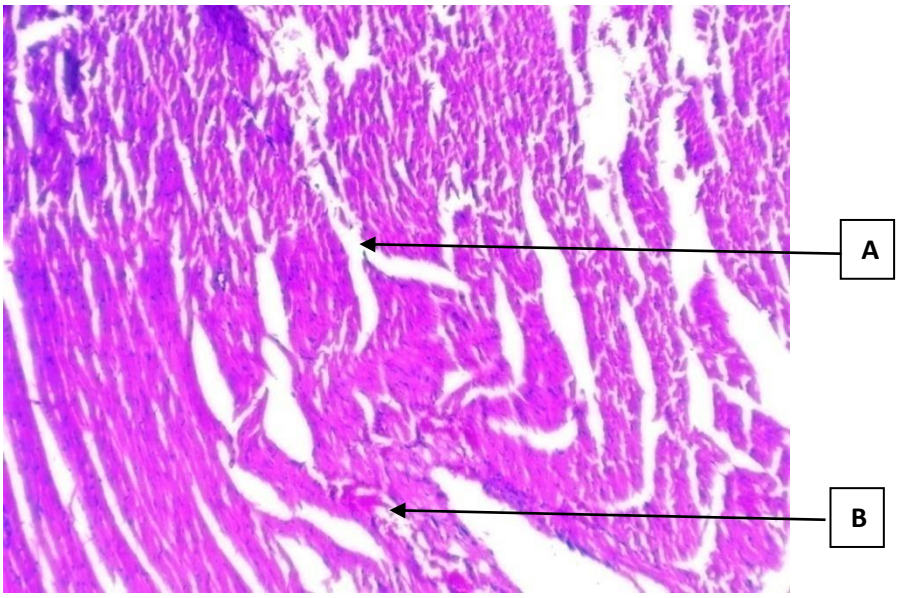

Figure 6: Photomicrograph of rat heart exposed to 10 g cement dust (Group C) showing: A, myocardium and B, intramyocardial haemorrhage (H&E x 100)

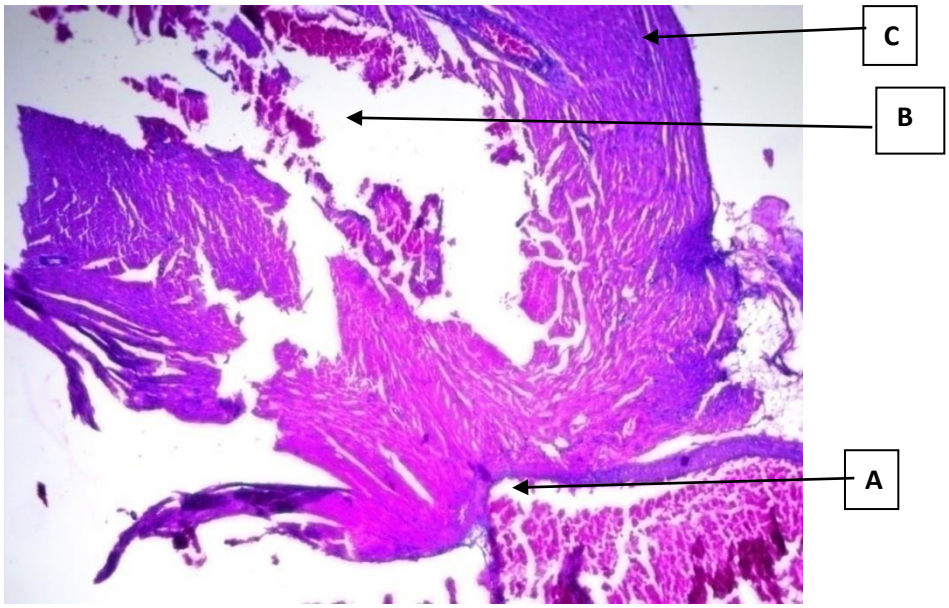

Figure 7: Photomicrograph of rat heart exposed to 20 g cement dust (Group D)showing: A, myocardium, B, intramyocardial haemorrhage and C, vascular congestion (H&E x 40)

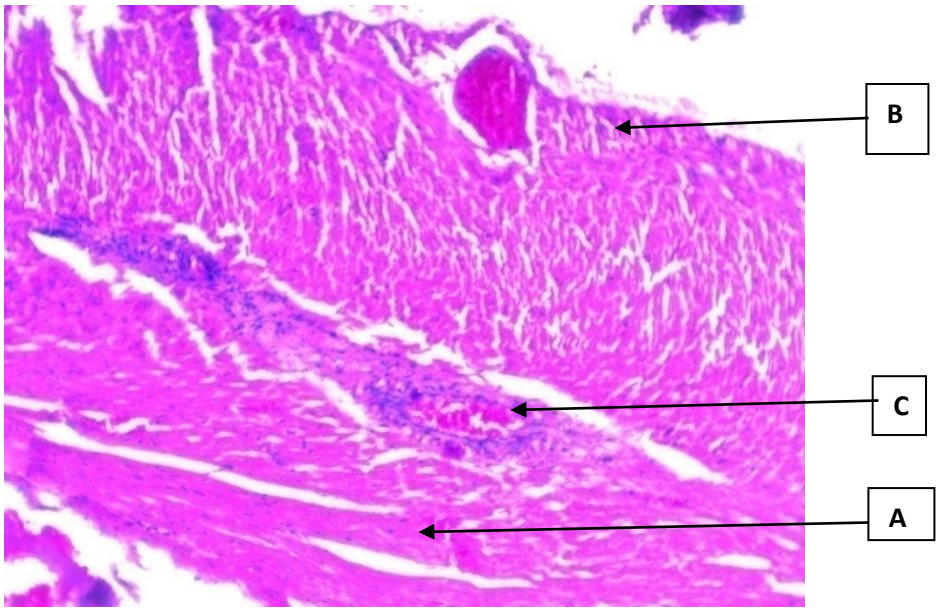

Figure 8: Photomicrograph of rat heart exposed to 20 g cement dust (Group D) showing: A, myocardium, B, intramyocardial haemorrhage and C, vascular congestion (H&E x 100)

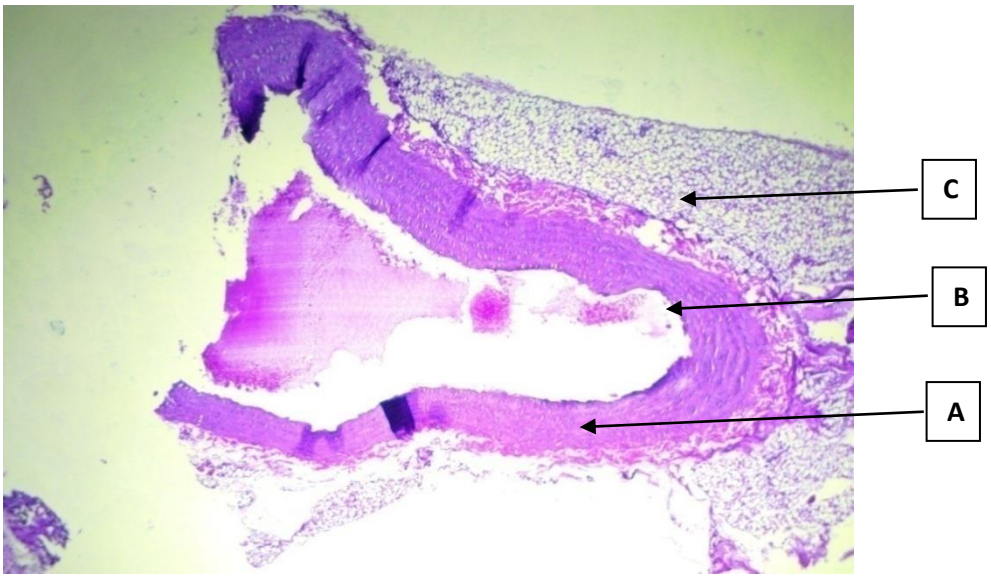

Figure 9: Photomicrograph of rat aorta in the control group (Group A) showing: A, lumen with blood in it, B, the wall and C, fibro-fatty tissue and adventitia (H&E x 40)

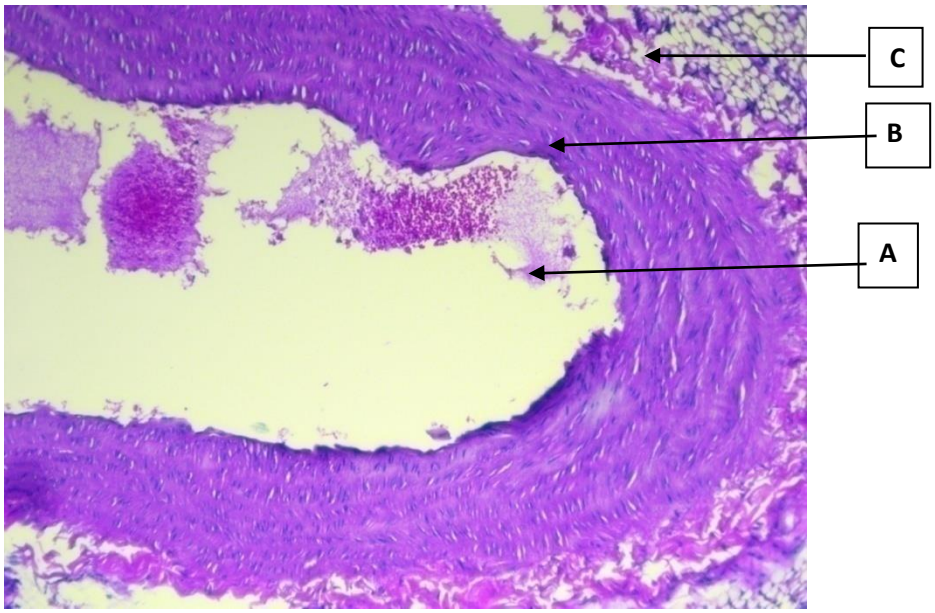

Figure 10: Photomicrograph of rat aorta in the control group (Group A) showing: A, lumen with blood in it, B, aortic wall, C, fibro-fatty tissue and adventitia (H&E x 100)

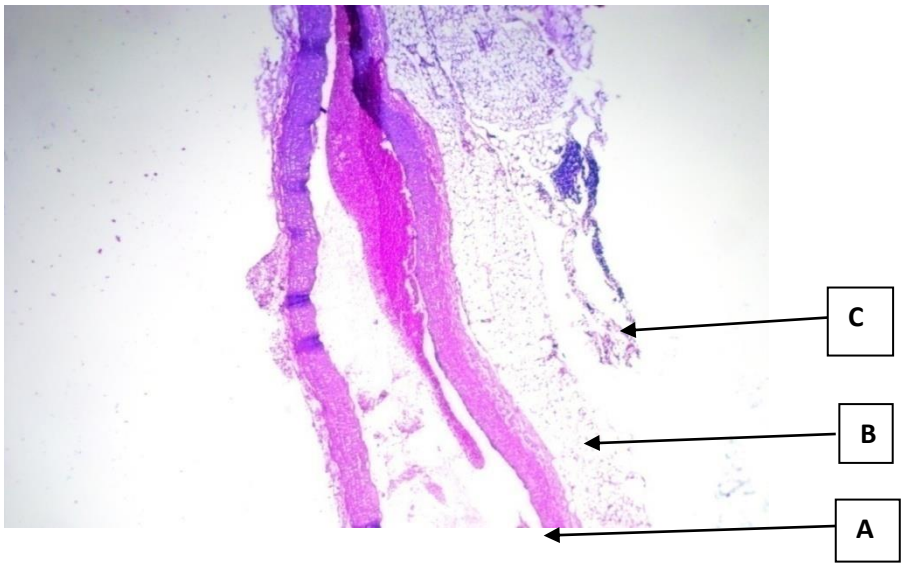

Figure 11: Photomicrograph of rat aorta exposed to 5 g cement dust (Group B) showing: A, aortic lumen containing blood, B, tunica media, C, fibro-fatty tunica adventitial tissue (H&E x 40)

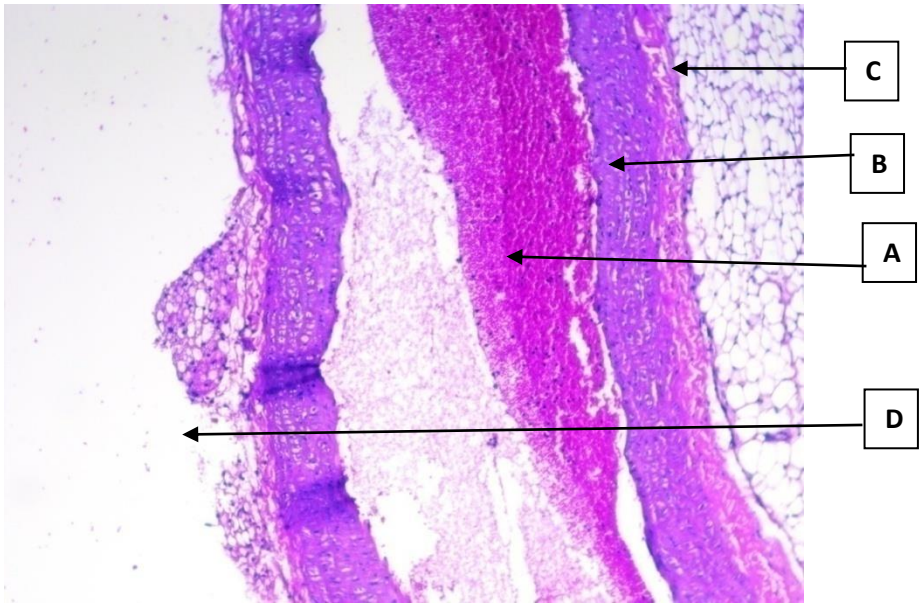

Figure 12: Photomicrograph of rat aorta exposed to 5 g cement dust (B) showing: A, aortic lumen containing blood, B, tunica media, C, fibro-fatty tunica adventitial tissue, D, heavy lymphocytic infiltrates of inflammatory cells surrounding the vessel(H&E x 100)

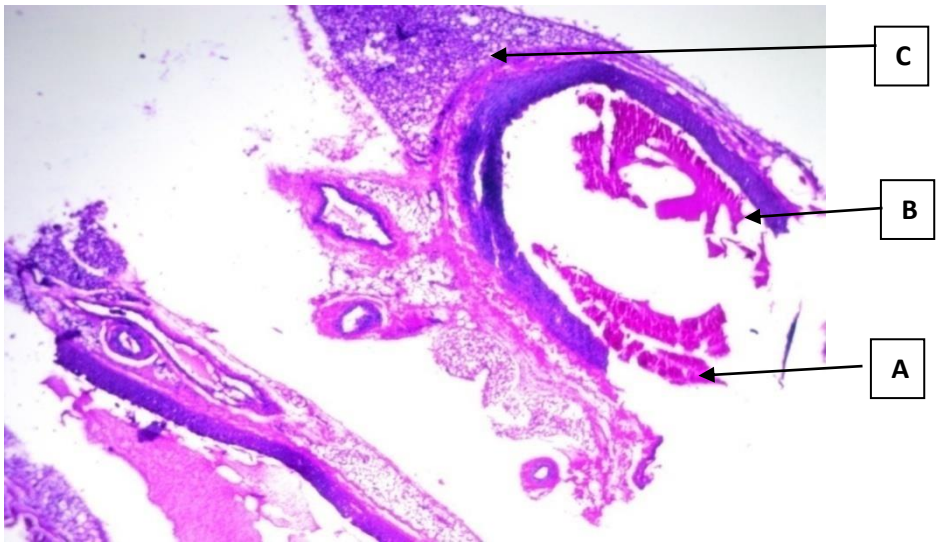

Figure 13: Photomicrograph of rat aorta exposed to 10 g cement dust (GropC) showing: A, aortic lumen containing blood, B, aortic wall, C, lymphocytic infiltrates of inflammatory cells surrounding the vessel (H&E x 40)

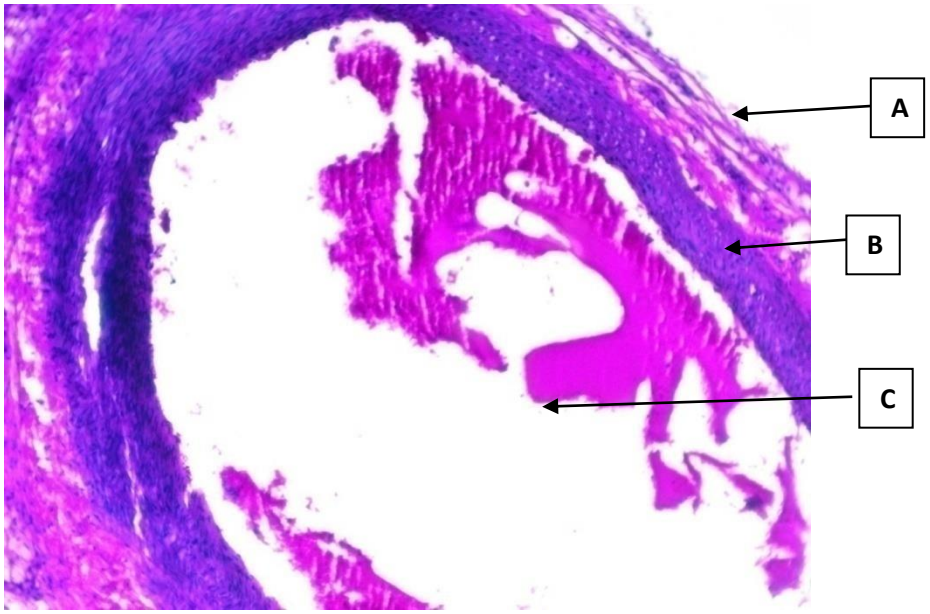

Figure 14: Photomicrograph of rat aorta exposed to 10 g cement dust (Grop C) showing: A, aortic wall, B, lymphocytic infiltrates of inflammatory cells surrounding the vessel, C, blood within the aortic lumen (H&E x 100)

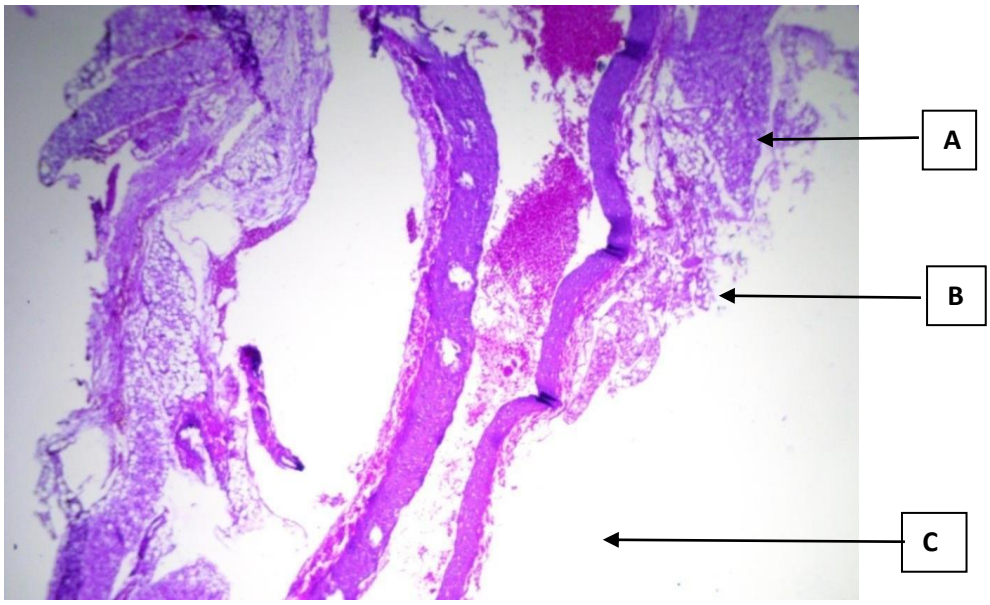

Figure 15: Photomicrograph of rat aorta exposed to 20 g cement dust (Group D) showing: A, lymphocytic infiltrates of inflammatory cells surrounding the vessel, B, blood in the lumen, C, aortic wall (H&E x 40)

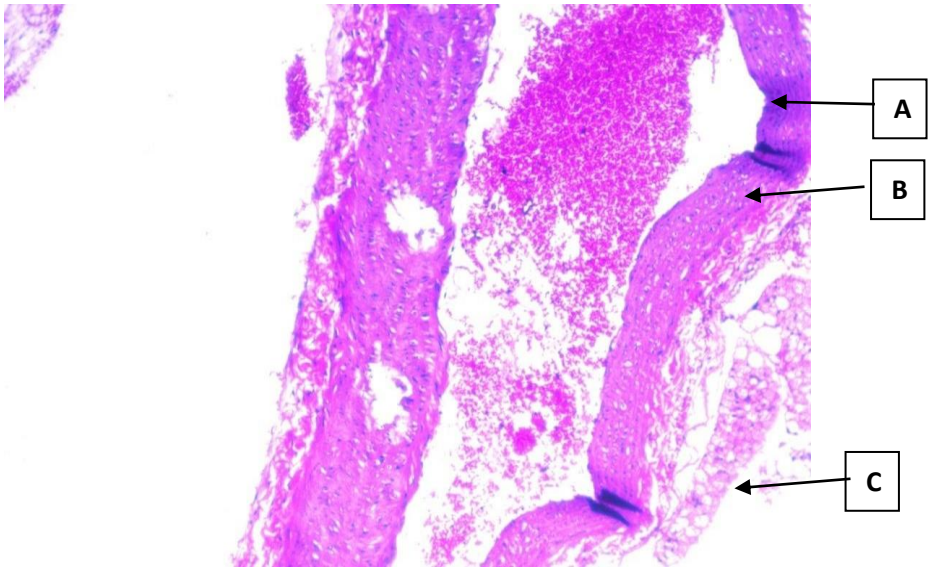

Figure 16: Photomicrograph of rat aorta exposed to 20 g cement dust (Group D) showing: A, the aortic wall, B, blood in the aortic lumen, C, lymphocytic infiltrates of inflammatory cells surrounding the vessel (H&E x 100).

Conclusion

Cement dust causes body weight loss and distorted heart and aorta histoarchitecture which are consistent with usual histological findings in myocarditis and aortitis which may ultimately lead to mortality of the research animals.

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