

Histopathological Analysis of Talc in Doxorubicin Induced Cardiac Remodelling

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Abstract—

Cardiac remodelling may be defined as apparently changes in size, shape and function of the heart after cardiac damage. Present study was aimed as to evaluate the histological assessment of Talc in doxorubicin induced cardiac remodelling in albino Wistar rat. Albino Wistar Rats (100-150gm) were separated in 8 groups each group have 6 rats, cardiac remodelling were induced by Doxorubicin with a 2.0 mg/kg i.p. two times in a week for 20 weeks and oral dose of talc (0.14mg/kg) for the period of 20 weeks and treated with Paracetamol for 5 day. Doxorubicin and Talc administration were dilated chamber and valve region of wistar rats. In the conclusion, Talc may promote cardiac remodelling in Wistar rats.

Keywords— Cardiac remodelling, Talc, left ventricles, Doxorubicin, albino wistar rats.

I. INTRODUCTION

Cardiac remodelling is a major determinant of heart failure (HF) [1] and is involves myocyte hypertrophy along with interstitial cells proliferation and extracellular matrix remodelling, induces structural and functional changes mainly in the left ventricle. Remodelling is initially a beneficial compensatory process due to decreased wall stress and increased cardiac output, but ultimately results in the inability of heart to meet hemodynamic demands [2].

Talc is a hydrated magnesium and layered silicate with chemical formula $Mg_3Si_4O_{10}(OH)_2$, that has unique properties, such as softness, chemical inertness, high thermal stability, low electrical conductivity, lamellar habit, adsorption properties, and occurrence of a wide range of particle sizes thus can be easily reduced by milling [3], [4] and high specific surface area, therefore talc is widely used in many industries, such as ceramics, paint, papermaking, roofing, plastics, rubber, and cosmetic productions [5]-[7].

This protocol was based on results of earlier research work which on the pharmacological action of excipient in anticancer drug, have given an influence that Talc may induced Type-2 diabetes [8]; furthermore many researcher said that diabetes act as promoter for a significant percentage of patients with a diagnosis of heart failure in numerous epidemiological studies [9]. The United Kingdom Prospective Diabetes Study [10], Cardiovascular Health Study [11], and Euro Heart Failure Surveys [12] all suggested that the presence of diabetes may independently increase the risk of developing incident cardiac disorder; Thus Cardiac remodelling protocol was designed.

II. MATERIAL AND METHOD

Albino Wistar male/female rats weighing about 100-150 gm, were acquired from Siddhartha institute of pharmacy, Dehradun. The Albino Wistar rats were housed under temperature ($25 \pm 2^\circ C$) and relative humidity (30-70%) with a 12:12 light-dark cycle, and acclimatized in the animal house facility of the department under ambient condition. The animals were fed with standard semi purified diet and water. All the experiments on animals were conducted in accordance with the internationally accepted principles for laboratory animal use and as per the experimental protocols duly approved by the Institutional Ethical Committee (IAEC No. 1435/PO/a/11/CPCSEA) with the project approval no- "SIP/IAEC/02/2014" by the Institutional Animal Ethical Committee (IAEC) of Siddhartha College of pharmacy, Dehradun. The rats were acclimatized and randomly divided into 08 groups and each group have 6 rats; (1) **Normal control (NC)** - Rats were given with normal diet and were not given any treatment during the research study. (2) **DOX Control**- Rats of this group were administered Doxorubicin with a 2.0 mg/kg i.p. twice in a week for 20 weeks (3) **Talc control**- Rats of this group were administered with Talc with a 0.14mg/kg oral dose for 20 weeks. (4) **PCM control**- Rats of this group were administered with normal diet for 20 weeks and after that were given PCM (500 mg/kg/d) for 5 days. (5) **DOX +Talc control**- Rats of this group were administered Doxorubicin with a 2.0 mg/kg body weight i.p. twice in a week and also administered Talc with a 0.14mg/kg oral dose for 20 weeks. (6) **DOX +PCM control**- Rats of this group were administered Doxorubicin with a 2.0 mg/kg body weight i.p. twice in a week and were given PCM (500 mg/kg/d) for 5 days. (7) **Talc+PCM control**- Rats of this group were administered Talc with a 0.14mg/kg oral dose for 20 weeks and were given PCM (500 mg/kg/d) for 5 days. (8) **DOX+Talc+PCM control**- Rats of this group were administered Doxorubicin with a 2.0 mg/kg body weight i.p. twice in a week and administered Talc with a 0.14mg/kg oral dose for 20 weeks and were given PCM (500 mg/kg/d) for 5 days.

Induction of cardiac remodelling

Cardiac remodelling was induced by a Doxorubicin dose of 2.0 mg/kg body weight two times in a week [13].

Histopathological Analysis

On the day of termination, the animals were sacrificed and rat's hearts were isolated. After blotting free of blood and tissue fluids, organ was fixed in 5% formalin. After 24 hours the tissues were washed thoroughly in repeated changes of 70% alcohol and then dehydrated in ascending grades of alcohol (70-100%). Dehydration in absolute alcohol was followed by treatment of tissues with toluene: xylene (50:50) followed by 10%, 50%, 70%, 90% paraffin wax in toluene and finally embedded with 100% paraffin wax at 60-62°C. 5-15µm thick section was serially cut on a leitz microtome in horizontal plane and mounted in glass slide with the help of egg albumin in glycerin solution (50% v/v). The sections were deparaffined in xylene and downgraded through 100%, 90%, 70%, 50%, & 30% alcohol and finally in water. They were then stained with 10% hematoxylin for 3-5 minutes and the staining was intensified by placing in running water. The hematoxylin stained sections were stained with 10% eosin for 2 minutes and were then quickly passed through ascending grades of alcohol and finally treated with xylene followed by mounting in DPX, The sections were observed by an experienced.

III. RESULTS

Histological assessment demonstrated the following results:

- 1. Normal Control** showed bundles of cardiac muscle fibers in syncytium, nuclei are centrally located, no atypia is seen, no inflammatory cells are noted, pericardium and endocardium are histologically are not significant, chambers and valves are histologically unremarkable, no evidence of toxicity is seen [Fig. no 1-(I)].
- 2. DOX Control** illustrated that bundle of cardiac muscle fibers in syncytium, nuclei are centrally located, no atypia is seen, pericardium and endocardium are histologically are not significant, chambers and valves are histologically remarkable, occasional toxic changes are seen especially in valve region [Fig. no 1-(J)].
- 3. Talc Control** demonstrated that bundles of cardiac muscle fibers in syncytium, nuclei are centrally located, no atypia is seen, no inflammatory cells are noted, pericardium and endocardium are histologically are not significant, chambers and valves both appear dilated [Fig. no 1-(K)].
- 4. PCM Control** explained that bundles of cardiac muscle fibers in syncytium, nuclei are centrally located, no atypia is seen, no inflammatory cells are noted, pericardium and endocardium are histologically are not significant, chambers and valves are histologically unremarkable, no evidence of toxicity is seen [Fig. no 1-(L)].
- 5. DOX+Talc Control** presented that bundles of cardiac muscle fibers in syncytium, nuclei are centrally located, no atypia is seen, no inflammatory cells are noted, pericardium and endocardium are histologically are not significant, chambers and valves are histologically unremarkable, valve appears dilated, areas of haemorrhage are noted [Fig. no 1-(M)].
- 6. DOX+PCM Control** showed that bundles of cardiac muscle fibers in syncytium, nuclei are centrally located, no atypia is seen, no inflammatory cells are noted, pericardium and endocardium are histologically are not significant, chambers and valves are histologically unremarkable, chambers and valves are histologically remarkable, irregularity of valve is noted [Fig. no 1-(N)].
- 7. Talc+PCM Control** demonstrated that bundles of cardiac muscle fibers in syncytium, nuclei are centrally located, no atypia is seen, no inflammatory cells are noted, pericardium and endocardium are histologically are not significant, chambers and valves are histologically unremarkable, valve appears dilated [Fig. no 1-(O)].
- 8. DOX+Talc+PCM Control** displayed that bundle of Cardiac muscle fibers in syncytium, nuclei are centrally located, no atypia is seen, pericardium show mature chondroid element, endocardium is histologically not significant, chambers and valves are histologically remarkable; valve appears dilated [Fig. no 1-(P)].

Food and water intake

Talc treated rats increased water intake after 10th weeks of Talc administration, and food intake is low in Talc treated groups as compare to normal control. Urination is frequent in Talc treated rats.

IV. DISCUSSION

The current American Heart Association heart failure classification schema designates the presence of diabetes mellitus as stage a heart failure, which raises the risk of developing stage B heart failure or asymptomatic left ventricular (LV) dysfunction [14]. Individuals with diabetes frequently have LV remodelling; both increased LV mass and dilatation have been reported [15]. Ventricular remodelling, first described in animal models of LV stress and injury, occurs progressively in untreated patients after large myocardial infarction and in those with dilated forms of cardiomyopathy. The gross pathologic changes of increased LV volume and perturbation in the normal elliptical LV chamber configuration is driven, on a histologic level, by myocyte hypertrophy and apoptosis and by increased interstitial collagen [16].

Doxorubicin is an anticancer drug that is helpful in treating several types of cancer, although its clinical use has been limited due to cardiomyopathy induced by dose-dependent cardiotoxicity. Once cardiomyopathy occurs, treatment options are few, and doxorubicin-induced heart failure is usually refractory to conventional therapy. Therefore, to evaluate Talc effect on doxorubicin-induced cardiomyopathy is the goal of this study [17], [18].

Histopathological study of albino wistar rats were deciphered the normal articulator in normal control and PCM control [Fig. no-1 (I, L)] which indicated that PCM has no effect on cardiac wall, chambers and valve also; while Doxorubicin treated rats showed toxic changes in valve region that supported the theory in which researcher suggested its limited use due to cardiomyopathy induced by dose-dependent cardiotoxicity [Fig. no-1 (J, M, N, P)]. With the Talc treatment, biopsy report proven that Talc administration was dilated the heart chamber and valve region in albino wistar rats (Fig. no-1 (K, M, O, P)). Histological assessment is deciphered that due to increased blood glucose level and elevated triglyceride and HDL level in blood [8] increased liver fat and increased islets triglyceride which diminished insulin response to ingested glucose; it may activated rennin angiotensin system and leads the ROS production in rats heart, resulted dilation of valve and chamber is occurred; this condition is known is diabetes cardiomyopathy, which further go ahead the myocardial infarction and then cardiac remodelling.

V. CONCLUSIONS

It is clear that talc may promote left ventricular dysfunction which is result of myocardial infarction by diabetes cardiomyopathy, and for a period it will converted in cardiac remodelling but further research is requisite for accepting the role of Talc as a supporter of cardiac remodelling.

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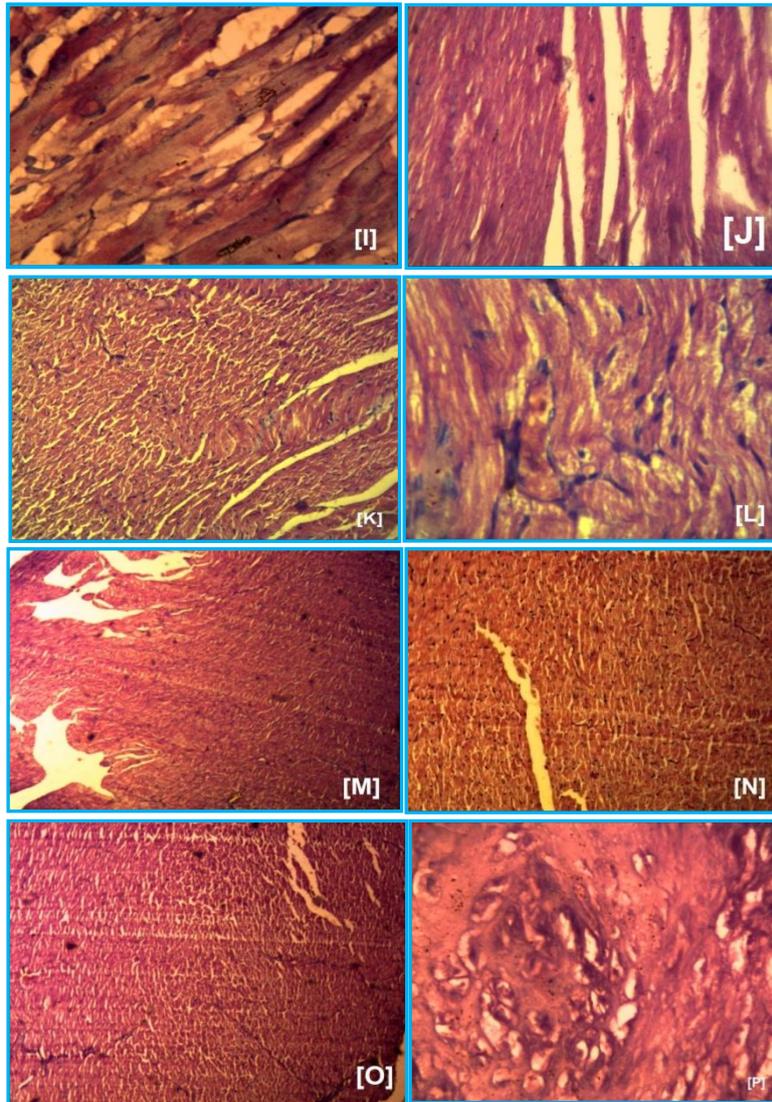


Fig no- 1: Representative Photo micrographs (45X) of histopathological changes demonstrate the effect of Doxorubicin and Talc on heart of wistar rat.