Comparative Histological Study on the Effect of Mesenchymal Stem Cell and Losartan on Cardiac Injury Induced by Doxorubicin in Male Albino Rats

Original Article

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ABSTRACT

Introduction: Cardiovascular disease is the leading cause of death worldwide. Mesenchymal stem cells (MSCs) could be a promising therapy for treating cardiomyopathy.

Aim of Work: To compare the therapeutic effects of MSCs and losartan (LOS) on doxorubicin (DOX) induced cardiomyopathy.

Material and Methods: Thirty-nine male albino rats were divided into group I as a control group, group II received DOX 5 mg/kg ip once, group III received DOX as in group II `after 3 weeks LOS was given 30 mg/kg daily by gastric tube for 3 weeks and group IV received DOX as in group and after 3 weeks rats injected by 1ml of Paul Karl Haron (PKH)26 labeled MSCs iv. Blood samples were collected for estimating the creatine kinase-myocardial band (CK-MB) value. Cardiac muscle sections from all groups were examined by fluorescence microscope. All sections were processed for histological study using Hx&E and Masson's trichrome stains in addition to immunohistochemical staining for Cx43 and CD44.Morphometric and statistical studies were done.

Results: Group II revealed fragmented muscle fibers and cytoplasmic vacuolation in addition to loss of transverse striation. These changes were confirmed by significant increase in the mean value of CK-MB and the mean area % of collagen fibers when compared to all other groups. In group III, regression of the previous changes was noticed and there was a significant decrease in the mean area % of collagen fibers when compared to group II. Group IV cardiomyocytes appeared healthy with normal arrangement and non-significant difference in mean area % of collagen fibers versus the control. Studying all groups immunohistochemically, revealed increase in positive reactions in LOS and SC groups when compared to DOX group with more increase in SC group than LOS group.

Conclusion: Losartan improves cardiac injury with little effect on cardiac regeneration. MSCs have promising potential in cardiac regeneration.

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Key Words: CD44; Cx 43; DOX; LOS; MSCs.

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INTRODUCTION

Cardiovascular diseases seem to be the major cause of disability and death in the whole world[1].

Doxorubicin (DOX) is a potent and widely used anthracycline chemotherapeutic agent used in treatment of several malignancies. The clinical use of this drug is limited because of its cardiotoxicity. The most frequent form of cardiotoxicity usually presented as a dilated cardiomyopathy[2].

Losartan (LOS) is an angiotensin II type 1 receptor (AT1) antagonist is commonly prescribed for the treatment of hypertension and its sequelae as pulmonary fibrosis and post infarction cardiomyopathy[3].

Mesenchymal stem cells (MSCs) are multipotent stem cells that can be isolated from bone marrow, umbilical cord and many other tissues and have the potential to differentiate into several cell types such as mesodermal lineage cells e.g. adipocyte, osteoblast and myogenic lineage[4].

Preclinical studies indicate that administered MSCs significantly ameliorated the cardiotoxic manifestations as shown by biochemical, functional and structural cardiac improvement and accelerate repair[5].

The current study was designed to evaluate and compare the possible therapeutic regenerative effect of both MSCs and LOS on experimentally induced cardiotoxicity using DOX in male albino rats.

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MATERIALS AND METHODS

I-Drugs

• Doxorubicin

Trade name Adriamycin, manufactured by Pharmacia Italia Co., in a form of vials containing (each 10 mg). It was dissolved in distilled water.

• Losartan

Trade name Cozaar, manufactured by Amriya Pharmaceutical Industries Alexandria-Egypt, in form of tablets (each 50 mg LOS potassium), that will crushed and dissolved in distilled water.

II- Stem cells (SCs)

Allogenic PKH26 labeled rat bone marrow SCs were purchased from stem cell research unit at the Biochemistry Department, Faculty of Medicine, Cairo University.

III-Animals

Thirty-nine adult male albino rats with an average weight of 200g were housed in Kasr Al Aini Animal house Faculty of Medicine, Cairo University and treated in accordance with guidelines approved by Animal use committee of Cairo University. All animals were kept under the same environmental conditions with free access to food and water.

IV- Experimental design

The rats were divided into four groups: Group I (Control group): 9 rats were subdivided into 3 subgroups, 3 rats for each experimental group.

Subgroup Ia: 3 rats, each received 0.5 ml distilled water by IP as a single dose.

Subgroup Ib: 3 rats, each received a single IP injection of 0.5 ml distilled water and after 3 weeks received 0.5 ml distilled water orally for 3 weeks.

Subgroup Ic: 3 rats, each received a single dose of 0.5 ml distilled water IP and after 3 weeks they were injected with 0.5ml of PBS IV in the tail vein for two successive days.

Group II (DOX group): 10 rats, given DOX in a dose of 5 mg/kg IP[6&7] as a single dose. Each rat received 0.5ml distilled water containing 1mg DOX.

Group III (LOS group): 10 rats, each received DOX as in group II and after 3 weeks LOS 30 mg/kg/day[8] was administered orally for 3 weeks[7] by the oral route through gastric tube[9]. Each rat received 0.5ml distilled water containing 6 mg LOS.

Group IV (SC group): 10 rats, each received DOX as in group II and after 3 weeks each rat received ml of PKH26 labeled MSC suspension in BPS (3x106 cells/ml) for two successive days[10] through the tail vein[11].

Preparation of the stem cells

A. Isolation and Propagation in culture[12&13]

B. Morphological identification of BM- derived MSCs[14].

C. Labeling of stem cells with PKH26 dye[15].

D. Detection of homing of injected cells in the cardiac muscle of rat

A-Biochemical study

Before scarification blood samples were drawn from tail veins of all rats and collected in heparinized capillary tubes. Samples were analyzed for creatine kinase -MB (CK –MB), a marker of cardiac muscle damage[16] at the Biochemistry Department, Faculty of Medicine, Cairo University.

B-Histological study

Scarification was done by 100 mg/kg ketamine-xylazine IP[17]. Ventral middle incision was made, and the heart was extracted and washed in saline. Cardiac muscle specimens were obtained from the ventricles and the apex of the heart. They were fixed in 10% formol saline for 48 hours. Paraffin sections were cut at 5 µm thickness and exposed to the following:

1. Haematoxylin and Eosin (Hx&E) stain[18]
2. Masson's trichrome stain[19&20]
3. Fluorescence detection (labeled MSC): fluorescent microscope was used to detect the injected SC labeled with a PKH26 dye[15].

C-Immunohistochemial Study

Connexin 43 (Cx43) Antibody

It is a major component of gap junctions showing clear and specific labeling of intercalated discs[21]. It was obtained from (Santa Cruz Biotechnology, Inc, Europe (C-20) sc 6560 Ab)

CD44 Antibody

It is the marker for MSCs[22&23]. It was obtained from (IHC world corporation, catalogue no IW-PA1021)

D-Morphometric study

Data were obtained using "Leica Qwin 500 C" image analyzer computer system Ltd. (Cambridge, England). For each group, five slides of five different specimens were examined. From each slide, ten non-overlapping fields were measured in a standard measuring frame. The following parameters were measured:

1. Mean area percent of collagen fibers Masson's trichrome stained sections x 100
2. Mean area percent of Cx43 positive reaction x 100

3. Mean number of CD44 positive immunostained cells x 400

E-Statistical Analysis

The measurements obtained were analyzed using SPSS software version 16 (SPSS, Chicago, IL). Comparison between different groups was made using analysis of variance (ANOVA) followed by post hoc Tukey test. The results were expressed as means ± standard deviation (SD). The differences were considered statistically significant when "*p value*" was < 0.05[24].

RESULTS

I-General Observation

No deaths were observed the rats during the experiment.

II-Biochemical Results

Marked affection of cardiac injury characterized by a significant increase of serum CK-MB in all experimental groups. Mean value of CK-MB in the group I was 123.14+0.68, while in group II was 298.16+6.57. In group III was 189.58+6.86 and group IV was 128.12 +4.82.

III-Histological results

1) H&E stain (Plate1)

Examination of sections of rat cardiac muscle of the group I showed elongated branching cardiac cells with cross striations exhibited rounded vesicular centrally located nuclei

Group II showed rarified widely separated muscle fibers. Many sections showed attenuated muscle fibers with wide endomysium. Focal areas of cytoplasm of different muscle fibers appeared either deeply acidophilic or vacuolated in others. Darkly stained pyknotic nuclei appeared peripheral in some fibers and lost in others. Thickened dilated congested blood vessels were seen, as well as areas of blood extravasation. There was localized large homogenous material with lost striation (infarction).

Group III showed many well-organized muscle fibers with central vesicular nuclei, while some fields showed few muscle fibers with peripheral dark nuclei and slightly rarified cytoplasm. There were many dilated congested blood vessels present between muscle fibers as well as many areas of blood extravasation.

Group IV showed well organized branched muscle fibers separated by fine endomysium and possessed vesicular centrally located nuclei. Some fibers possessed dark central nuclei surrounded by a halo. There was minimal cellular infiltration.

2) Masson'sTrichrome stain (plate 2)

Examination of sections of cardiac muscle from the group I showed fine collagen fibers between the muscle fibers.Group II demonstrated dense collagen fibers between the muscle fibers and around blood vessels. Group III revealed few dense collagen fibers between cardiac muscle fibers. While Group IV showed minimal collagen fiber between cardiac muscle fibers as well as around multiple dilated blood vessels.

2) Fluorescent Microscopic Results

Sections of control, DOX and LOS groups showed absence of PKH26 labeled stem cells while sections of cardiac muscle in SC group showed the presence of PKH26 labeled stem cells among muscle fibers (Plate 3).

IV-Immunohistochemical Results

Cx43 immunohistochemical stained sections (Plate 4)

Examination of sections of cardiac muscle from the group I showed multiple immunostained positive Cx43 at intercalated discs. Group II showed few immunostained positive Cx43 at intercalated discs in the disrupted muscle fiber, sections of cardiac muscle from group III showed some immunostained positive Cx43 at intercalated discs .While the group IV showed multiple immunostained positive Cx43 at intercalated discs between the cardiomyocytes.

CD44 immunohistochemical stained sections (Plate 5)

Examination of sections of cardiac muscle from the group I showed positive immunostaining reactions at the endothelial lining of blood vessels. Group II positive CD44 endothelial cells of blood vessels and among cardiac muscle were observed. Sections of cardiac muscle in group III contained multiple positive CD44 endothelial cells of blood vessels and between the muscle fibers. On the other hand, sections in the cardiac muscle of rats in group IV revealed numerous positive cells among the muscle fibers and in vessels in multiple fields.

V-Morphometric and Statistical Results

1) The mean values of serum CK-MB

Statistical analysis revealed that the mean values of serum CK-MB showed a significant increase ($P \leq 0.05$) in group II when compared to all other groups. It was a significant increase ($P \leq 0.05$) in group III when compared to both groups I and IV. However, in group IV CK-MB level was significantly decrease ($P \leq 0.05$) comparable to groups II and III with non-significant difference when compared to group I (Histogram 1).

2) Mean area % of collagen fibers (Table 1)

The mean area % of collagen in Masson stained sections of group II indicated a significant increase ($P \leq 0.05$) compared to all other groups. In the group III, there was significant decrease ($P \leq 0.05$) in mean area% of collagen fibers when compared to group II with non-significant difference versus the group IV. In group IV, there was significant decrease ($P \leq 0.05$) in mean area% of collagen fibers when compared to group II and non-significant difference when compared to the group I (Histogram 2)

3) Mean area % of Cx43 immunohistochemical stain (Table 1)

Regarding the mean area % of Cx43 immunoexpression showed a significant decrease (*P≤0.05*) in both groups II &III when compared the control group. However, group IV showed a significant increase (*P≤0.05*) in Cx43 immunostaining area % when compared to both groups II& III, but there was a non-significant difference versus the control group (Histogram 3)

4) Mean number of CD44 immunopositive cells (Table 1)

The mean number of CD44 positive cells in both groups III & IV was significantly increased (*P≤0.05*) when compared to group II. While group IV showed a significant increase (*P≤0.05*) in positive CD44 cells when compared to group III (Histogram 4)

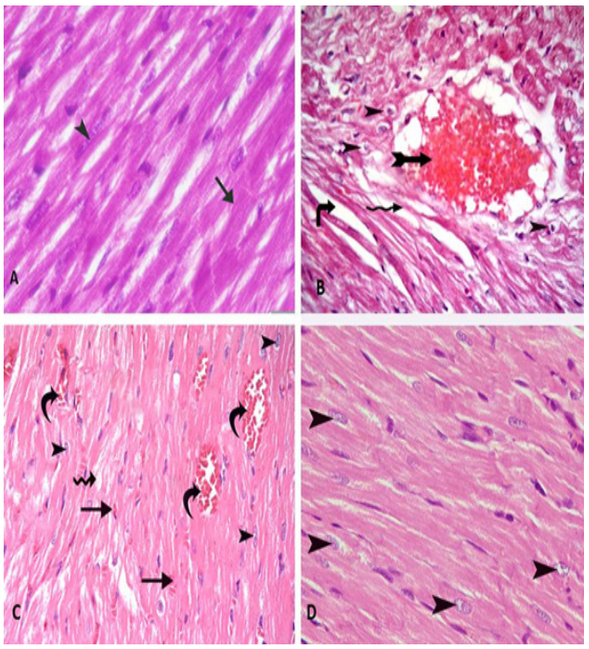


Plate 1: (A) Photomicrograph of the cardiac muscle from Group I) showing branched fibers (arrow) with cross striation and centrally located vesicular nuclei (arrow head). (B) Group II showing dilated congested blood vessel (bifid arrow), attenuated muscle fibers (spiral arrow) and some fibers show pyknotic nuclei surrounded by cytoplasmic halos (arrow heads). Note the presence of wide endomysium (right angled arrow). (C)Group III showing many well organized muscle fibers with central vesicular nuclei (arrow head), while some fibers show slightly rarified cytoplasm (spiral arrow). There are many dilated congested blood vessels present between muscle fibers (curved arrow), as well as many areas of blood extravasation (arrow) can also be seen.(D) Group IV showing apparently normal cardiac muscle fibers with central vesicular nuclei (arrow heads) (Hx&E,x 400).

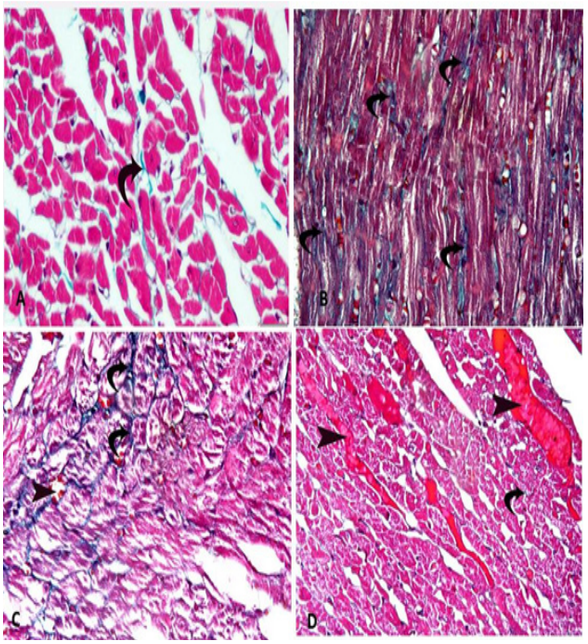


Plate 2: (A) Photomicrograph in the cardiac muscle of a rat in group I showing fine collagen fibers between the muscle fibers (curved arrow) B)Group II showing dense collagen fibers between the muscle fibers (curved arrows).(C) Group III showing few dense collagen fibers between cardiac muscle fibers (curved arrows), and around the dilated blood vessels (arrow head).(D) Group IV showing fine collagen fibers between the muscle fibers (curved arrow). There are also multiple dilated congested blood vessels (arrow heads) (Masson's trichrome, x 400)

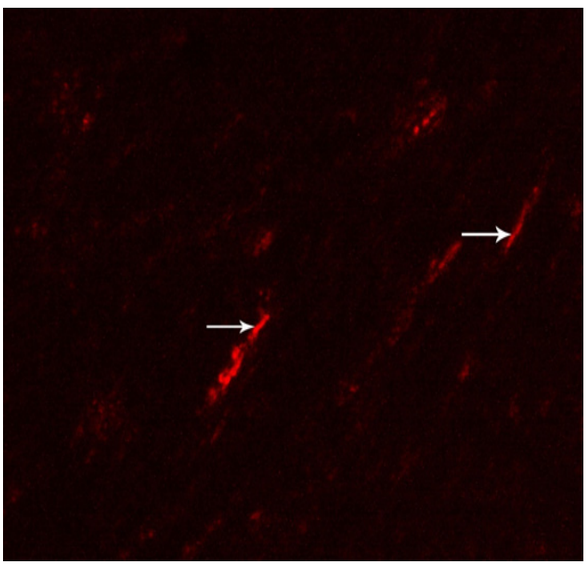


Plate 3: Photomicrograph of a section in the cardiac muscle of a rat from group IV showing few fluorescent labeled cells among the fibers (thin arrows) (PKH26, x 100).

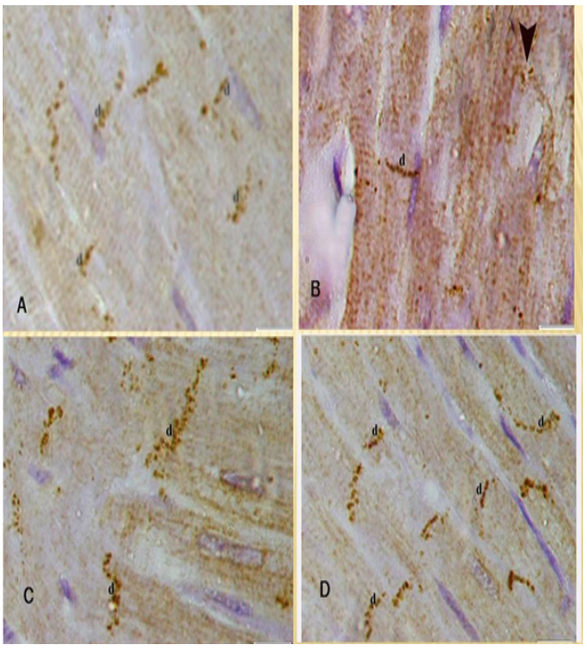


Plate 4: (A)Photomicrograph of a section in the cardiac muscle of a rat from group I showing multiple Cx43 +ve intercalated discs between the cardiac myocytes (d).(B) Group II showing few Cx43 +ve intercalated discs (d), while many cardiac muscle fibers show disrupted intercalated discs(arrow head).(C)Group III showing some Cx43 +ve intercalated discs (d) between the cardiac myocytes.(D) Group IV showing multiple Cx43 +ve intercalated discs (d) between the cardiac myocytes (Cx 43 immunostaining, x1000).

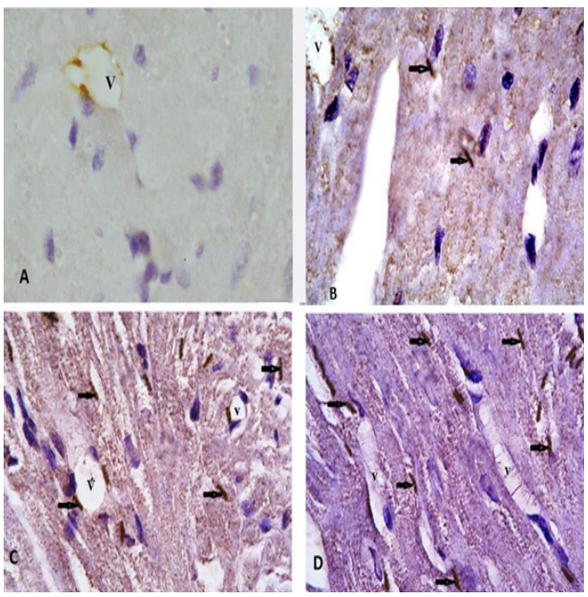
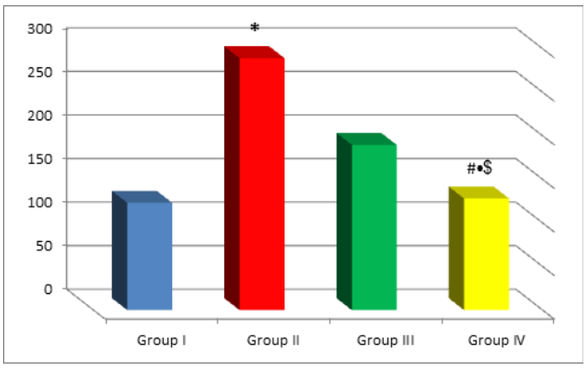
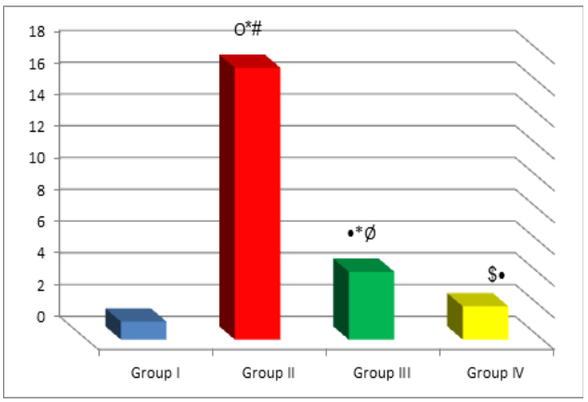


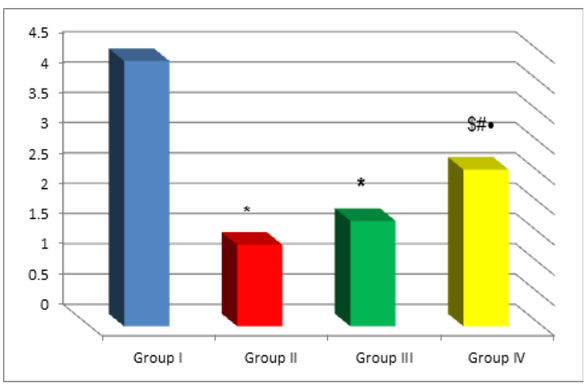
Plate 5: (A) Photomicrograph of a section in the cardiac muscle of a rat from group I showing few +ve cells at the endothelial cells lining of blood vessels (v) .(B) Group II showing few +ve cells (arrows) among the muscle fibers and in the endothelial cells lining the blood vessels (v).(C) Group III showing multiple + ve immunostaining cells (arrows) among the fibers and in the endothelial cells lining blood vessel (v). (D) Group IV showing numerous + ve immunostaining cells (arrows) among the fibers, and in endothelial cells lining blood vessel (v) (CD44 immunostaining, x1000).



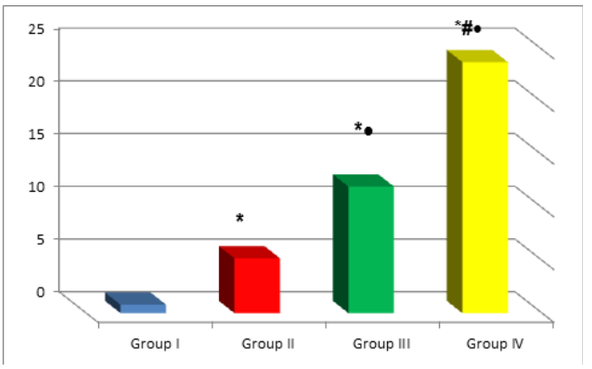
Histogram 1: Mean values of serum CK-MB in all studied groups
*Significant (*P≤0.05*) compared to group I
• Significant (*P≤0.05*) compared to group II
Significant (*P≤0.05*) compared to group III
O Significant (*P≤0.05*) compared to group IV
$ Non-significant difference when compared to a group I



Histogram 2: Mean area % of collagen fibers in all studied groups
*significant (*P≤0.05*) compared to group I
• Significant (*P≤0.05*) compared to group II
Significant (*P≤0.05*) compared to group III
O Significant (*P≤0.05*) compared to group IV
Ø Non-Significant (*P≤0.05*) compared to group IV
$ Non-significant difference when compared to the group I



Histogram 3: Mean area % of Cx43 immunostaining
*significant (*P≤0.05*) compared to group I
• Significant (*P≤0.05*) compared to group II
Significant (*P≤0.05*) compared to group III
$ Non-significant difference when compared to the group I



Histogram 4: Mean number of CD44 immunopostive cells
*significant (*P≤0.05*) compared to group I
• Significant (*P≤0.05*) compared to group II
Significant (*P≤0.05*) compared to group III

Table 1: Mean area % of collagen fibers, mean area% of Cx43 immunostainning and mean number of CD44 positive cells

Groups	Mean area % of collagen fibers	Mean area % Cx43 immunoreactivity	Mean number of CD44 immunostained cells
Control group	1.1340±0.46	4.380±1.762	0.8±0.44
Group II (DOX group):	17.1087±1.817$^{\#O*}$	1. 346±0.73*	5.2±1.3*
Group III (Losartan group):	4.266±2.41$^{*\emptyset}$	1.740±0.291*	12±1.58$^{*\bullet}$
Group IV (SC group):	2.094±1.6$^{\bullet\$}$	2.589±1.66$^{\bullet\#\$}$	23.8±3.1$^{*\bullet\#}$

* Significant (*P≤0.05*) compared to group I

• Significant (*P≤0.05*) compared to group II

Significant (*P≤0.05*) compared to group III

O Significant (*P≤0.05*) compared to group IV

$ Non-significant difference when compared to the group I

Ø Non-Significant (*P≤0.05*) compared to group IV

DISCUSSION

Cardiovascular disease is the leading cause of mortality and morbidity worldwide[25].

Many pharmacological agents may acutely lesion the myocardium either causing myocarditis or a chronic lesion like dilated idiopathic cardiomyopathy. Among these agents, we point out DOX[26].

The current study was designed to evaluate and compare the possible therapeutic and regenerative effect of both MSCs and LOS on experimentally induced cardiotoxicity using DOX in male albino rats. Biochemical, histological and immnunohistochemical techniques in addition to morphological measurements and statistical analysis of data were done.

In the present study, analysis of serum CK-MB revealed a significant increase in the0 mean values of DOX group when compared to all groups. This is in consistent with previous reports where DOX- induced cardiotoxicity characterized by an elevation of CK-MB[27]. There was a significant increase in group III when compared to I and IV group. This could be explained by the study of[28] that the ameliorating effect of SCs over that produced by LOS and also documented that LOS neither stimulated nor prevented cardiomyocyte regeneration.

There was significant decrease in group IV when compared to groups II and III and non-significant difference when compared to group I.[29] speculated that MSCs secret certain cytokines, thus promote myocardial glucose metabolism, stimulate cardiomyocytes regeneration and enhance cardiac function.

By LM examination, cardiomyopathic findings were in the form of fragmentation with loss transverse striations of muscle fibers. Many sections showed attenuated muscle fibers with wide endomysium. Some muscle fibers showed focal areas of deep cytoplasm staining indicating cytoplasmic protein denaturation[30] As well as, other fibers showed vacuolated cytoplasm that might be related myofibril loss or vacuolar degeneration caused by intracellular water and electrolytes redistribution[31]. It was associated with multiple pyknotic nuclei might be due to myocardial apoptosis, some of which were eccentric might be due to peripheral chromatin condensation. These changes were recorded by[32]. A number of studies have suggested that apoptosis is associated with oxidative stress[33].

The present study revealed congested blood vessels and wide areas of extravasated RBCs among the muscle fibers in group II. Congestion of the vessel might be due to weakness of the vessel's walls as a result of inflammation with consequent damage of the vessel wall and interstitial hemorrhage[34]. This is in agreement with[35] who reported focal hyalinization of myocardial bundles with focal hemorrhages in-between the cardiac fibers.

Homogenous material was noticed among the fibers, diffuse interstitial oedema and pale myocytes with fading nuceli indicating the coagulative necrosis (infarction) this finding was in agreement with[36]. This could be explained by the immediate effect of DOX on the morphology of the regional vasculature manifested by disintegration of the vessel's wall and activation of coagulation and platelet aggregation pathways and microthrombi formation, leading to acute reduction in vascular blood flow and could be followed by infarction as was recorded [37].

In the LOS group showed many well-organized muscle fibers with central vesicular nuclei this explained by[38] that LOS could inhibit the oxidative stress via upregulation of NO synthase enzyme.

As regards dilated congested blood vessels[39], reported that LOS induced a stimulation of angiogenesis via inhibitory function of Ang II on the AT1 receptor. But[40], stated that the exact role of LOS in angiogenesis has been contradictory.

In the current work, homing of MSCs was confirmed by the fluorescent microscope which exhibited some PKH26 labeled cells among the fibers. And this is in agreement with[41].

Section from SC group revealed apparently normal arrangement that well-organized cardiac muscle. Transplanted MSCs have ability to differentiate into cardiomyocytes and vasculature. This could be explained by[42]. Mesenchymal stem cells injection promotes the cardiac progenitor cells (CPCs) and helps in the improvement of myocardium as CPCs could develop into the three main types of adult heart cells; cardiomyocytes, endothelial cells and smooth muscle cells that are required for heart regeneration[43].

It was reported that MSCs secrete a wide array of paracrine factors which led to immunosuppressive, anti-fibrotic, pro-angiogenic, and anti-oxative responses[44].

Examination of Masson's trichrome stained sections of cardiac muscle from group II demonstrated dense collagen fibers between the muscle fibers and around blood vessels. This is in accordance with[45] who explained that fibrosis was due to excessive cardiac fibroblast accumulation and extracellular matrix deposition .Moreover, DOX increase expression of tumor necrosis factor-beta (TGF-β)[46] reported that the TGF-β is a strong stimulator of pathological collagen production as well as a cytokine that has been associated to fibro-inflammatory signaling.

Group III revealed few dense collagen fibers between cardiac muscle fibers[47] mentioned that LOS is an anti-fibrotic agent; it directly antagonizes TGF-β via AT1. While group IV showed minimal collagen fiber between cardiac muscle fibers this could be via paracrine signaling from MSCs mediated by anti-fibrotic factors. In addition, the cultured MSCs express hepatocyte growth factor that has been shown to exert antifibrotic, antiapoptotic and angiogenic effects that may participate in amelioration of cardiac fibrosis[48].

The expression of Cx43 in the heart muscle is critical for synchronized contraction between cardiomyocytes[49] and it is an important molecule in the regenerative healing of heart[50].

In group II showed few Cx43 positive reaction at intercalated discs in the disrupted muscle fiber, this was confirmed by the morphometric results where there was a significant decrease mean area % of immunoreactivity, which reflects gradual decrease in percentage of healthy cardiac muscle fibers in group II when compared to the control group. These results are in agreement with[51] who explained that DOX treatment induced slower proliferation rates, decreased Cx43 production and hindered SC capacity to respond to cardiomyogenic differentiation stimuli.

Group III showed some Cx43 positive reaction between the myocyte. The mean area % of Cx43 immunoractivity in this group showed a significant decrease when compared to the control group; this might be due to decreased production of Cx43. It was reported that upregulation of Cx43 protein in response to Ang II and also stated that LOS reduced the over-expressions of Cx43 in the myocardium[52].

Group IV showed multiple Cx43 positive reaction at intercalated discs between the cardiomyocyte. There was a significant increase in the mean area % of Cx43 immunoexpression, which reflect increase in percentage of healthy cardiac muscle fibers when compared to both groups II and III, and a non-significant difference versus the control group. MSCs have been documented to modulate conduction of cardiomyocytes by increase in the conduction velocity of cardiomyocyte by paracrine signaling, via upregulation of Cx43 and nerve growth factor[53].

In group II sections of cardiac muscle stained with CD44 immunohistochemical stain showed few positive spindle cells confined to the endothelial cells in the blood vessels walls and among the muscle fibers. The mean number of CD44 positive cells was a significant decrease in group II when compared to all groups. This could be explained by[54] who demonstrated that hypoxia improves MSC self-renewal and therapeutics resulted in SCs migration, increased cell adhesion and activation of target genes coding for paracrine factors, which accelerate tissue regeneration.

As regards the group III, sections of cardiac muscle revealed multiple CD44 positive cells among the fibers and the endothelial cells of blood vessels. The mean number of CD44 positive cells in group III was significantly increased when compared to the group II, but significantly decreased when compared to IV group as angiotensin receptor blockers improved the efficiency of cardiomyogenic transdifferentiation and improved the cardiac function via neovascularization that restored the contractility[55].

Sections from group IV displayed multiple CD 44 positive cells among the muscle fibers and in endothelial cells of blood vessels. The mean number of CD44 positive cells in group IV were significantly increased when compared to both group II&III. This could be explained by[56] who that BM-MSCs could express important SCs surface markers such as CD44, CD90 and CD105. In addition[57], stated that CD44 is a cell adhesion receptor and is involved in cell–cell interactions, cell adhesion and migration. The improved CD34 and CD44 expression reflects a recovery in bone marrow function[58].

CONFLICT OF INTEREST

There are no conflicts of interest.

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الملخص العربى

دراسة هستولوجية مقارنة بين تاثير الخلايا الجذعية المزنشيمية وعقار اللوسارتان علي اصابة عضلة القلب المحدث بعقار الدكسوروبسين في ذكور الجرذان البيض

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الخلفية: تعتبر أمراض القلب والاوعية الدموية السبب الرئيسي للوفاة علي مستوي العالم. ويمكن للخلايا الجذعية المزنشمية ان تكون علاجا واعدا لعلاج اعتلال عضلة القلب.

الهدف من العمل: لمقارنة التأثير العلاجي للخلايا الجذعية المزنشيمية واللوسارتان على اعتلال عضلة القلب المستحدث بعقار الدوكسوروبيسين.

مواد وطرق البحث: تم تقسيم تسعة وثلاثون من ذكور الجرذان البيض قسمت الي

المجموعة الأولي كمجموعة ضابطة، تلقت المجموعة الثانية الدوكسوروبيسين ٥ ملج/كج داخل تجويف الغشاء البريتونى لمرة واحدة. وتلقت المجموعة الثالثة الدوكسوروبيسين كما في المجموعة الثانية وبعد ٣ أسابيع تم اعطائهم ٣٠ ملج / كج / يوميا اللوسارتان عن طريق انبوبة المعدة لمدة ثلاثة أسابيع. وكذلك تلقت جرذان المجموعة المجموعة الرابعة الدوكسوربيسين كما في المجموعة الثانية وبعد ثلاثة اسابيع تم حقنهم بالخلايا الجذعية المزنشمية المميزة بـ PKH26 عن طريق الوريد وتم تجميع عينات الدم لقياس قيمة CK-MB ثم فحص الشرائح عضلة القلب في كل المجموعات بواسطة المجهر الفلورنسي MB وتمت معالجة كل شرائح للدراسة الهستولوجية باستخدام صبغة الهيماتوكسيلين والايوسين وماسون ثلاثية الألوان بالاضافة الي الصبغة هستوكيميائية مناعية CD43 و Cx44 كما تم اجراء دراسة مورفومترية واحصائية.

النتائج: اظهرت المجموعة الثانية الالياف العضلية مجزأة كما ظهرت فجوات في السيتوبلازم بالاضافة الي فقدان التخطيطية العرضية ولقد تاكدت هذة التغيرات بزيادة ذو دلالة احصائية في قيم CK-MB ومتوسط النسبة المئوية لمساحة الياف الكولاجين مقارنة بكل المجموعات الاخري ولقد لوحظ في المجموعة الثالثة تراجع ملحوظ في التغيرات السابقة مع انخفاض ذو دلالة احصائية في متوسط النسبة المئوية لمساحة الياف الكولاجين مقارنة بالمجموعة الثانية. ظهرت في المجموعة الرابعة الالياف العضلية بشكل صحي و ترتيب طبيعي وكانت غير ذات دلالة احصائية في متوسط النسبة المئوية لالياف الكولاجين مقارنة بالمجموعة الاولي ولقد عكست الدراسة الهستوكيميائية المناعية لكل المجموعات ان مجموعة اللوسارتان ومجموعة الخلايا الجذعية تفاعل ايجابي مقارنة بمجموعة الدكسوروبسين كما اظهرت الخلايا الجذعية المزيد من الزيادة في التفاعل الايجابي عنه في مجموعة اللوسارتان

الاستنتاج: ولقد وجد ان عقار اللوسارتان يحسن من اصابة عضلة القلب وان كان له تاثير ضئيل علي تجدد خلاياه بينما وجد ان للخلايا الجذعية امكانية واعدة في تجدد عضلة القلب.