

The Protective Effect of Bone Marrow-Derived Mesenchymal Cells on Nephrotoxicity Induced by Carbon Tetrachloride in Adult Male Albino Rats: Histological and Ultrastructural Study

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Abstract: Background: Exposure to carbon tetrachloride (CCl₄) initiates nephrotoxicity in rats. The purpose of the present work was to study the role of bone marrow-derived mesenchymal cells (BM-MSCs) on CCl₄-induced nephrotoxicity in adult male albino rat judged by light and electron microscope using histological, immunohistochemistry and ultrastructure parameters. **Material & methods:** Thirty adult male albino rats were classified randomly into four experimental groups. Group I: Control group which was equally divided into two subgroups; Ia (that was fed on the standard diet) and Ib (that injected intraperitoneally with 0.1 ml of olive oil twice weekly for 4 weeks). Group II: treated with 1.0ml /kg b.w of 10% CCl₄ dissolved in olive oil, IP, twice a week for 4 one month. Group III: was injected intraperitoneally with the same dose of CCl₄ twice a week for 4 weeks and then were given a single dose intraperitoneally of 3×10⁶ BM-MSCs. Group IV: served as recovery group, were treated IP with 5.0ml /kg b.w of 10% CCl₄ twice a week for 4 weeks and left for a further 4 weeks. At the end of experimental period, all of the rats were scarified and both kidneys were prepared for histological, immunohistochemical and ultrastructural study. **Results:** In CCl₄treated animals, there were several pathological changes included: dilatation of tubular lumens with intraluminal cell debris and vacuolation of tubular epitheliumas compared with the control rats. Mallory's trichrome staining showed increased amount of collagen fibers in the glomerulus and interstitium in CCl₄ treated animals. While the rest of groups showed minimal collagen fibers around glomerulus and renal tubules. Immunohistochemical staining using α-Smooth muscle actin (α-SMA) showed negative reaction in all groups except CCl₄ treated animals and recovery group. EM evaluation of CCl₄ induced changed revealed significant condensation of chromatin in the nucleus and shortened brush border of proximal convoluted tubules, swollen mitochondria and loss of apical plasma membrane micro projection of distal convoluted tubules, also effacement of secondary foot processes on basement membrane of glomerular capillary with loss of slit diaphragms. In contrast, these deleterious histopathological alterations resulting from CCl₄nephrotoxin were absent after BM-MSCs treatment in CCl₄ + BM-MSCs group of rats. **Conclusion:** Our results demonstrated the protective potential of BM-MSCs on CCl₄-induced nephrotoxicity in adult male albino rat with better recovery findings in BM-MSCs received group than in those rats that were left to recover without treatment.

[Eman E. Elwakeel, Amira Z. Mohamed. **The Protective Effect of Bone Marrow-Derived Mesenchymal Cells on Nephrotoxicity Induced by Carbon Tetrachloride in Adult Male Albino Rats: Histological and Ultrastructural Study.** *J Am Sci* 2018;14(6):13-24]. ISSN 1545-1003 (print); ISSN 2375-7264 (online). <http://www.jofamericanscience.org>. 2. doi:[10.7537/marsjas140618.02](https://doi.org/10.7537/marsjas140618.02).

Keywords: Carbon tetrachloride; bone-marrow; renal dysfunction; nephrotoxicity; stem cell.

1. Introduction

Chronic kidney disease (CKD) is thought to be a global health burden with a high monetary cost to health systems. All phases of CKD are related with increased dangers of cardiovascular morbidity, premature mortality, and diminished nature of life [1].

One of the most commonly used for induction of intoxication in experimental animals is carbon tetrachloride (CCl₄) as a model that leading to oxidative stress in most pathophysiological conditions. The poisoning by CCl₄persuaded toxic lesions in the kidney and liver [2]. Hepatic injury may be dominated by acute renal tubular necrosis, which may cause renal oliguria of most of animal species. Many different researches noted that CCl₄ intoxication caused free

oxygen radical formation in most vital organ tissues like liver, heart, brain, kidney, blood and lung [3].

There is no specific treatment for chronic kidney disease, except for the adjuvant therapy [4]. Although these treatments can improve renal function and promote renal repair, they require administration in high doses [5]. Nowadays, renal additional treatment represented in either renal dialysis or transplantation of kidney, which is greatly restricted due to decrease in the number of suitable donors [6]. The use of stem cell therapy has been proposed as a possible way to advance the course and overcome kidney damage. MSCs (Mesenchymal stem cells) show special promise in renal repair, as the nephrons are of mesenchymal origin [7].

Stem cells are known as biological cells that present in every multicellular organism. Bone marrow is considered as one of the main sources for obtaining stem cells and can harvest at least two types of stem cells, hematopoietic stem cells and non-hematopoietic stem cells or mesenchymal stem cells (MSCs) [8].

Mesenchymal stem cells (MSCs) are defined as multipotent adult stem cells which can be harvested from all organs tissues, in addition it is known as a specific aggregations of mural cells/pericytes which found on the abluminal side of blood vessels. Initially recognized in the stroma of bone marrow (BM), and also make a support for the development of hematopoietic stem cells (HSCs), These cells can be distinct into dissimilar mesodermal lineages. Researchers reported that MSCs can routinely be harvested from the BM and multiply and developed in vitro culture system and can exhibit interesting immunomodulatory properties, thus developing a new interesting application as a therapy for different body disorders [9].

MSCs can differentiate, give rise to many kinds of cell and therefore, can be experimentally potentially used as a cell and gene therapy in the treatment of some diseases [10].

The present work aimed to investigate and analyze the defensive impact of bone marrow- derived mesenchymal cells in kidney of carbon tetrachloride intoxicated adult male albino rats.

2. Materials and Methods

Materials

1. Carbon tetrachloride

CCl₄ of 100% concentration (Sigma, St Louis, Missouri, USA) and pure olive oil (Saporito Foods Inc., Markham, Ontario, Canada) were purchased from Algomhoria Company. CCl₄ was diluted in equal volume of pure olive oil (1:1) and it was diluted at the time of use. It was given to rats by intraperitoneal injection twice weekly at a dose of 0.1 ml/kg for 4 weeks [11].

2. Bone marrow-derived mesenchymal stem cells

Rat bone marrow-derived MSCs were obtained from the Biochemistry Department, Kasr Al-Ainy Medical School, in the form of vials. Each vial contained 3×10^6 BM-MSCs suspended in 0.5 ml PBS. The vials were maintained in an ice box until injection within 5 h [12].

Animals and experimental design

The study was performed on 30 adult male albino rats (*Rattus norvegicus*) aged eight weeks and weighted about (120g-150g). Rats were purchased from the laboratories of ministry of agriculture, and kept in individual special plastic rodent cages in the laboratory under constant condition of temperature of

25 °C. Animals were fed on a standard rodent diet. Cages were maintained daily. The use of animals in this study conformed to the guidelines and bioethics of the animal ethical committee in Benha University, Faculty of medicine. The rats were divided into four groups:

Group I (Control group): This group was further divided into two subgroups of 5 rats each.

Subgroup Ia: Animals were fed on the standard diet and were served as control group.

Subgroup Ib: This group was given 0.1 ml of olive oil by intraperitoneal injection twice weekly for 4 weeks.

The rats in both subgroups were then sacrificed after 4 weeks.

1. Group II (CCl₄ treated group): 7 Rats were injected intraperitoneally with 1.0ml /kg b.w of 10% CCl₄ dissolved in oliveoil twice a week for 4 weeks and then sacrificed [11].

2. Group III (CCl₄ and BM-MSCs treated group): 7 Rats were injected intraperitoneally with 1.0ml /kg b.w of 10% CCl₄ dissolved in oliveoil twice a week for 4 weeks and then the rats in this group were given a single intraperitoneal injection of 3×10^6 BM-MSCs suspended in 0.5 ml PBS. They were then sacrificed after a further 4 weeks (i.e. 8 weeks from the beginning of the experiment).

Group IV (Recovery group): 6 Rats were injected intraperitoneally with 5.0ml /kg b.w of 10% CCl₄ dissolved in oliveoil twice a week for 4 weeks and then left as a recovery group for a further 4 weeks and then sacrificed.

Histological and immunohistochemical examinations

At the end of experiment according to timing mentioned in each group, the rats were sacrificed by decapitation. Their kidneys were dissected and fixed in 10% neutral formalin. Fixed specimens were embedded in paraffin wax and sections of 4-5 micrometer thickness were cut. Slides were stained with haematoxylin and eosin for histological examination, with modified Mallory's trichrome and modified avidin-biotin immunoperoxidase for α smooth muscle actin (α -SMA). Mallory's trichrome stain is a stain utilized in histology to aid in revealing different macromolecules that make up the cell. It utilizes the three stains: aniline blue, acid fuchsin, and orange G. As a result this staining technique can reveal collagen, red blood cells, and ordinary cytoplasm. It is useful, therefore, in examining the collagen of connective tissue. Positive immunoreactivity for α -SMA showed up as dark brown cytoplasmic staining of varying degrees. Negative control was gotten by omitting the step involving the primary antibody. Microscopic

examination of the stained sections was carried out by Olympus Light Microscope [13].

Ultrastructure examinations

For Transmission electron microscope (TEM) examination, Small renal cortex specimens (1 mm³) were fixed with 2.5% glutaraldehyde in 0.1 M cacodylate buffer, at that point post-fixed in 1% osmium tetroxide, got dried out in an evaluated arrangement of ethanol, then embedded in epon araldite [14]. One micrometer semithin sections were prepared by LEICA Ultra microtome and were mounted on glass slide then stained with toluidine blue. Ultra-thin sections were cut; at that point they stained with 8% uranyl acetic acid dissolved in 70 % ethanol lastly stained with lead citrate. The stained sections were analyzed and photographed with a JEM 100SX (JEOL, Japan) transmission electron microscope at the electron microscopic unit, Faculty of medicine, Tanta University, Egypt.

3. Results

In the current study, the results pointed to the possible protective effect of BM-MSCs against CCL₄-induced hepatotoxicity in experimental animals through retrogressive CCL₄ toxicity in the kidney cells as seen in the sections under light and electron microscope different methods, histological, immunohistochemistry and ultrastructure parameters.

The examination of kidney tissues in the present study under microscope revealed that the renal cortex contains multiple renal corpuscles and renal tubules which comprises proximal, distal convoluted tubules, and collecting tubules (Fig. 1A & 1B).

In the current work, the group administered CCL₄, showed that the renal cortex of the CCL₄ treated group showed congestion of blood capillaries, many degenerated proximal and distal convoluted tubules (PCT) with dilated tubular lumens, vacuolated tubular epithelium and flattened tubular epithelium (Fig. 1C). The renal glomerulus showed hypercellularity with dilated luminal spaces and infiltration with inflammatory cells (Fig. 1D).

BM-MSCs-treated rats showed enhancement of glomerular structure which surrounded by Bowman's capsule, filtration space. The proximal convoluted tubules were most probably normal with preserved brush border. Most renal tubules were seen almost similar to the control group (Fig. 1E). While the recovery group showed obliterating the Bowman's space. Congestion of peritubular

In the present study, Mallory's trichrome stained sections revealed the normal distribution of collagen fibers around glomerulus and renal tubules within the renal cortex in control rats (Fig. 2A & 2B). In contrast to the CCL₄ treated rats, showed increased amount of collagen fibers in the glomerulus and interstitium

within the renal cortex (Fig. 2C). BM-MSCs-treated rats showed the same finding compared to rats of control group (Fig. 2D). Moderately increased amount of collagen fibers around the glomerulus and interstitium were observed in the recovery group (Fig. 2E).

The current study indicated that the maximum effect was present in epithelial tubular cells and in the renal glomeruli in CCL₄-treated rats, which may be due to strong positive reaction of α -SMA (Fig. 3B), followed by the withdrawal group which showed negative reaction in glomerulus and positive reaction in renal tubules (Fig. 3D). While the control and BM-MSCs-treated groups revealed a negative reaction in glomerulus, renal tubules or interstitium, positive reaction is only seen around blood capillaries (Fig. 3A & 3C).

In the current work, the kidney of control group showed by EM examination that the PCTs had a normal appearance, where it PCTs were lined with cuboidal cells with even microvilli, abundant elongated mitochondria bounded in the regular basal infolding (Fig. 4A). However in rats treated with CCL₄ revealed apical shortened brush border and dark nuclei (N) with condensed chromatin. Many lysosomes and endosomes were too found in their cytoplasm (Figs. 4B & 4D). In contrast, examination of PCT in the group that treated with stem cells (MSCs) showed an indication for histological rebuilding of typical kidney structure with regular microvilli compared with the control group (Fig. 4C).

In the control group, examination of distal convoluted tubules (DCTs) exhibited little cuboidal cells with regular basal infoldings, euchromatic nuclei, and elongated mitochondria amongst can be observed (Fig. 5A). In contrast, the DCTs of CCL₄ treated rats showed unsettled basal infoldings carrying some swollen disrupted mitochondria within them, dilated infolding of the basolateral plasma membrane. Also shrunken lumen with loss of apical plasma membrane microprojection can be observed (Fig. 5B). The same findings were observed in the DCTs of recovery group; however, a few area of cytoplasm were rarefied (Fig. 5D). The DCTs of BM-MSCs-treated rats had preserved tubular cells ultrastructure in this group compared to control group (Fig. 5C).

The renal corpuscle of control rats revealed normal ultrastructure (Figs. 6A & 6B). The podocyte of the internal layer of Bowman's capsule had central euchromatic nucleus surrounded by cytoplasm. A thin capsular space was seen between the podocyte and the basal lamina of the capillary tuft. On the other hand the renal corpuscle in CCL₄-treated rats revealed congestion of the glomerular capillary with fusion of the secondary foot processes with loss of slit diaphragms and widening of the capsular space. Also

the renal corpuscle showed numerous mesangial cells in-between the capillary loops (Fig. 6C & 6D).

The same histological finding were seen in the recovery group (Fig. 7C & 7D)

The renal corpuscle of BM-MSCs-treated rats showed more or less normal podocyte with

euchromatic nucleus. This podocyte had primary thick foot process and numerous secondary foot processes on basement membrane of glomerular capillary compared to control group (Fig. 7A & 7B).

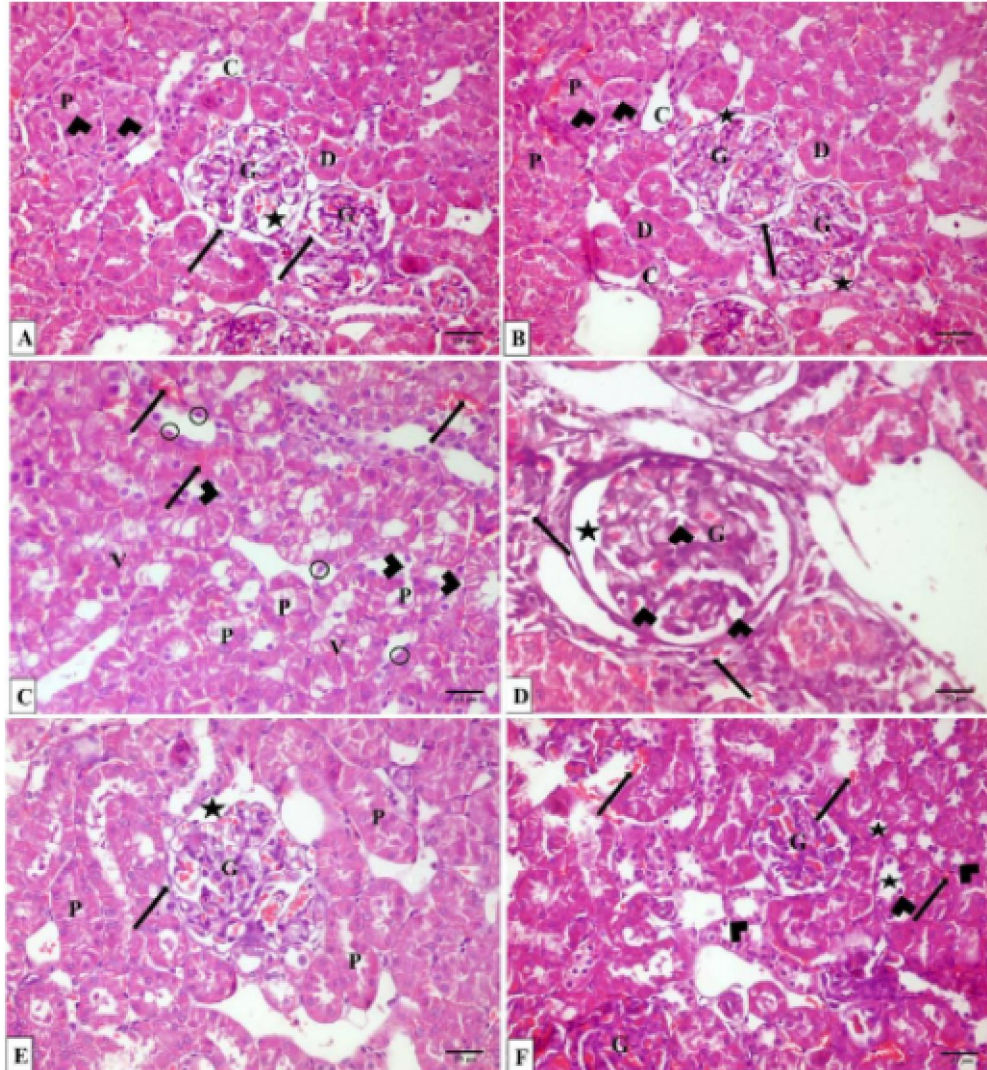


Figure 1. Effect of bone marrow- derived mesenchymal cells on histopathological damage Induced by CCl_4 on the kidneys of rats. Kidney sections were stained using the haematoxylin–eosin method. **A & B.** Control rats (subgroup Ia & Ib) without any signs of kidney damage. normal glomerular structure (G), surrounded by Bawman's capsule (arrow), filtration space (astiric), proximal convoluted tubules (P) with intact brush border (arrow head), distal convoluted tubules (D), and collecting ducts (C) (x 400). **C.** After 4 weeks following CCl_4 treatment, kidney sections presented congested peritubular blood capillaries (arrow), dilated tubular lumens with intraluminal cell debris (P), vacuolated tubular epithelium (V), flattened tubular epithelium (Closed circle), separated nuclei and denuded spaces (arrow head) (x 400). **D.** Carbon tetrachloride group showing hypercellularity of renal glomerulus (G) with dilated luminal spaces (arrow head) and widened filtration space (astiric), inflammatory cells infiltration (arrow) (x 1000). **E.** Bone marrow-derived mesenchymal stem cells treatment: kidney section with normal glomerular structure (G), surrounded by Bowman's capsule (arrow), filtration space (astiric), proximal convoluted tubules (P) with preserved brush border (x 400). **F.** the recovery group: the kidney sections showed obliteration of glomerular space (G), congested peritubular blood capillaries (arrow), dilated tubular lumens (astiric), separated nuclei and denuded spaces (arrow head). (x 400)

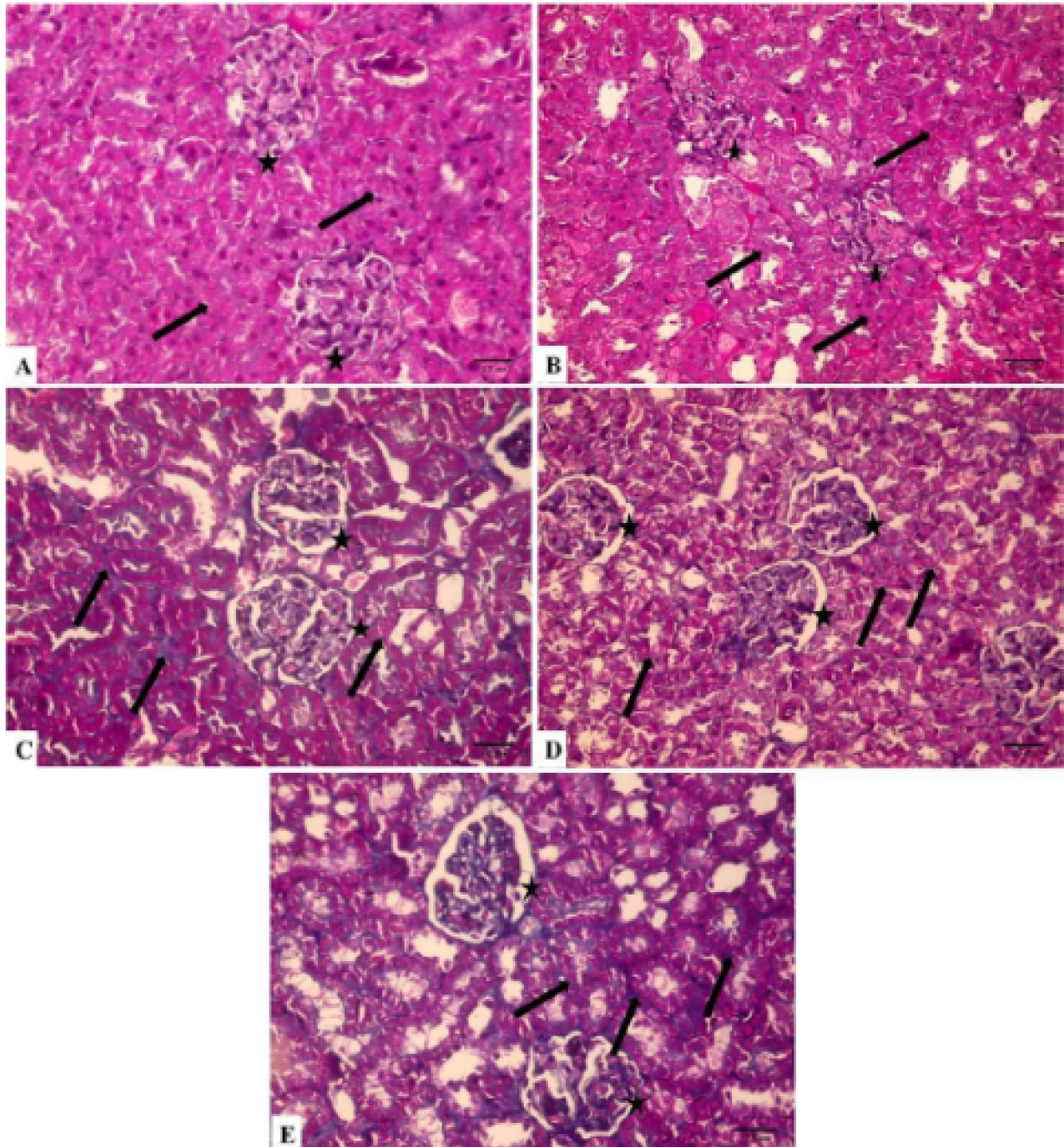


Figure 2. Mallory's trichrome staining of rat kidney sections showing: **A & B.** minimal collagen fibers around glomerulus (astric) and renal tubules (arrow) in normalcontrol group. **C.** increased amount of collagen fibers in the glomerulus (astric) and interstitium (arrow) in group II. **D.** minimal collagen fibers around glomerulus (astric) and renal tubules (arrow) in group III. **E.** moderately increased amount of collagen fibers in the glomerulus (astric) and interstitium (arrow) in group IV. (x 400).

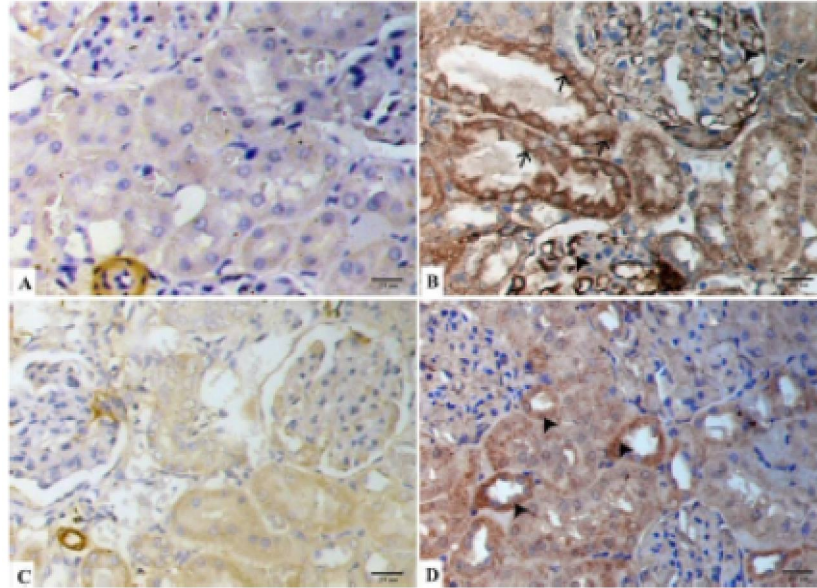


Figure 3. Immunohistochemical staining of rat kidney using α -Smooth muscle actin (α -SMA) showing: **A.** Negative reaction in glomerulus, renal tubules or interstitium, positive reaction was only seen around blood capillaries in group I. **B.** Strong positive reaction in glomerulus (arrow head), renal tubules (arrow) in group II. **C.** Negative reaction in glomerulus, renal tubules or interstitium, positive reaction was only seen around blood capillaries in group III. **D.** Negative reaction in glomerulus, positive reaction in renal tubules (arrow) in group IV. (x 400).

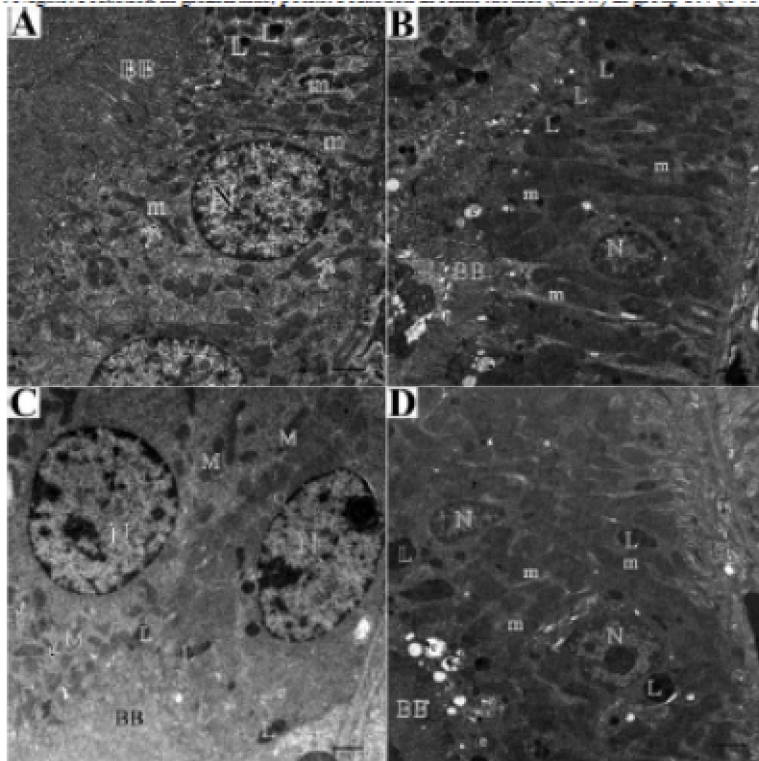


Figure 4. Transmission electron micrograph of proximal convoluted tubules showing: **A.** Normal aspect of the cuboidal epithelial cells lining with euchromatic nuclei (N), apical long brush border (BB), elongated vertical mitochondria (M) rough endoplasmic reticulum (arrow) and apical lysosomes (L) in group I. **B.** Dark nuclei (N) with condensed chromatin, apical shortened brush border (BB), elongated vertical mitochondria (M) and increased lysosomes (L) and endosomes (e) in group II. **C.** Preserved tubular cells ultrastructure in group III compared to control group. **D.** Dark nuclei (N) with condensed chromatin, apical shortened brush border (BB), elongated vertical mitochondria (M), increased lysosomes (L) and endosomes (e) in group IV. (x 8000).

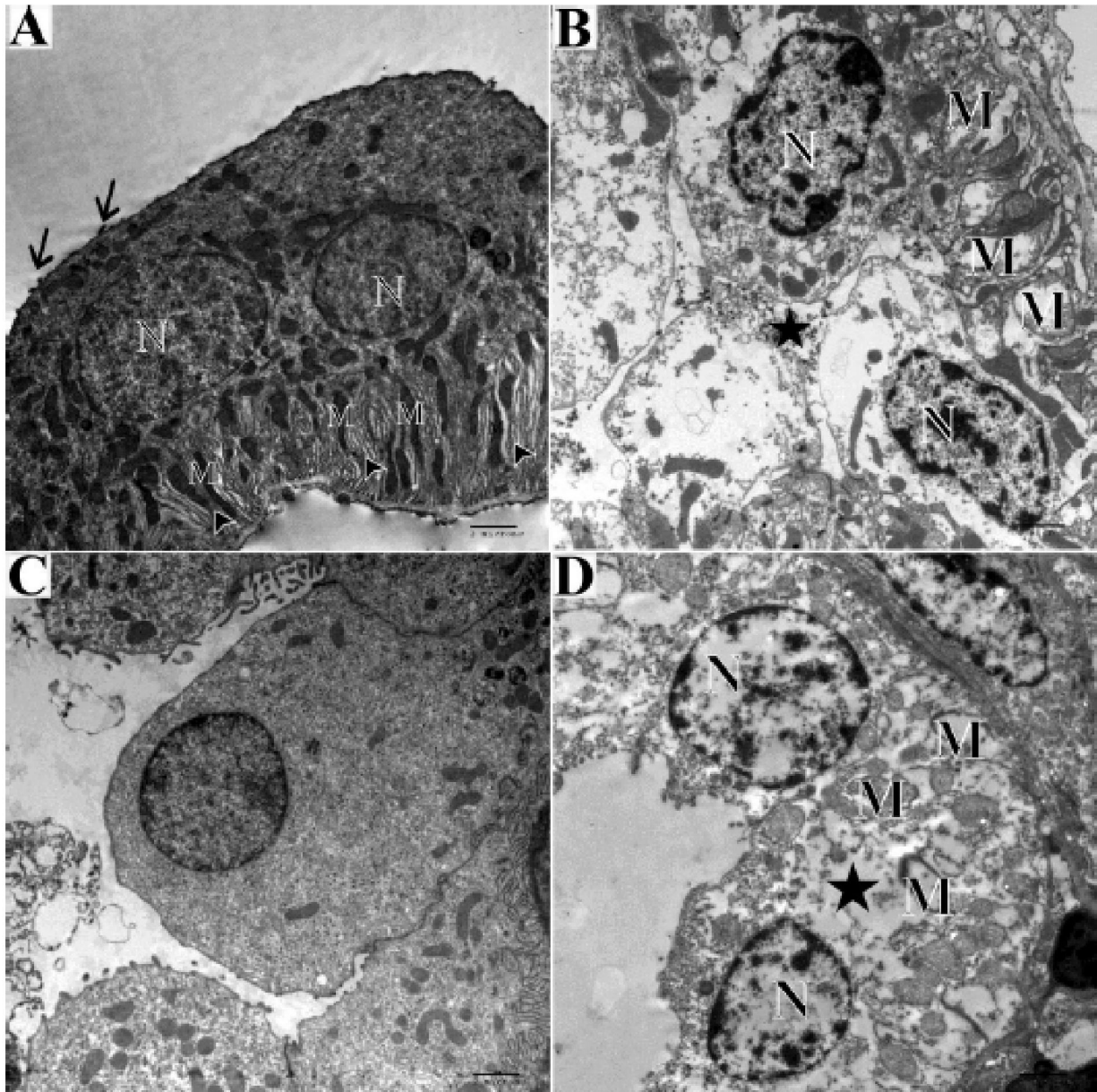


Figure 5. Transmission electron micrograph of distal convoluted tubules showing: **A.** Cells with euchromatic nuclei (N), long mitochondria (M) oriented vertical among the numerous deep enfoldings of the basolateral plasma membrane (arrowhead), short apical plasma membrane microprojection (arrow) in group I. **B.** Swollen disrupted mitochondria (M), dilated enfoldings of the basolateral plasma membrane (arrowhead), shrunken lumen with loss of apical plasma membrane microprojection (asteric) of CCl_4 treated group. **C.** Preserved tubular cells ultrastructure in group III compared to control group. **D.** Cells with euchromatic nuclei (N), swollen disrupted mitochondria (M), dilated enfoldings of the basolateral plasma membrane, and region of cytoplasm rarefaction (asteric) in group IV. (x 8000).

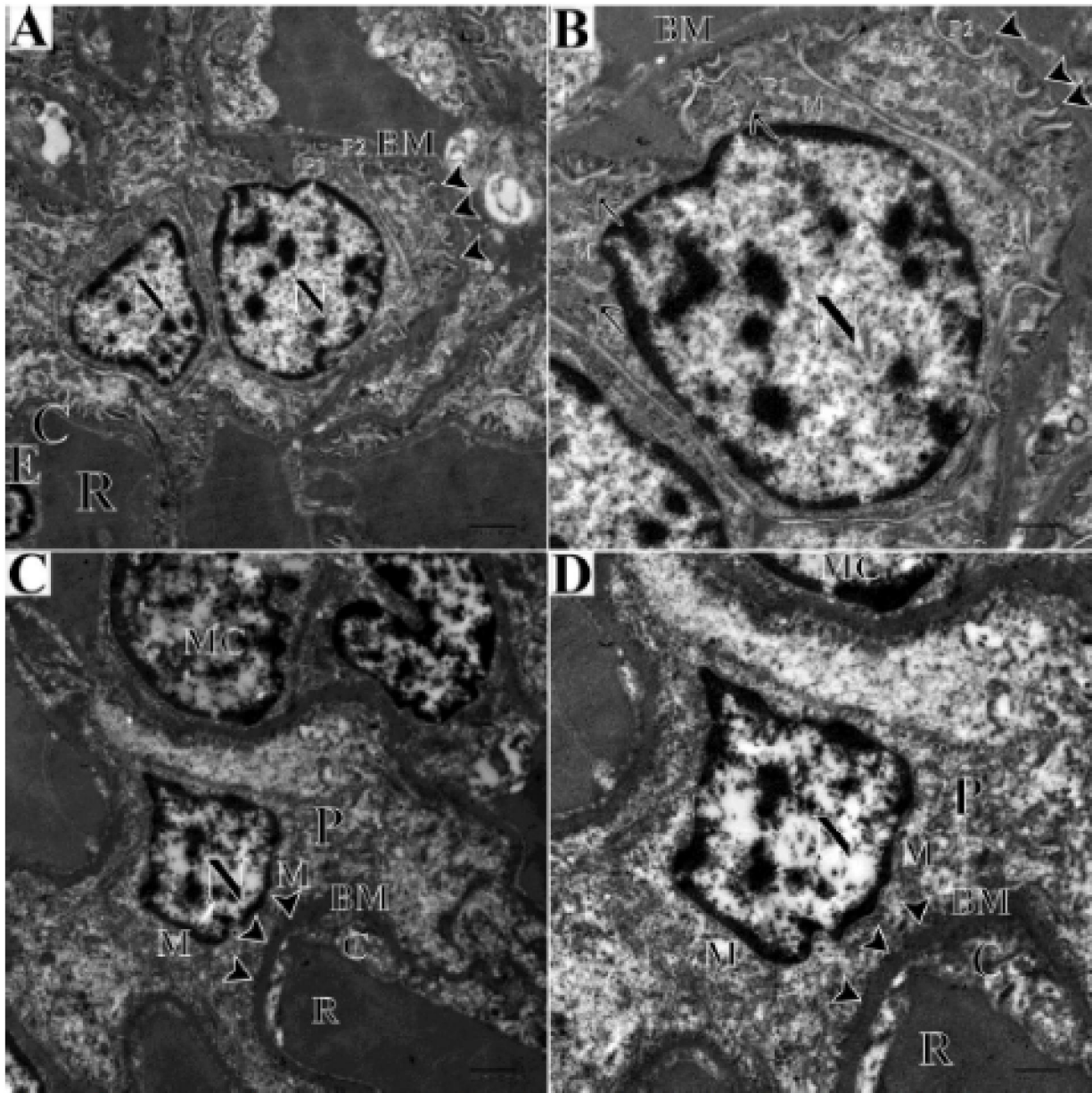


Figure 6. Transmission Electron micrograph of renal glomerulus Showing: **A.** Podocyte (P) with euchromatic nucleus (N), primary thick foot process (p1) and numerous secondary foot processes (p2) on basement membrane (BM) of glomerular capillary (C) with slit diaphragms (arrow head) in-between. Capillary was lined by discontinuous endothelial cells (E) and contained RBCs (R) in group I (x 8000). **B.** The higher magnification of the previous micrograph (x 17500). **C.** Podocyte (P) with euchromatic nucleus (N), effacement of secondary foot processes on basement membrane (BM) of glomerular capillary (C) with loss of slit diaphragms (arrow head) in-between. Capillary showed numerous mesangial cells (MC) and contained RBCs (R) of CCl₄ treated group (x 8000). **D.** The higher magnification of the previous micrograph (x 17500).

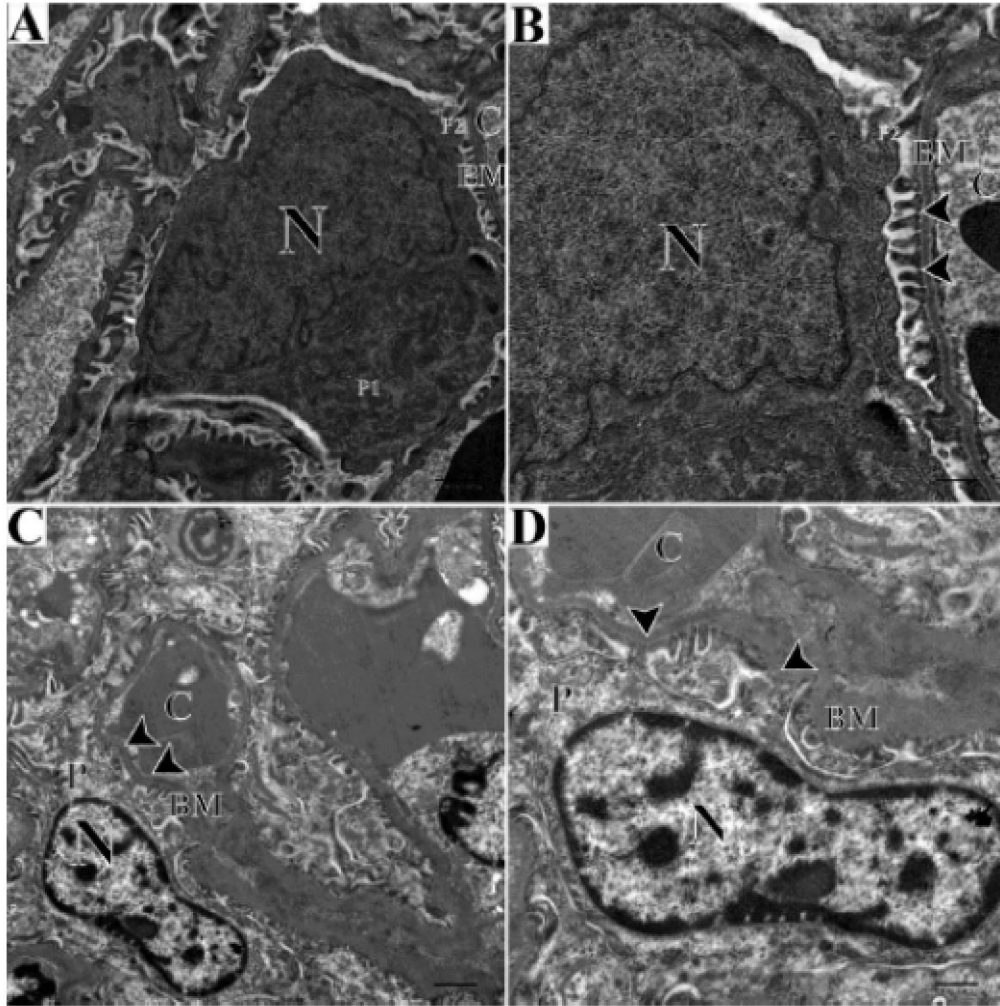


Figure 7. Transmission Electron micrograph of renal glomerulus Showing: **A.** Podocyte (P) with euchromatic nucleus (N), primary thick foot process (p1) and numerous secondary foot processes (p2) on basement membrane (BM) of glomerular capillary (C) with slit diaphragms (arrow head) in-between of group III (x 8000). **B.** The higher magnification of the previous micrograph (x 17500). **C.** Podocyte (P) with euchromatic nucleus (N), fusion of secondary foot processes on basement membrane (BM) of glomerular capillary (C) with loss of slit diaphragms (arrow head) in-between of group IV (x 8000). **D.** The higher magnification of the previous micrograph (x 17500).

4. Discussion

CCl_4 has been commonly utilized as a part of rat experimental models to investigate the toxicity induced in various tissues and organs. Previous studies had been recorded that contact to CCl_4 brings about kidney damage. The mechanism of action of CCl_4 in the tissues including reductive dehalogenation due to biotransformation of CCl_4 . Where electrophils, free radicals initiate the process of lipid peroxidation, in these condition, if the lipid peroxidation overcomes the antioxidant defense mechanism, renal damage will be occurred [15]. The harmful effects of kidneys is considered a great public health problem. Therefore, in the current investigation on rats we confirmed the possible role for protection of the renal tissues from hazard effects of CCl_4 by using of BM-MSCs

through reversing CCl_4 -induced nephrotoxicity as confirmed by microscopic examination and immunohistochemical assay.

Rats in group III by histopathological examination, showed different deviations in the structure of renal cortex, such as apparent congested dilated capillaries, flattening of the epithelial lining of some tubules, also infiltration of renal glomerulus with inflammatory cells with dilated luminal. These results go in hand with Tugba et al., (2016), who evaluated the damage effect of carbon tetrachloride on kidney and protective effect of *Amaranthus lividus* L. in rats and found that *A. lividus* has defensive and cell reinforcement impacts against CCl_4 -prompted oxidative kidney damage [16].

In the current study, BM-MSCs-treated rats showed most probably normal structure of renal tubules compared to the control group. Similar results were observed by Hamam (2015) and other authors, who investigated the impact of bone marrow-derived mesenchymal stem cells on CCl₄ in the renal cortex in adult male albino rat [17, 18]. While congestion of peritubular blood capillaries could also be seen in recovery group. These results agreed with Makni et al., (2012) who reported the nephrotoxic effect of carbon tetrachloride and DNA damage in rats [19].

Mallory's trichrome stain can reveal collagen, red blood cells, and ordinary cytoplasm. It is useful, therefore, in examining the collagen of connective tissue [20].

In the present study, Mallory's trichrome stained sections revealed increase of collagen fibers around Bowman's capsules and interstitium in CCl₄ treated rats. While no noticeable deviations were seen in the scattering of collagen fibers in the renal cortex in BM-MSCs-treated rats in comparison with control rats. Moderately distribution of collagen fibers in the renal cortex around the glomerulus and interstitium were seen in the recovery group. These findings are in accordance with D'Angelo et al., (2016) who studied the anti-inflammatory effect of GW501516 to ameliorates the renal damage, and use the Mallory trichrome stain to address the deposition of collagen around glomeruli [21].

The available data stated that MSCs play an importance role in the recent remedy as a recent approach for treating of dangerous diseases due to its ability to migrate to the site of lesion, and also due to their multi-lineage differentiation prospective [22]. Our investigation concentrated on impacts of MSCs therapy on ultrastructural and α -SMA immunohistochemical changes in nephrotoxicity-induced by CCl₄ rats. Actin is one of the components present in eukaryotic cells and considered one of the greatest plentiful cellular proteins obtainable. There are more than six isoforms were identified, between them α -SMA which is considered as a good biomarker for smooth muscle differentiation and is specified for detection of smooth muscle tissue [23]. Due to the great similarity between the mesangial cells and smooth muscle cells and their contractile properties [24], α -SMA might be expressed in mesangial cells. In the present investigation, positive reaction of α -SMA were seen in CCl₄-treated rats. While the control and BM-MSCs-treated rats revealed a negative reaction in glomerulus, renal tubules or interstitium. These results were in line with Strutz and Muller, (1996), who found that increased α -SMA immunoreactivity discovered in fibrotic areas might be due to the existence of the interstitial myofibroblasts, pericytes or perivascular adventitial cells [25]. Also Floege et al., (1992) found

that α -SMA is not shown in the glomerulus of the normal rats [26].

In this study, ultrastructural examination of the CCl₄ treated rats revealed shortened brush border of PCT and pyknosis of its nucleus. These ultrastructural changes agreed with the work of Anca et al., (2013), who found an alteration in both proximal and distal tubular epithelial cells, represented in presence of many lysosomes and dense granular bodies, altered mitochondria, appearance of "myeloid bodies" and basal enfolding dilatation instigated by CCl₄[27]. In contrast, rats treated with mesenchymal stem cell (MSCs) the proximal convoluted tubules showed improvement in the structure of kidney tissues as demonstrated by histological examination, it appears more or less normal in structure as compared with the control rats. These results were similar to Donizetti-Oliveira et al., (2012) and Huuskas et al., (2015) [28, 29].

Swollen degenerated basal infolded mitochondria of DCTs were observed in CCl₄ treated rats in addition to dilatation of basolateral plasma membrane and loss of apical plasma membrane microprojection. Similar results were observed by Gooneratne, (1986) [30]. The DCTs of BM-MSCs-treated rats had preserved tubular cells ultrastructure in this group compared to control rats. With respect to rats treated with stem cells, the PCT and DCT appeared the euchromatic nuclei and normal cytoplasmic appearance, which indicated that MCSs carry the antiapoptotic properties. In addition, some authors revealed that MCSs has the ability to release microvesicles which seemed to be an integral component of the intercellular microenvironment [31]. Other researchers (Hao et al.) reported that MCSs has an important role in cell-to-cell communication between the damaged and stem cells which appears to be bi-directional [32]. Consequently, this require to reprogramming of stem cell phenotype to gain specific characters of the tissue or inducing self-repair.

In the present study, in rats treated with CCl₄, the renal corpuscle demonstrated dilatation of the capsular space and congested glomerular capillary. Also numerous mesangial cells were observed in-between the capillary loops. While the renal corpuscle of BM-MSCs-treated rats showed most probably podocyte with euchromatic nucleus. On the contrary, other investigators found that the direct impact of transplanted MSCs to tissue restoration was minimal. The same investigators reported that 85% of MSCs disastrous to express endothelial, mesangial or monocyte/macrophage markers even through renal artery injection [33]. In this way, several investigations revealed another idea that paracrine actions of MCSs are accountable for the described

improvement of renal functions mediated by their mitogenic, angiogenic and antiapoptotic effects [34].

Conclusions

The present study demonstrated the protective potential of BM-MSCs on CCl₄-induced nephrotoxicity in adult male albino rat with better recovery findings in BM-MSCs received group than in those rats that were left to recover without treatment.

Conflict of interest

The authors declare that they have no competing interests.

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6/10/2018