R.M. Matkivska	DOI: https://doi.org/10.26641/1997-9665.2024.4.72-75
Bogomolets National Medi- cal University Kyiv, Ukraine	UDC: 616-001.43;616-022.912/.913 MICROSCOPIC CHARACTERISATION OF STRUCTURAL COMPONENTS OF
Надійшла: 14.10.2024 Прийнята: 15.12.2024	THE KIDNEYS 1 HOUR AFTER EXPERI- MENTAL RATS' EXPOSURE TO THE SCORPION LEIURUS MACROCTENUS
Прийнята: 15.12.2024	VENOM

Matkivska R.M. ២ 🖂 Microscopic characterisation of structural components of the kidneys 1 hour after experimental rats' exposure to the scorpion Leiurus macroctenus venom.

Bogomolets National Medical University, Kyiv, Ukraine.

ABSTRACT. Background. The kidneys function in the human body as important organs for maintaining the constancy of the internal environment. With their participation, constant control is carried out over such parameters as the constancy of the circulating blood volume, electrolyte composition, osmotic pressure, ion concentration, acid-base balance, blood pressure, etc. Under the influence of pathogens of various genesis, these constants undergo pronounced changes due to disruption of the structural organisation of the kidneys. Objective. Study of microscopic changes in rat kidney tissue 1 hour after exposure to the venom of the scorpion Leiurus macroctenus. Methods. The study used 10 white male laboratory rats weighing 200 g (± 10 g) grown in the vivarium of the Educational and Scientific Center "Institute of Biology and Medicine" of the Taras Shevchenko National University of Kyiv. The venom of scorpions of the Buthidae family, genus Leiurus, species Leiurus macroctenus, was administered to rats once intramuscularly (0.5 ml of venom solution previously dissolved in saline; 28.8 µg/ml; LD50=0.08 mg/kg). Kidney samples from animals of all groups were taken for microscopic examination. Histological preparations of the heart were stained with hematoxylin and eosin. Results and conclusion. It has been established that when the venom of the scorpion Leiurus macroctenus is administered, after 1 hour, toxic substances cause the development of significant hemodynamic disorders and the initiation of inflammatory processes in the organ in experimental rats, acute renal failure as a result of acute damage to the glomerular apparatus, disruption of the cytoskeleton of podocytes and the tubular apparatus of nephrons with manifestations of desquamation, hydropic, hyaline-droplet dystrophy of the tubular epithelium.

Key words: venom, scorpions, kidneys, hemodynamic disorders, rats.

Citation:

Matkivska RM. Microscopic characterisation of structural components of the kidneys 1 hour after experimental rats' exposure to the scorpion Leiurus macroctenus venom. Morphologia. 2024;18(4):72-5. DOI: https://doi.org/10.26641/1997-9665.2024.4.72-75

D Matkivska R.M. 0000-0002-4082-2899 ⊠ rujena011279@gmail.com © Dnipro State Medical University, «Morphologia»

Introduction

The kidneys are essential organs in the human body for maintaining the constancy of the internal environment. With their participation, constant control is carried out over such parameters as the constancy of the circulating blood volume, electrolyte composition, osmotic pressure, ion concentration, acid-base balance, blood pressure, etc. Under the influence of pathogens of various genesis, these constants undergo pronounced changes due to disruption of the structural organisation of the kidneys [1].

Kidney pathology is a common phenomenon.

The influence of physical and chemical environmental factors causes the development of morpho-functional disorders of the organ, which have unpredictable consequences. As is known, the kidneys are characterised by a fairly significant blood supply (about 25% of the total cardiac output), which makes them highly susceptible to the action of toxic substances since the concentration of the latter during the period of their maximum content in the blood in the renal vessels is higher than in other tissues, which contributes to the rapid damage to the endothelial layer and basal membranes of the capillary walls. The presence of a well-developed system of anastomoses is often

the cause of the vulnerability of the interstitium of the renal medulla to the effects of toxins. Compounds absorbed by the nephron tubules, interacting with the transport system of the tubular epithelium and actively accumulating in the basal parts of the cells, cause the development of degenerative and dystrophic changes in them [2]. In addition, substances secreted into the lumen of the tubules by secretion can affect the enzyme system of the nephron tubules, disrupting the normal course of biochemical reactions in them. It should also be noted that the high level of energy processes in the renal tissue determines their increased susceptibility to circulatory disorders and hypoxia [3, 4, 5]. In response to these factors, the development of stress, disruption of the microscopic structure of the organ and shifts in the physiological constants of the body are characteristic [6, 7, 8].

According to WHO, about 1.5 million cases of poisoning due to scorpion stings are registered in the world annually, leading to 2000-3000 deaths. Cases of scorpionism are common in the United States of America, Canada, Europe, and Australia. Still, this problem has become most widespread in Africa, India, Mexico, Brazil, Iran, Saudi Arabia, and Venezuela, where more than 2 billion people are under constant threat [9, 10, 11]. Among the currently known pathological processes associated with scorpionism, according to experimental studies, kidney damage is considered one of the most critical complications, which is often fatal [12, 13].

The study aims to examine microscopic changes in rat kidney tissue one hour after exposure to Leiurus macroctenus scorpion venom.

Materials and methods

The venom of scorpions of the Buthidae family, genus Leiurus, species Leiurus macroctenus, was administered to rats once intramuscularly (0.5 ml of venom solution previously dissolved in saline; 28.8 μ g/ml; LD₅₀=0.08 mg/kg) [14, 15].

The study used 10 white male laboratory rats weighing 200 g (± 10 g), raised in the vivarium of the Educational and Scientific Center "Institute of Biology and Medicine" of Taras Shevchenko National University of Kyiv. The rats were kept on a standard diet in an accredited vivarium by the "Standard Rules for the Arrangement, Equipment and Maintenance of Experimental Biological Clinics (Vivaria)". The experiments were conducted by the current regulatory documents regulating work organisation with experimental animals and compliance with the principles of the "European Convention for the Protection of Vertebrate Animals Used for Experimental and Other Scientific Purposes". Also, all work with animals was carried out by the Law of Ukraine dated February 21, 2006, No. 3447-IV, "On the Protection of Animals from Cruelty and Ethical Norms and Rules for Working with Laboratory Animals".

The rats selected for the experiment were divided into two groups: control—5 rats; no poison was administered; the material was collected one hour after the administration of saline; and experimental—5 rats; histological material was collected one hour after the administration of poison. The rats were euthanised by carbon dioxide inhalation. The kidneys of the rats were isolated at 4 °C immediately after euthanasia.

Kidney samples from animals of all groups were collected for microscopic examination. The pieces were fixed in a 10% formalin solution for 1 day. Then, they were dehydrated in alcohols of increasing concentration and embedded in paraffin blocks. Histological preparations of rat kidneys were stained with hematoxylin and eosin. Histological preparations were studied using an SEO SCAN light microscope and photographed using a Vision CCD Camera with an image output system from histological preparations.

Results and conclusions

Histological studies of kidney preparations 1 hour after the introduction of scorpion venom into experimental animals established the onset of acute renal failure as a result of acute damage to the glomerular apparatus of nephrons, the onset of significant hemodynamic disorders and the initiation of inflammatory processes in the organ.

The fibrous capsule is edematous; it easily peels off from the organ. In the interstitium of the organ around the vessels of the microcirculatory bed, there is dilation and engorgement of the vessels of the venous bed and ischemia of the vessels of the arterial bed. Perivascular oedema with infiltration by lymphocytes and macrophages-histiocytes is also present. Hemocapillaries are spasmodic and engorged, with increased intracapillary aggregation of erythrocytes.

In the cortical substance, next to the renal corpuscles, which have preserved the typical histological structure, hypertrophied renal corpuscles are found where the lumen of the afferent and efferent arterioles is spasmodic with moderate anaemia. In their lumens, erythrocyte aggregation is enhanced. The wall of the arteries and arterioles is thickened due to hypertrophy and slight leukocyte infiltration of the media, smooth myocytes, losing their spindle-shaped shape, become rounded, their nuclei are hyperchromic, and the cytoplasm is brightly oxyphilic. There is slight oedema and lymphohistiocytic infiltration in the subendothelial layer of the intima and adventitia. The endothelial cells of their intima had a cubic shape; in the cytoplasm, there was slight oedema. The nuclei are hyperchromic, and their plasmalemma is indistinct and forms significant protrusions into the lumen and invaginations. The basement membrane is continuous, but in some places, there are signs of endothelial desquamation (Fig. 1).

In the glomerular vascular glomeruli, there is anaemia; in their lumens, there is stasis. In some places, their lumens have collapsed due to spasms of the afferent arteriole. The urinary spaces were sharply dilated. In the wall of the glomerular capillaries, initial signs of focal endothelial dystrophy are observed. The cytoplasm of endothelial cells is swollen; the nuclei are hypertrophied and hyperchromic, which significantly protrude into the vessel's lumen. The basement membrane is thickened and swollen. Due to the increase in the proliferative activity of mesangiocytes, there is an expansion of the mesangium and an increase in the amount of matrix. Hyperplasia of the mesangium and pleurisy of the glomerular capillaries caused a sharp narrowing of the urinary space of the renal corpuscles; in most fields of view, it looked slitlike or was not visualised at all (Fig. 2).



Fig. 1. Microscopic structure of the kidney of white rats 1 hour after the introduction of scorpion venom. 1 - dystrophy of proximal tubules with an accumulation of cellular detritus in the lumen, 2 - the destruction of distal tubules, 3 - hypertrophied renal corpuscle, 4 - an area of haemorrhage, 5 - destructured artery. Staining with hematoxylin-eosin. ×200.

The cells of the outer leaf of the Shumlyansky-Bowman capsule are sharply flattened, their nuclei are hyperchromic with invaginations of the karyolemma, the basement membrane is thickened. The growth of the mesangium leads to a disruption of the cytoskeleton of podocytes of the inner leaf of the capsule. There is a disorganisation of the cytopodia and cytotrabeculae of podocytes, and as a result, the process of filtration disorders begins. In the proximal and distal renal tubules, the lumens are unevenly expanded with thinning of the walls. In the proximal and distal tubules, the epithelium is damaged. The brush border and basal striation are especially noticeable dystrophic changes. Focal cells almost completely lose them, and the epithelium's desquamation into the tubule's lumen is observed. The cells show signs of hydropic and hyaline-droplet dystrophy, characterised by the appearance of vacuoles and single acidophilic granules in the weakly oxyphilic cytoplasm,

indicating cytoplasmic oedema. The nuclei are hyperchromic, compacted, and have indistinct contours. Fibrin accumulations were detected in the lumens of the tubules. In thin tubules, hyperplasia of epithelial cells is due to oedema of their cytoplasm. On the crosssection, thin tubules with slit-like lumens. When tubules with moderately dilated lumens are collected, epithelial cells almost completely lose microvilli and basal striation in their cytoplasm, oedema, and single vacuoles in the weakly oxyphilic cytoplasm. The nuclei are hyperchromic and compacted. Along the course of the vessels of the microcirculatory bed of the interstitium, both in the cortex and the medulla, local single haemorrhages occur (Fig. 2).



Fig. 2. Microscopic structure of the kidney of white rats 1 hour after the introduction of scorpion venom. 1 - dystrophy of proximal tubules, 2 - destruction of distal tubules, 3 - renal corpuscles with no urinary space, 4 - area of hemorrhage. Staining with hematoxylin-eosin. ×200.

Summary

When the venom of the scorpion Leiurus macroctenus is administered, after 1 hour, toxic substances cause the development of significant hemodynamic disorders and the initiation of inflammatory processes in the organ in experimental rats, acute renal failure as a result of acute damage to the glomerular apparatus, disruption of the cytoskeleton of podocytes and the tubular apparatus of nephrons with manifestations of desquamation, hydropic, hyalinedroplet dystrophy of the tubular epithelium.

Prospects for further development are related to studying histological changes in rat kidneys under the influence of Leiurus macroctenus scorpion venom at later stages of the experiment.

Information on conflict of interest

There are no potential or apparent conflicts of interest related to this manuscript at the time of publication and are not anticipated.

References

1. Assmus AM, Mullins JJ, Brown CM, Mullins LJ. Cellular plasticity: A mechanism for homeostasis

in the kidney. Acta Physiol (Oxf). 2020;229(1):13447. doi: 10.1111/apha.13447

2. Tatarina O, Chulak O, Chulak Y, Nasibullin B. Changes in the kidney and liver structure and functions during the experimental, non-lethal load of carbon tetrachloride (CCL₄). Georgian Med News. 2021;314:150-155. PMID: 34248046

3. Abd El-Aziz FEA, El Shehaby DM, Elghazally SA, Hetta HF. Toxicological and epidemiological studies of scorpion sting cases and morphological characterization of scorpions (Leiurusquin questriatus and Androctonus crassicauda) in Luxor, Egypt. Toxicol Rep. 2019;6:329-335. doi: 10.1016/j.toxrep.2019.03.004

4. Basile DP, Yoder MC. Renal endothelial dysfunction in acute kidney ischemia reperfusion injury. Cardiovasc Hematol Disord Drug Targets. 2014;14(1):3-14. doi:

10.2174/1871529x1401140724093505

5. Bhargava P, Schnellmann RG. Mitochondrial energetics in the kidney. Nat Rev Nephrol. 2017;13(10):629-646. doi: 10.1038/nrneph.2017.107

6. Honda T, Hirakawa Y, Nangaku M. The role of oxidative stress and hypoxia in renal disease. Kidney Res Clin Pract. 2019;38(4):414-426. doi: 10.23876/j.krcp.19.063

7. Scammell MK, Sennett CM, Petropoulos ZE, Kamal J, Kaufman JS. Environmental and occupational exposures in kidney disease. Semin Nephrol. 2019;39(3):230-243. doi: 10.1016/j.semnephrol.2019.02.001

8. Thompson RH, Lane BR, Lohse CM, Leibovich BC, Fergany A, Frank I, et al. Renal function after partial nephrectomy: effect of warm ischemia relative to quantity and quality of preserved kidney. Urology. 2012;79(2):356-360. doi: 10.1016/j.urology.2011.10.031

9. Galíndez-Cerón JD, Jorge RJB, Chavez-Acosta MH, Jorge ARC, Alves NQ, Prata MMG, et al. Renal alterations induced by the venom of Colombian scorpion Centruroides Margaritatus. Curr Top Med Chem. 2019;19(22):2049-2057. doi: 10.2174/1568026619666190731143523

10. Radi ZA. Kidney pathophysiology, toxicology, and drug-induced injury in drug development. Int J Toxicol. 2019;38(3):215-227. doi: 10.1177/1091581819831701

11. Reis MB, Zoccal KF, Gardinassi LG, Faccioli LH. Scorpion envenomation and inflammation: Beyond neurotoxic effects. Toxicon. 2019;167:174-179. doi: 10.1016/j.toxicon.2019.06.219

12. Naqvi R. Scorpion sting and acute kidney injury: case series from Pakistan. Br J Med Med Res. 2015;9(10):1-6. Doi: 10.9734/BJMMR/2015/19611

13. Ranaweera GG, Bavanthan V, Nazar AL, Lokuhetty MD. Acute renal insufficiency after scorpion sting. Ceylon Med J. 2015;60(1):31-32. doi: 10.4038/cmj.v60i1.7487

14. Gunas V, Maievskyi O, Raksha N, Vovk T, Savchuk O, Shchypanskyi S, Gunas I. Study of the Acute Toxicity of Scorpion Leiurus macroctenus Venom in Rats. The Scientific World Journal. 2024;1:9746092. doi: 10.1155/2024/9746092

15. Özkan Ö, Filazi A. The determination of acute lethal dose-50 (LD50) levels of venom in mice, obtained by different methods from scorpions, Androctonus crassicauda (Oliver 1807). Turkiye Parazitol Derg. 2004;28(1):50-53.

Матківська Р.М. Мікроскопічна характеристика структурних компонентів нирок через 1 годину після впливу на експериментальних щурів отрути скорпіонів Leiurus macroctenus.

РЕФЕРАТ. Актуальність. Нирки функціонують в організмі людини як важливі органи для підтримання сталості внутрішнього середовища. За їх участю здійснюється постійний контроль таких параметрів, як сталість об'єму циркулюючої крові, електролітного складу, осмотичного тиску, концентрації іонів, кислотно-основної рівноваги, артеріального тиску, тощо. За умов впливу патогенів різного генезу дані константи зазнають виражених змін внаслідок порушення структурної організації нирок. Мета. Вивчення мікроскопічних змін тканини нирок щурів через 1 годину після впливу отрути скорпіонів Leiurus macroctenus. Методи. У дослідженні використано 10 білих лабораторних щурів-самців масою 200 г (±10 г), вирощених у віварії Навчально-наукового центру "Інститут біології і медицини" Київського національного університету імені Тараса Шевченка. Отруту скорпіонів родини Buthidae роду Leiurus виду Leiurus macroctenus вводили щурам одноразово внутрішньом'язово (0.5 мл розчину отруги попередньо розчиненому у фізіологічному розчині; 28.8 мкг/мл; ЛД50=0.08 мг/кг). Для мікроскопічного дослідження забирали зразки нирок тварин всіх груп. Фарбування гістологічних препаратів серця здійснювали гематоксиліном та еозином. Результати та підсумок. Встановлено, що при введенні отрути скорпіона Leiurus macroctenus через 1 годину токсичні речовини спричиняють в експериментальних щурів початок розвитку значних гемодинамічних розладів та запуску запальних процесів в органі, гострої ниркової недостатності як наслідок гострого ураження гломерулярного апарату, порушенням цитоскелету подоцитів та тубулярного апаратів нефронів з проявами десквамації, гідропічної, гіаліново-крапельної дистрофій епітелію канальців.

Ключові слова: отрута, скорпіони, нирки, гемодинамічні розлади, щури.