UDC 602.9 : 591.81 :[636.92.09 : 612.12 : 616.74-001.4] https://doi.org/10.31548/ujvs2021.04.010

SERUM CREATINE PHOSPHOKINASE ACTIVITY IN RABBITS DURING REGENERATION OF EXPERIMENTALLY DAMAGED MUSCLE TISSUE AND AFTER ITS STIMULATION BY TRANSPLANTED MSC

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Abstract. *According to statistics, in modern veterinary practice, the percentage of muscle injuries among sport and working animals ranges from 40–70% of sports injuries. Quite often there are cases with muscle injuries of skeletal muscles, namely extremities. This scientific work describes the research methodology, stages of research step-by-step, and studies the relationship of dynamics of the activity of a single biochemical blood*

indicator. The essence of the method was to model the injury of muscle tissue performed by the skin and fascia dissection and cutting off in the area of the midplane of the pelvic head of the biceps femoris muscle, measuring 1.5×1.5 cm to a depth of 1.5 cm of muscle tissue in 105 laboratory animals, divided into 4 groups with the use of various treatment methods. We analyze the results of one of the most effective biochemical methods for diagnosing damage of muscle fibers in the skeletal system and compare the activity of the creatine phosphokinase iso-enzyme depending on the stage of the study. Other research methods such as clinical, biochemical, ultrasonographic, and histological were recorded on 4, 7, 10, 14, 21, and 28 days. We analyzed the latest literature sources and concluded that on the 4th and 7th days, the level of creatine phosphokinase in the groups with intravenous and intramuscular administration of allogeneic mesenchymal stem cells is higher than the reference values but significantly lower compared to the control groups and the traditional method of treatment. But we observe a significant decrease in serum creatine phosphokinase levels 2 times in rabbits on the 10th day in the intravenous administration group compared with the control group of animals and in 1.6 times compared with traditional treatment. The group of animals with intramuscular administration has reference values on the 14th day, compared with the control in 1.3 times lower, traditional treatment in 1.2 times. And on the 21st day, we get reference values for a group of animals with traditional treatment. The level of creatine phosphokinase activity decreases in the control group of animals on the 28th day of the research, which indicates a complete muscle rupture. The results of studies showed that the highest activity of the creatine phosphokinase enzyme during the study was shown by groups of animals with control and traditional treatment, which indicated significant structural, functional, and destructive disorders of the muscle fibers of skeletal tissues with severe trauma. Thus, it is noted that the activity of the enzyme in conditions of damage of skeletal muscle tends to increase in accordance with the severity of the injury.

Keywords: *allogenic mesenchymal stem cells, muscle tissue regeneration, laboratory animals, activity, isoform*

Introduction

In recent years, such sports as equestrian sports have become very popular among animals. Among dog sports: agility, weight pulling, dog sled racing, dog frisbee, dog races among Greyhound breeds of dogs, and cynological training (IGP international service dog testing system, mondioring), and other training of service dogs. The relevance of this topic lies in the fact that in modern veterinary practice, muscle injuries are quite common among sports animals, namely ruptures, damage of the skeletal muscle tissue of the extremities. Therefore, the study, analysis, and effectiveness of treatment of animals with damaged muscle tissue using cellular regenerative therapy methods are very relevant, since it can significantly speed up the recovery time of animals after injury compared to traditional methods of injury treatment.

Analysis of recent researches and publications

Motion activity of animals is a coordinated contraction of skeletal muscles, which is accompanied by a change in the position of the body in space.

It is commonly known that the motion activity of a living organism is provided by the organs of the motor system. The central nervous system is responsible for body movement coordination, and the movements themselves are performed by muscles with the help of skeletal bones, their connections (joints, as well as the immobile and arthrodial connection of bones) (Mazurkevych et al., 2013; Mazurkevych et al., 2015). Muscles account for about 40–45% of the animal's body weight (Fabri et al., 2014).

Cell therapy using mesenchymal stromal cells (MSCs) is a promising approach to skeletal muscle regeneration after injuries and diseases. MSCs, which are initially present in muscle tissue or come to it from the bone marrow in response to damage, secrete various biologically active regulatory compounds that stimulate the survival, reproduction, and differentiation of cells, enhance the formation of new blood vessels (capillaries), have anti-inflammatory and antifibrotic effects (Mazurkevych et al., 2015). The ability of MSCs to produce various factors that affect all stages of the reparative process allows them to be used to accelerate regeneration, which has been repeatedly shown in various experimental models of muscle damage (Mazurkevych et al., 2009). The effect of increasing the effectiveness of transplanted MSCs can be enhanced by improving the method of cells delivering to the tissue and improving their survival; in addition, the secretory profile of cells can be changed in the appropriate direction by the method of influence of various physical or chemical stimuli or genetic modification. A new direction of regenerative medicine is the use of extracellular vesicles produced by MSCs and the regulatory molecules contained in them, primarily micro-RNAs. Activation of the regenerative potential of MSCs can be used as a tissue engineering tool in vivo that stimulates tissue repair using internal reserves (Mazurkevych et al., 2010).

Biochemical changes that occur in skeletal muscles during work are usually determined by the content of muscle metabolic products in the blood, urine, exhaled air, or directly in the muscles. For example, the maximum oxygen consumption is often used as an indicator of the intensity of aerobic processes. The intensity of glycolysis is determined by measuring the content of lactic acid. The creatine phosphokinase reaction is determined by the content of creatine and creatinine in blood; the inclusion of fats in energy exchange is identified by the content of free fatty acids and ketone bodies in blood. Conclusion about the ability of the body to resist the adverse effects of acidic products of anaerobic metabolism is made according to the indicators of acid-alkaline balance. Long-term work leads to an increase in the content of proteins, ammonia, and urea in blood, which indicates significant changes in amino acid exchange and protein metabolism (Skidanov et al., 2016).

Laboratory markers of structurally functional disorders of muscle tissue are a small number of biochemical parameters, which in a certain way reflect the processes of energy exchange and muscle damages during pathological processes (Mazurkevych et al., 2008).

Creatine kinase (CK) molecule is a dimer consisting of two types of subunits: M (muscle) and B (brain). From these subunits are formed 3 isoenzymes: BB, MB, and MM. The BB isoenzyme is mainly found in the brain, MM – in skeletal muscle, and MB – in the heart muscle. Normal CK activity should not

exceed 140-372 U/L in rabbits. The amount of the MM isoform may increase with injuries and damages of skeletal muscles (Payushina et al., 2011).

Creatine phosphokinase (CPK) is an enzyme that catalyzes the creatine phosphorylation reaction, which provides an energy substrate for muscle contraction. CPK is found in the cytoplasm and mitochondria of myocardial cells, skeletal muscles, and brain tissues, where it catalyzes the reaction:

$$\text{Creatine} + \text{ATP} \leftarrow \text{CPK} \rightarrow \text{ADP} + \text{creatinephosphate}.$$

CPK is a heterogeneous protein that consists of two types of subunits – B (brain) and M (muscle). Therefore, CPK is found in four forms: mitochondrial and three fractions of cytosolic isoenzymes (MM is found in skeletal muscles and myocardium, BB – mainly in the brain and smooth muscles, MB – in the heart muscle). Enzymes are distinct in their physicochemical and immunological properties. The level of total CPK in healthy people is represented almost entirely by CPK-MM (Boncharuk et al., 2009; Payushina et al., 2011).

Materials and methods of research

The study was conducted in the conditions of the Research and Educational Laboratory Center of Cell Technologies in Veterinary Medicine of the Faculty of Veterinary Medicine of the National University of Life and Environmental Sciences of Ukraine. Keeping animals and conducting research was carried out at the hospital on the basis of the Department of Surgery and Pathophysiology Named after Academician I. O. Povazhenko. Laboratory methods of research were carried out in the conditions of the educational and research and development clinical center Vetmedservis and the veterinary laboratory Bald.

In experiments, we used rabbits of the English Spot breed, 105 males, 3 months of age, with a weight of 2.5–3.0 kg. All selected animals had the same standard conditions of keeping and breeding, care and feeding. Rabbits are carefully selected for research and their overall health is checked. All animals were healthy according to performance in vivo. The conditions for keeping experimental animals and using them in experiments comply with the requirements and provisions of the current domestic regulatory documents and Directive No. 2010/63/EU On Protection of Animals Used for Scientific Purposes.

Prior to the study, all animals were vaccinated with the first dose at 6 weeks of age and again after 4 weeks with a combined preventive vaccine Pestorin Mormix preheating to +25 ºC, which is effective against viral hemorrhagic disease and rabbit myxomatosis. Experimental animals were kept in the appropriate conditions of the hospital of the Department of Surgery and Pathophysiology Named after Academician I. O. Povazhenko of the National University of Life and Environmental Sciences of Ukraine, all animals had identical conditions of keeping, care, and feeding. The animals were kept individually in a separate cage with the animal's number on it. The animal feeding diet was comprehensive. It consisted of sweet-smelling hay, enriched granulated feed Purina, and clean tap water from automatic drinking bowls. Everything was freely available for animals. Sterile sawdust from non-coniferous species was used for bedding. Cleaning of rabbit hutches and ventilation of the room took place

2 times per day. The room temperature ranged from +21–23 ºC, the indoor air humidity is about 70–80%. Experimental rabbits had normal conditions of changing darkness and light (12-hour cycle).

Animal experiments were conducted in compliance with the requirements of the Law of Ukraine On the Protection of Animals from Brutal Treatment and General Ethical Principles of Animal Experiments approved by the National Congress on Bioethics (Kiyv, Ukraine) on the care of laboratory animals in accordance with the Directive of EU on Protection of Vertebrate Animals used for Experimental and other Scientific Purposes (Strasbourg, 1987). The research was confirmed, verified, and approved by the local commission on Bioethics of the National University of Life and Environmental Sciences of Ukraine in accordance with Protocol No. 110/3-pr – 2018 from November 2, 2018.

Allogeneic MSCs culture was performed according to the methodological recommendations developed by the staff members of the Department of Surgery and Pathophysiology Named after Academician I. O. Povazhenko of the National University of Life and Environmental Sciences of Ukraine on Obtaining of mononuclear cells' fraction of rabbit bone marrow with high proliferative activity (Mazurkevych et al., 2017). MSCs were obtained from the red bone marrow of donor animals, taken according to the designed method of intravital obtaining of red bone marrow in small animals. It includes the selection of bone marrow in the area of the proximal and distal epiphysis of the corresponding bones of the shoulder and femur, sedation of the animal and tissues anesthesia in the area of surgical access, skin shaving, and its treatment with 5% iodine solution in the area of the proximal and distal epiphysis of the corresponding bones (Mazurkevych et al., 2016). After preparation and cleaning of the operating field, a bone marrow aspiration is performed using a medical needle for spinal anesthesia and a diagnostic puncture with Pencil-point needle bevel with a mandrel (Mazurkevych et al., 2013).

Procedures of cell isolation and manipulation of cellular material were performed in Class II Biosafety Cabinet of the Research and Educational Laboratory (ESCO). Cell culture was carried out in a CO_2 incubator (HERA CELL, Germany), which provided absolute humidity, a temperature of +37 °C, and a content of 5% of CO_2 in the air. Cell counting was performed in a Goriaev chamber using a PrimoVert microscope (Germany) with a magnification of 200 times. The total number of cells was calculated using the formula (Jennifer et al., 2011).

A cryogenic storage dewar with liquid nitrogen (SDS-20, Ukraine) was used to freeze the cells. Defrosting of the cells was carried out at a temperature of +37 °C in a water bath (EL 20, Poland). Culture media, other solutions, components, preparations were stored at a temperature of 4 °C and -18 °C in a household refrigerator Nord (Ukraine). Centrifugation of cell-rich fluids was performed by centrifuge (UNICO, USA). A TC-80M thermostat (Ukraine) was used to heat the solutions. Dehumidification and sterilization of laboratory utensils were carried out in a hot-air sterilizer HS–62A (Poland) and in an air sterilizer GPO-50 (Ukraine).

The animal was kept on a starvation diet for 12 hours before the operation. The operation of experimental muscle tissue injury was performed under general anesthesia. After fixing the animal

on the operating table, the operation field was freed from hair fibers by shaving with an electric machine and treated with a solution (Kutasept F).

Before introducing the animal to general anesthesia, an anticholinergic agent was intramuscularly introduced. It blocks mainly peripheral cholinoreactive systems, as well as atropine sulfate at a dose of 0.05 mg/L kg of animal body weight for prevention of bronchospasm and laryngospasm, decreasing gland secretion, reflex reactions, and side effects caused by excitation of vagus nerve. In due course of time, the drug Zoletil 100 was administered intramuscularly at a dose of 8 mg/L kg of animal body weight.

At the site of surgical access, infiltration anesthesia was previously performed with a 0.5% novocaine solution (a dose of 3 mL/animal) in the planned part of the simulated defect. Modeling of muscle tissue injury was performed by skin and fascia dissection process and cutting off the midplane of the pelvic head of the biceps femoris muscle in the measure of 1.5×1.5 cm to a depth of 1.5 cm of muscle tissue, which is shown in Fig. 1.

After cutting off 1.5 cm in width and 1.5 cm to the deep of muscle tissue (from the pelvic head of the biceps femoris), the wound was measured using a stationery ruler and a surgical scalpel with a replaceable blade. During the operation, each of the animals was placed on the operating table covered with a wool blanket to prevent excessive heat loss. All surgical procedures were performed in accordance with the requirements of aseptic and antiseptics (Mazurkevych et al., 2015).

Animals with experimental muscle tissue injuries were divided into 4 main groups. Rabbits of the 1st group were injected into the site of the experimental defect using an insulin syringe with pre-prepared doses (in the amount of 3 million/animal) of allogeneic MSCs, which were obtained according to methodological recommendations developed by the staff of the Department of Surgery and Pathophysiology Named after

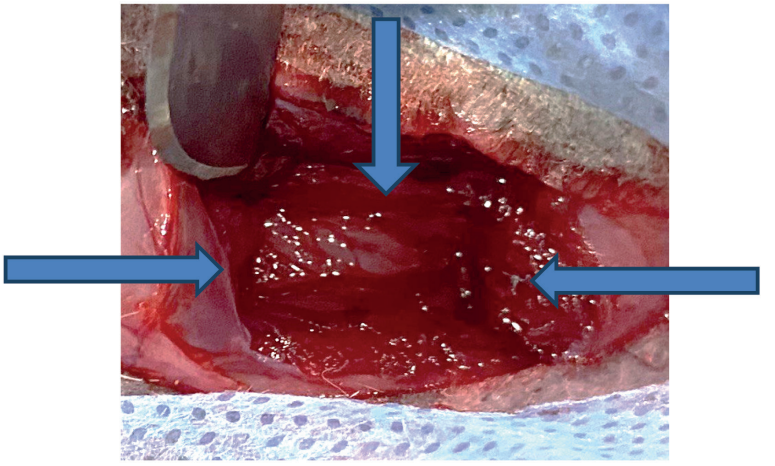

Fig. 1. Arrows show a simulated wound defect on the photo (size of 1.5×1.5 cm to a depth of 1.5 cm)

Academician I. O. Povazhenko of the National University of Life and Environmental Sciences of Ukraine on Obtaining of mononuclear cells` fraction of rabbit bone marrow with high proliferative activity (Mazurkevych et al., 2009). Cultivation took place from biological material (red bone marrow of donor animals), taken according to the designed method of intravital production of red bone marrow in small animals. It includes a selection of bone marrow in the area of the proximal and distal epiphysis of the corresponding bones of the shoulder and femur, sedation of the animal and tissues anesthesia in the area of surgical access, skin shaving and its treatment with 5% iodine solution, in the area of the proximal and distal epiphysis of the corresponding bones (Mazurkevych et al., 2016; Pristupa et al., 2018). After preparation and cleaning of the operating field, a bone marrow aspiration is performed using a medical needle for spinal anesthesia and a diagnostic puncture with Pencil-point needle bevel with a mandrel (Mazurkevych et al., 2017; Payushina et al., 2019).

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A cryogenic storage dewar with liquid nitrogen (SDS-20, Ukraine) was used to freeze the cells. Defrosting of the cells was carried out at a temperature of +37 °C in a water bath (EL 20, Poland). Culture media, other solutions, components, preparations were stored at a temperature of 4 °C and -18 °C in a household refrigerator Nord (Ukraine). Centrifugation of cell-rich fluids was performed by centrifuge (UNICO, the USA). A TC-80M thermostat (Ukraine) was used to heat the solutions. Dehumidification and sterilization of laboratory utensils were carried out in a hot-air sterilizer HS–62A (Poland) and in an air sterilizer GPO-50 (Ukraine).

After that, the fascia and skin were stitched with synthetic polyfilament absorbable ligatures and sutures Vicryl (Belgium).

Animals of the 2nd experimental group were injected with the same amount of allogeneic MSCs into the bloodstream by puncturing the jugular vein at the edge of the upper and middle third of the neck. After that, the fascia and skin were stitched with synthetic polyfilament absorbable ligatures and sutures Vicryl (Belgium).

Animals of the 3rd experimental group were prescribed traditional trauma treatment, namely:

1. Surgical method: interrupted stitching on the site of muscle tissue rupture; fascia and skin were sewn with synthetic polyfilament absorbable ligatures and sutures Vicryl (Belgium), muscle tissue was sewn with absorbable material (wicker Chirasorb braided).

2. Pharmacological method: in the place where the wound was sewn up, we smeared 0.5 ml of surgical glue Dermabond. There were intramuscularly administered 0.2 mL/kg of animal body weight once a day for 5 days. And Tylosin 5% at a dose of 6 ml/L animal once a day for 5 days.

Animals of the 4th experimental group (control group) were given an

intravenous 0.9% sodium chloride solution. In addition, there was the 5th experimental group – intact animals.

In the postoperative period, the animals were provided with complete rest, proper conditions for keeping, feeding and watering in accordance with the requirements.

Animal from each experimental group was removed with the use of euthanasia (after deep anesthesia) at a certain stage. In such a way, we took muscle tissue samples for morphological and histological studies. In the postoperative period, the animals were provided with complete rest, proper conditions for keeping, feeding and watering in accordance with the requirements. A clinical examination of animals was conducted during the entire study period.

Samples of biological material for laboratory tests were taken at the initial condition (before the start) and on the 4th, 7th, 10th, 14th, 21st, and 28th days of the experiment (Table 1). The animal was fixed, the skin was stretched in the area of the jugular vein passage at the edge of the upper and middle third of the neck, and the animal's head was taken to the other side for obtaining blood samples for laboratory tests. The puncture was performed by needling the skin (inserting the needle into the blood flow at an angle of 45 ° and the vein wall. The selected fresh blood was placed in a coagulant tube (Fig. 2).

Biochemical studies were conducted in the veterinary laboratory Bald by the kinetic method using a biochemical analyzer RT-9600, using commercial kits of the company Filisit-Diagnostics (Ukraine) according to the instructions.

Statistical processing of the obtained digital data was carried out using a package of statistical programs Microsoft Excel. The arithmetic mean was calculated using the Student's t-test. The difference between the values was considered a statistically reliable result, in accordance to which coefficient (P) was no more 0.05 that is common in biological research.

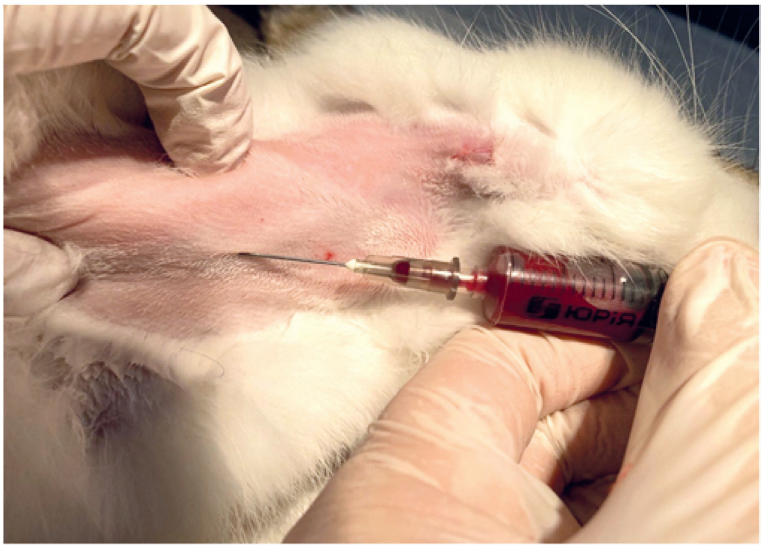


Fig. 2. Blood sampling from the jugular vein

1. The number of animals in experimental design

Animal groups	Initial condition	4th day	7th day	10th day	14th day	21st day	28th day
Administration of MSCs in the muscles	3	3	3	3	3	3	3
Administration of MSCs into blood	3	3	3	3	3	3	3
Traditional treatment	3	3	3	3	3	3	3
Control group	3	3	3	3	3	3	3
Intact animals	3	3	3	3	3	3	3
Total animals used:			105				

Results of the research and their discussion

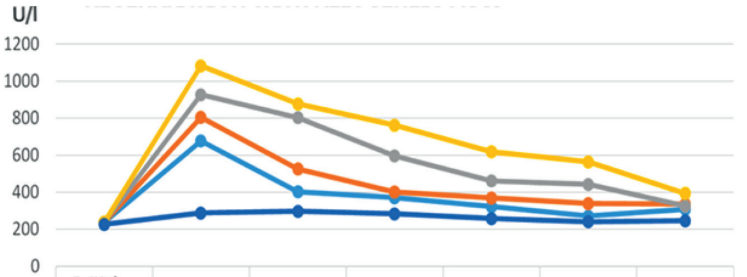
The findings of CPK activity in rabbit blood associated with the activity of regeneration of experimentally injured muscle tissue in different periods of the experiment are shown in Table 2.

The results of the analysis of the obtained data indicate that changes in the activity of the creatine phosphokinase type (MM) isoenzyme occurred during the studies. As can be seen from Table 1 and Fig. 3, on the 4th day after transplantation of allogeneic MSCs, the level of creatine phosphokinase in blood of animals of all groups was the highest for the entire experimental period. CPK activity in blood serum of animals of the 1st and 2nd experimental groups significantly exceeds the reference val-

2. Creatine phosphokinase activity in blood of rabbits after application of allogeneic MSCs for stimulating myogenesis in experimentally damaged muscle tissue, U/L (M ± m, n = 3)

Animal groups	Initial condition	After applying MSCs					
		4th day	7th day	10th day	14th day	21st day	28th day
Administration of MSCs in the muscles	237.8 ± 52.4	676.8 ± 8.6 ***	402.6 ± 3,7**	370.6 ± 3**	322.5 ± 18.3**	271.4 ± 52.7**	308 ± 21.9*
Administration of MSCs into blood		804.3 ± 54.4**	525.7 ± 32.5**	400.9 ± 8.1**	368.5 ± 4.5**	338.7 ± 34 **	335.5 ± 26.4
Traditional treatment		926.3 ± 39.6*	802.6 ± 11	596.2 ± 69.5	460.7 ± 61.5	442.6 ± 31.7	325.1 ± 49
Control group		1082.6 ± 40.2	877 ± 63.3	761.8 ± 36.5	618.4 ± 47.7	563.9 ± 15.2	393.6 ± 6.5
Intact animals	226.6 ± 21.2	287.5 ± 15.4**	296.9 ± 27.2**	283.3 ± 19.5**	257.7 ± 41.2**	240.9 ± 33.5**	245.9 ± 43*
Total animals used:			105				

Note: * P < 0.05; ** P < 0.01; *** P < 0.001.



	Initial condition	4th day	7th day	10th day	14th day	21th day	28th day
MSCs into muscle	237,8	676,8	402,6	370,6	322,5	271,4	308
MSCs into blood	237,8	804,3	525,7	400,9	368,5	338,7	335,5
Traditional treatment	237,8	926,3	802,6	596,2	460,7	442,6	325,1
Control group	237,8	1082,6	877	761,8	618,4	563,9	393,6
Intact animals	226,6	287,5	296,9	283,3	257,7	240,9	245,9

Fig. 3. Creatine phosphokinase activity in blood of rabbits after application of allogeneic MSCs for stimulating myogenesis in experimentally damaged muscle tissue

ues but is significantly lower compared with this indicator in animals of the 3rd groups (traditional treatment) and the 4th (control group). Later, on the 7th, 10th, 14th, 21st, and 28th days of the experiment, there is a gradual decrease in the enzyme activity with slight fluctuations in the groups, but with unchanged dynamics between the groups.

It is known that an increase in the activity of this enzyme in blood indicates the destruction of myocytes. The dynamics of its activity allow to determine the degree of muscle injury and the intensity of recovery of damaged muscle tissue (Francchi et al., 2021). In the first 4 days of the experiment, the highest activity of CPK obviously indicates significant destruction of damaged muscle tissue as a result of the active phase of the inflammatory process, stage of its alteration, and vascular reaction.

The lowest level of CPK activity in blood of animals was observed during the entire observation period in the 1st experimental group, which gives grounds to conclude that allogeneic MSCs transplanted directly into the inflammatory zone are reliably highly effective.

The second place in terms of effectiveness is the animals of the 2nd group that were administered MSCs intravenously.

The traditional method of treating experimentally damaged muscle reliably ranks third place. It's significant to pay attention to the dynamics of indicators in the intermediate observation period between the 4th and 28th days. In particular, CPK activity in the blood decreased against the initial condition in animals of the 1st and 2nd experimental groups, while in animals of the 3rd experimental group (traditional treatment), this indicator only approached but did not reach this level only on the 14th day. It indicates a significantly lower activity of regenerative processes in experimentally injured muscle tissue.

Creatine phosphokinase activity of the control group decreases on the 28th

day of the study. It indicates severe consequences of an injury of this nature without medical care for the body.

Conclusions and future prospects

Application of allogeneic mesenchymal stromal cells for stimulation regenerative processes in experimentally damaged muscle tissue by the method of administration it directly into the area of damage significantly increases the regeneration activity of damaged tissue and reduces the healing time more effectively than the method of administration it into the blood, as well as in comparison with the method of traditional treatment.

Method for modeling of experimental damage in muscle tissue by damage of muscle tissue in the pelvic head of the biceps femoris of rabbits is a possibility to study the comparative effectiveness of the stimulating effect of transplanted allogeneic mesenchymal stem cells.

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Стадник Н. В., Бокотько Р. Р. , Савчук Т. Л. , Куліда М. А., Мазуркевич А. Й. (2021). АКТИВНІСТЬ КРЕАТИНФОСФОКІНАЗИ В СИРОВАТЦІ КРОВІ КРОЛІВ ЗА РЕГЕНЕРАЦІЇ ЕКСПЕРЕМЕНТАЛЬНО УШКОДЖЕНОЇ М'ЯЗОВОЇ ТКАНИНИ ТА ПІСЛЯ ЇЇ СТИМУЛЯЦІЇ ТРАНСПЛАНТОВАНИМИ МСК.

Ukrainian Journal of Veterinary Sciences, 12(4): 127–139,
https://doi.org/10.31548/ujvs2021.04.010

Анотація. *За статистикою в сучасній ветеринарній практиці відсоток м'язових травм серед спортивних та робочих тварини складає від 40–70% спортивних травм. Досить часто трапляються випадки із м'язовими травмами скелетних м'язів, а саме кінцівок. У цій науковій праці описано методику досліджень, етапи досліджень та вивчено взаємозв'язок динаміки активності певного біохімічного показника крові. Суть методу по-*

лягала в моделюванні травми м'язової тканини проведеного методом розтинання шкіри та фасції і відсікання в ділянці серединної площини тазової голівки двоголового м'яза стегна, розміром 1,5×1,5 см на глибину 1,5 см м'язової тканини у 105 лабораторних тварин, поділених на 4 групи за застосування різних методів лікування. Проаналізовано результати одного із найефективніших біохімічних методів діагностики ушкодження скелетних м'язових волокон та порівняно активність ферменту ізоферменту креатинфосфокінази (ММ) залежно від етапу дослідження. Інші методи досліджень такі як, клінічні, біохімічні, ультрасонографічні та гістологічні реєструвалися на 4, 7, 10, 14, 21, 28 добу. Нами проаналізовано останні літературні джерела та зроблено висновки, що на 4 та 7 доби рівень креатинфосфокінази в групах із внутрішньовенним і внутрішньом'язовим введенням алогенних мезенехімальних стовбурових клітин вище референтних значень, але значно нижче, ніж у групах контролю та з традиційним методом лікування. А от значну динаміку зниження рівня креатинфосфокінази в сироватці крові у кролів ми спостерігаємо на 10 добу в групі із внутрішньовенним введенням порівнюючи із контрольною групою тварин у 2 рази та в 1,6 раза порівнюючи із традиційним лікуванням. Група тварин із внутрішньом'язовим введенням має референтні значення на 14 добу, порівнюючи із контролем нижче в 1,3 раза, традиційним лікуванням – в 1,2 раза.

Та на 21 добу отримуємо референтні значення в групи тварин із традиційним лікуванням. Рівень активності креатинфосфокінази знижується в контрольної групи тварин на 28 добу дослідження, що свідчить про повний м'язовий розрив. Результати досліджень показали, що найбільш високу активність ферменту креатинфосфокінази впродовж дослідження показали групи тварин контрольна та із традиційним лікуванням, що свідчило про значні структурно-функціональні й деструктивні порушення м'язових волокон скелетної м'язової тканини із тяжкою травмою. Отже відзначено, що активність ферменту за умов ушкодження скелетної м'язової тканини має тенденцію збільшуватися відповідно до ступеня тяжкості травми.

Ключові слова: *алогенні мезенхімальні стовбурові клітини, регенерація м'язової тканини, лабораторні тварини, активність, ізоформа*