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Locomotor activity and spasticity level of the limb in female mice with a spinal cord injury model

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ABSTRACT

Spinal cord injuries (SCI) lead to large-scale physical, physiological, psychological, and professional losses. Therefore, today, one of the urgent aims of neurophysiology is the study of the consequences of spinal cord injury. For a comprehensive and detailed study of the functional post-traumatic recovery of the spinal cord, various options of its hemisection as a most common type of SCI are modeled.

***THE PURPOSE** of the study was to analyze the level of locomotor activity and changes in spasticity of the ipsilateral hindlimb (IH) after SCI modeling by hemisection in female mice.*

***MATERIALS AND METHODS.** The injury of the left-side spinal cord hemisection was modeled at the level of the lower thoracic segments (T10-T11) in female FVB mice. The locomotor activity and spasticity of the IH were determined every week using the Basso-Beattie-Bresnahan (BBB), the Basso (B) and the Ashworth scales during the first 1-12 weeks of the post-traumatic period.*

***RESULTS.** At a later term (the 11th-12th weeks) compared with the first weeks of the post-traumatic period, there was a marked recovery of the IH function: 4.39 ± 0.61 points out of 21 possible on the BBB scale and 2.22 ± 0.31 points out of 9 possible on the B scale. However, at all studied time intervals after SCI, a consistently high level of hindlimb spasticity in experimental animals was noted, on the 12th week 3.03 ± 0.39 out of 4 possible by the Ashworth scale.*

***CONCLUSION.** After modeling spinal cord injury by hemisection in mice, spontaneous post-traumatic recovery of locomotor activity was observed (since the 2nd week), which had scores corresponding to the early recovery phase. On the other hand, the levels of IH spasticity were quite high throughout the entire study period, although already at the 2nd week there was a slight decrease in this parameter compared to the 1st week of the post-traumatic period.*

***KEY WORDS:** spinal cord injury; modeling of spinal cord hemisection; locomotor function; spasticity; the BBB scale; the B scale*

Spinal cord injuries (SCI) are one of the most serious conditions which lead to long-term functional complications, chronic pain and a general decline in the quality of life. The general consequences of SCI are influenced by the patient's age, the severity of the injury, as well as social and economic conditions the affected person lives [1, 2].

Every year about 2.4 thousand new cases of SCI are recorded in women in the United States. In its turn today, 39,000 out of 27 million women with a physical disability in the United States have been disabled due to various types of spinal cord injury [3]. In women, neurological changes due to SCI can adversely affect reproductive health and fertility. Usually, the reaction of the reproductive system to SCI is manifested in abnormal menstruation and uterine bleeding, neurogenic prolactinemia, and galactorrhea. It is also more difficult for women with SCI to get pregnant and carry a pregnancy. In addition, SCI in women causes respiratory diseases, inflammation of the urinary tract, and even thrombophlebitis. At later stages of the post-traumatic period, contracture develops, which complicates the movement of the limbs and there is ossification of the knee, shoulder, and elbow joints [4].

Different models *in vitro* and *in vivo* are being developed to further study the pathogenesis and identify key aspects of physiological recovery from SCI. The modeling of SCI is mainly performed on laboratory animals, in particular rodents. The modeling of SCI *in vitro* during short- and long-term cultivation is usually performed either on organotypic slices or on monolayer adhesive cultures of neural tissue cells. The most common ones are the model of compression on organotypic cultures of spinal cord tissue, the hemisection of organotypic cultures, and the model of glial scar formation *in vitro* [5, 6]. In its turn, the modeling of spinal cord segment removal, contusion [7-9], compression [7, 10], ischemia-reperfusion [11-13], photochemically-induced SCI [14, 15], complete section, an incomplete section of the spinal cord *in vivo* allow to more comprehensively elucidate the pathogenesis of neural tissue damage and assess its endogenous regenerative potential [5, 6].

Various modifications of the spinal cord transection model are used for detailed morphofunctional research, as well as to establish the level of neural tissue recovery [9, 16, 17]. The following variants of the spinal cord transection model are used: complete and partial transection. The complete transection of the spinal cord in humans is much less common. According to the literature, this experimental model in rats and mice is most appropriate for the study of axon regeneration after injury or selection of different types of polymer matrices to determine the characteristics of neural tissue regeneration. It should be noted that one of the important advantages of this model is the exclusion of the development of spontaneous neural plasticity and ease of reproduction of the experiment. However, the model of complete transection has a number of disadvantages, in particular, leads to high mortality and causes urinary incontinence in males due to anatomical features and innervation [6, 17]. Due to severe post-traumatic urinary incontinence, the complete transection is usually modeled on adult rats (with mandatory urethral catheterization) and adult female mice. This type of injury eventually leads to further retrograde urethral infection with *E. coli, Klebsiella, Enterococcus, and Staphylococcus,* which adversely affects the survival and behavior of laboratory animals [18, 19].

On the other hand, the modeling of SCI by partial transection does not lead to disruption of the urinary system as dramatically as at complete transection, and thus there is almost no retrograde infection of the urinary tract. Moreover, the hemisection model does not involve transection of the vertebral artery, which prevents the occurrence of unwanted hemorrhages, as is observed in a complete spinal cord transection. In addition, one of the advantages of such a model is the presence of a control part of the spinal cord within one experimental animal. Spinal cord hemisection is best modeled to study nerve fiber growth and synaptic plasticity. Compared with the complete spinal cord transection, the model of spinal cord hemisection is similar to clinical cases of neural tissue injury in humans. The model of spinal cord hemisection does not lead to severe post-traumatic consequences, as it is observed at complete spinal cord transection, but is more often characterized by spontaneous recovery. Modeling of spinal cord hemisection is usually performed to assess locomotor activity and study the restoration of spinal cord tracts [6, 17].

The model of partial transection is generally a hemisection of the spinal cord. This type of SCI leads to partial or complete loss of motor and vegetative functions and sensory sensitivity below the level of injury. The ratio of injuries with incomplete spinal cord injury is 52.8 % and 44.3 % according to European and American data, respectively. There are also five syndromes of incomplete SCI: central cord syndrome, Brown-Séquard syndrome, anterior cord syndrome, *conus medullaris* syndrome, and *cauda equina* syndrome [20]. The most common is Brown-Séquard syndrome, which was described in 1862 and manifested by weakness or paralysis, impaired proprioceptive function and ipsilateral body, and loss of pain and temperature sensitivity of the contralateral body [20, 21].

Currently, hemisection modeling is most often performed on rats and mice. SCI in males, in contrast to female mice, causes a higher mortality rate due to the anatomical features of the male urogenital system. While the recovery of motor function after SCI in females is probably better than in males, because the CNS responds differently to trauma in females and males, and features of pathophysiological processes may be due to gender differences [22].

The survival of laboratory mice after SCI also differs between the sexes: 58 % in males and 61 % in females. The natural death of animals usually occurs at the age of 15 months due to lung cancer or liver disease. In 20 % of females, spontaneous death is associated primarily with neuroendocrine disorders that can cause mutations, such as in the *FVB/Cr* substrain and the NCr-derived stock2 strain. According to another hypothesis, the syndrome of "spontaneous death" is a long-term condition only in female mice of *FVB* strain. In general, the described syndrome causes neuronal necrosis in the spinal cord, associated with hypoxinemia and, consequently, changes in animal behaviour. The syndrome is often accompanied by epileptic seizures, as well as tumors of the urethra and gallbladder, and adrenal hypertrophy [23, 24].

SCI models (*in vitro* and *in vivo*) can be used to study various cell therapy protocols for this injury; and, in general, to open new opportunities in the study of the regenerative ability of neural tissue at the spinal cord level. Given that the modeling of the spinal cord hemisection in males has more negative consequences; this affects the possibility of a long-term study of the implantation of scaffolds and stem cells of different origins to regenerate spinal cord tissue.

Therefore, the **PURPOSE** of our study was to analyze the level of locomotor activity and spasticity of the ipsilateral hindlimb (IH) after SCI modeling by hemisection in female mice.

MATERIALS AND METHODS

Modelling of SCI in vivo. The injury of the left-side spinal cord hemisection was modeled at the level of the lower thoracic segments (T10-T11) of female *FVB* mice, aged 2-3 months and weighing 20-25 g. Mice were divided into two groups: 1 – control, sham-operated animals (n = 9), 2 – animals with spinal cord hemisection (n = 64). It should be noted that this is the first time we have applied such a model of SCI to this strain of mice.

The animals were kept on a standard diet in the vivarium of Bogomoletz Institute of Physiology of the National Academy of Sciences of Ukraine. All experiments with animals were carried out in compliance with the Law of Ukraine "On Protection of Animals from Cruelty" [25], "European Convention for the Protection of Vertebrate Animals Used for Experimental and Other Scientific Purposes", principles of bioethics and biosafety standards [26]

The mixture of 50 mg/kg ketamine (*Alfasan*, the Netherlands) and 25 mg/kg of xylazine (*Biolec*, Ukraine) was used for anesthesia. All manipulations with animals were performed under aseptic conditions using disinfectant solutions to treat the site of surgery and sterile instruments for the actual SCI modeling.

After anesthesia, the animals' eyes were lubricated with 0.2 % Vidisik gel (*Dr. Gerhard Mann Chem.-pharm. Fabric GmbH*, Germany) to prevent them from drying out during surgery. Next, the last ribs of the animal were determined by palpation, which corresponded to the T11 vertebra. Then a longitudinal incision of the skin about 2 cm was made along the spine, and the subcutaneous fat and intervertebral muscles of 1-2 vertebrae were carefully dissected. One of the branches of ophthalmic scissors was inserted along the left edge of the posterior middle artery into the spinal cord at the level of T10-11 segments and the left part of the spinal cord was dissected. For the group of sham-operated animals, all the above steps were performed, except for spinal cord hemisection. After hemostasis, muscles, fat, and skin were sutured. The skin of animals was treated with 10 % solution of betadine (*Egis*, Hungary).

For 12 weeks after the modeling of spinal cord hemisection, the locomotor activity of the ipsilateral hindlimb was assessed weekly using the Basso-Beattie-Bresnahan (BBB) and the Basso (B) scales; the assessment of the level of the limb spasticity was performed using the Ashworth scale.

Determining of the functional recovery level by the BBB scale. The BBB scale, developed in 1995, is used to determine the consequences of various types of SCI in the study of locomotor activity of rodents [27]. The BBB scale reflects the functional features of the hindlimb, taking into account motor activity in the joint, movement, walking, coordination of fore and hindlimbs, body position and stability, as well as foot and tail position. There are three stages of the recovery of IH motor function after SCI within the BBB scale (0-21 points):

- early stage (score 0-7) is characterized by single movements in the knee joint with no or low level of motor activity of the hindlimb;

- middle stage (score 8-13) represents uncoordinated movements of the hindlimbs with/without holding the body;

- late stage (score 14-21) describes the coordination of the fore and hindlimbs and the position of the tail [28].

Determining of the functional recovery level by the B scale. The Basso scale, developed in 2006, allows us to assess the recovery of locomotor activity of mice hindlimb after modeling spinal cord hemisection. There are four stages of the recovery of motor function after injury by the B scale:

- the early phase of the post-traumatic period after hemisection modeling is characterized by hindlimb paralysis and/or paresis, lack of motor activity in the knee joint, which corresponds to 0-2 points on the B scale;

- at the middle phase of the post-traumatic period there is a plantar posture of the IH foot (3-4 points);

- at the late phase of recovery (5-8 points) there is the coordination of the fore-hindlimbs and the stability of the body;

- 9 points of the B scale correspond to the normal IH locomotor activity and stable body position [29].

Assessment of the spasticity level. The Ashworth scale, developed in 1964, was used to assess the level of hindlimb muscle spasticity in experimental animals. The Ashworth scale consists of four points:

- 0 points correspond to the lack of muscle tone;
- 1 point characterizes a slight increase in muscle tone and low resistance during flexion and extension of the limb;
- 2 points indicate an increase in muscle tone and moderate resistance during flexion and extension of the limb;
- 3 points reflect significant muscle tone and difficulty in performing passive movements;
- 4 points characterize the rigidity of the limb muscles during flexion and extension [30].

Statistical analysis of results. The results of the BBB, B, and Ashworth behavioral tests were compared by Wilcoxon within the group after hemisection each week with the previous weeks for 1-12 weeks. To determine the level of differences significance between the mean values in the control and experimental groups at different times of the study we used the t-Student test. Data are presented as Mean ± SEM. The differences were considered significant at $p \leq 0.05$.

RESULTS AND DISCUSSION

For our research, we chose *FVB* mice, as very few models have been developed so far for SCI in rodents, in particular in laboratory mice, which now account for about 70 % of experimental animals. *FVB* mice are widely used in many diverse studies. For example, females of the *FVB* strain, aged 5-8 weeks, were used to study the effect of encephalomyelitis virus on neural tissue, namely the development of acute and chronic inflammation, and axillary myelination [31, 32]. To study the consequences of spinal cord tissue infection with Theiler's virus, female mice of the *FVB* strain, aged 5-8 weeks, were also selected as experimental animals [33, 34]. In addition, the effects of streptozotocin- and albumin-induced models of diabetes were studied on female *FVB* mice [35]. Metabolic studies have also been performed on *FVB* mice with a deletion of angiotensin II type 2 receptor [36]. *FVB* mice are also actively used in the study of various tumors. Thus, squamous cell carcinoma and breast cancer are modeled in *FVB/N* mice [37, 38].

In order to study the features of functional recovery and the level of IH spasticity in *FVB* female mice, spinal cord hemisection was modeled. After modeling the hemisection of the spinal cord, IH locomotor activity of experimental animals was evaluated on the BBB and B scale, in turn, the level of spasticity of ipsilateral hindlimb was evaluated on the Ashworth scale weekly for the first 12 weeks. It should be noted that all operated animals (n = 64) survived during this period of post-traumatic examination (1-12 weeks).

The level of locomotor activity recovery of IH was determined by the BBB scale. Already on the 1st week of the post-traumatic period, we noted visible spontaneous motor activity, weak movements in one or two, and common movements in the third joint, which corresponded to the average score on the BBB scale – 1.29 ± 0.16 (**Fig. 1**). At the 2nd, 3rd and 4th weeks of the post-traumatic period, the mean score on the BBB scale was 1.36 ± 0.16, 1.75 ± 0.22 and 2.26 ± 0.28, respectively. At the functional level that was manifested in visible spontaneous motor activity, weak or widespread movements in one or two or all three joints. From the 3rd week the functional levels of IH in comparison with the 1st week were significantly higher ($p < 0.01$).

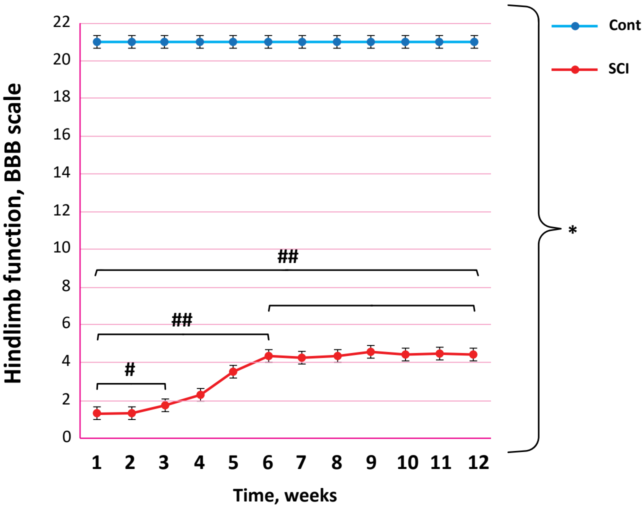

Fig. 1. The levels of locomotor activity of ipsilateral hindlimb in female FVB mice after spinal cord hemisection by the BBB scale.

Notes: * – $p < 0.001$ compared to the control at 1st-12th weeks; # – $p < 0.01$, ## – $p < 0.00001$ compared to the 1st week in the experimental group

During the 5th week of the post-traumatic period, a significant improvement in IH locomotor activity was observed in most mice compared with the 3rd week ($p < 0.00001$). That was detected in weak movements in one or two, or in all three joints of the limb, and step synergies while holding body balance. The average score on the BBB scale was 3.54 ± 0.44 (**Fig. 1**). At the 6th week of the post-traumatic period, we continued to note an increase in the rate up to 4.37 ± 0.54 compared with the 5th week ($p < 0.00001$) (**Fig. 1**). At the functional level it was manifested in mostly common movements in one or two or all three joints of the limb, step synergies, constant holding body balance and frequent coordination of the fore and hindlimbs.

At 7th, 8th, 9th, and 10th week there was no significant improvement in locomotor activity of female mice IH on the BBB scale compared with the 6th week: 4.24 ± 0.54, 4.41 ± 0.58, 4, 56 ± 0.60 and 4.46 ± 0.59, respectively (**Fig. 1**). The post-traumatic period during the 11th and 12th weeks was marked by slight fluctuations in the mean scores on the BBB scale: 4.43 ± 0.60 and 4.39 ± 0.61, respectively (**Fig. 1**). At the functional level, as previously (starting from the 7th week) characterized by widespread movements in all three IH joints, frequent or constant holding of body, plantar posture, step synergies sole downwards, frequent or constant holding of the body (over 95 % of movement time).

It should be noted that the BBB scale is also widely used to assess the locomotor activity of the mice limbs after modeling other types of SCIs. For example, after contusion to the thoracic spinal cord (T8) of male and female ddY mice at the 1st week of the post-traumatic period, weak movements of the knee joint of the hindlimb were observed, corresponding to

7-8 scores by the BBB scale [39]. In its turn, the B scale was used to assess locomotor characteristics after modeling spinal cord concussion at the T10 level in C57BL/6J mice. In the 1st week after the injury modeling, the average score on the B scale was 1-1.5, which at the functional level was manifested in active movements of the knee joint of the hindlimb, and plantar posture of the foot without holding body balance. In the 2nd week after the modeling of the spinal cord contusion, a periodic plantar posture of the experimental animal feet was observed, which corresponded to 2-2.5 scores on the B scale [40].

The B scale was directly developed to assess the locomotor activity of IH after SCI in mice, as these laboratory animals are most often used to develop clinically relevant models of spinal cord injury of different genesis. Unlike the BBB scale, the B scale allows a more detailed assessment of the motor activity of the knee joint and the ipsilateral limb, which largely determines the overall locomotor activity of the limb, foot posture, synergy in limb movement, and body position.

Thus, in the 1st week after spinal cord hemisection of mice, the average score on the B scale was 0.79 ± 0.09 (**Fig. 2**), which was manifested in the absence, of weak or intense movements in the knee joint and plantar posture of IH. However, already starting from the 2nd and during the 3rd and 4th weeks of the post-traumatic period in most injured animals there was a significant increase in motor activity compared to the 1st week ($p < 0.001$) in the IH knee joint of animals, frequent or constant plantar foot posture without coordination of the hindlimbs, which corresponded to 1.22 ± 0.15, 1.48 ± 0.18 and 1.75 ± 0.22, respectively (**Fig. 2**).

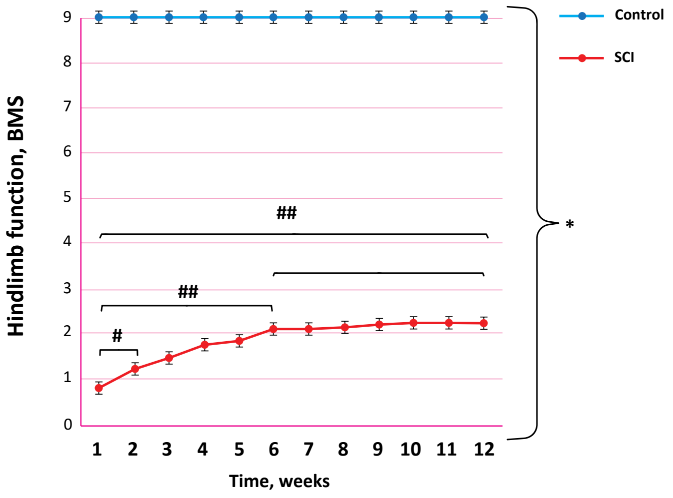

Fig. 2. The levels of locomotor activity of ipsilateral hindlimb in female FVB mice after spinal cord hemisection by the B scale.

Notes: * – $p < 0.001$ compared to the control, 1st-12th weeks; # – $p < 0.01$, ## – $p < 0.00001$ compared to the 1st week in the experimental group.

At the 5th week of the post-traumatic period, there were already active movements in the knee joint, frequent or constant plantar posture of the foot, slight coordination of the hindlimbs and their parallel posture – the average score on the B scale increased to 1.82 ± 0.22 compared to the 2nd week ($p < 0.00001$) (**Fig. 2**). At the 6th week, there was a further improvement in the IH locomotor activity compared with to 5th week (2.1 ± 0.26, $p < 0.004$), which was manifested by the active movements of the knee joint, plantar posture without or with temporary coordination of IH (**Fig. 2**).

However, already at the 7th and 8th week after SCI, female mice did not further improve IH locomotor activity in comparison with the 6th week - 2.11 ± 0.27 and 2.14 ± 0.28, respectively (**Fig. 2**). During the 9th and 10th weeks after SCI modeling, the mean score on the B scale was slightly higher – 2.21 ± 0.29 and 2.26 ± 0.30, respectively, compared to the 6th week (**Fig. 2**), which indicated an improvement in IH locomotor activity, and the detailed functional characteristics did not differ from those in the previous weeks of the post-traumatic period.

At the 11th and 12th week of follow-up, the mean score on the B scale also did not change: 2.25 ± 0.30 and 2.22 ± 0.31, respectively (**Fig. 2**), in this period of the postoperative period, as before, active work of the knee joint of the ipsilateral limb, frequent or constant plantar posture of the foot and coordinated parallel movements of IH were noted.

According to other authors, when modeling the spinal cord hemisection at the level of T10 on the 1st-3rd day of the post-traumatic period, the average score on the B scale was 0-1 points. Already from the 7th day of observation, the average score was 2-3 points, and for the period from the 14th to the 42nd day the average score on the B scale went to the plateau and reached 3.5-4.5 points. This study was performed on 10-15 week-old female *C57BL-6* mice [41].

In another study, female *C57BL/6* mice at the age of 6-8 weeks were used to model T8 spinal cord hemisection, after which experimental animals were injected with NgR agonist NEP1-40, which is known to promote the growth of corticospinal and serotonergic neurons of nervous tissue. The study found an improvement in locomotor function after the administration of NEP1-40 due to axonal sprouting. Thus, on the B scale in animals of the experimental group, the average score was 0-0.1 on the 1st day of the post-traumatic period. From the 7th day after injury modeling, the average score on the B scale gradually increased: 0.2-0.3 points on the 7th day, 0.5-0.7 points on the 14th day, 1.0-1.2 points on the 21st, 28th, and 35th days, and on the 42nd day, it increased to 1.4-1.6 points [42].

When in female *C57BL/6* mice, spinal cord hemisection at the C2 level was modeled with subsequent training, exercise has been shown to increase vascularization in all limb muscles for 6 weeks [43]. In order to study excitotoxicity in the nervous tissue due to neuronal destruction and glutamate release, spinal cord hemisection at the T12 level was modeled in 2-month-old *C57BL/6* mice, followed by training of animals 5 days a week for 3 months. Regular exercise reduces glutamate levels by 50 %, decreases axonal degeneration, scar, and transectional defect sizes as well as maintains axonal integrity, increases the number of synapses around motoneurons, and increases GAP-43 expression in neurons [44].

The spinal cord hemisection at the T10-11 level was also modeled in 9-10 week-old *C57BL/6* mice. The animals were trained during the 3rd, 6th, and 9th weeks after SCI. According to the results of the study, training helps to improve the plantar posture and active movements in the knee joint [45]. Moreover, training during the 3rd, 6th, and 9th weeks of the post-traumatic period leads to a reduction in muscle atrophy and improved locomotor function of the limbs [41]. It was also found that training for 3 weeks during the post-traumatic period does not affect the deep changes in the spinal neurons, although there is an increase in excitatory synapses [46].

The Ashworth scale was used to assess the spasticity level in mice IH. Thus, in the 1st week of the post-traumatic period, the average score was 3.63 ± 0.44, which at the functional level was found to increase muscle tone, passive movement difficult and some animals had rigid IH in flexion or extension (**Fig. 3**).

However, from the 2nd and 3rd week after SCI, compared with the 1st week ($p < 0.01$), there was a gradual decrease in the rate of IH spasticity – the average score was 3.35 ± 0.41, 3.2 ± 0.40, respectively, which corresponded to catch or significant violations of IH muscle tone, passive movements of the animal performed with some difficulty or freely (**Fig. 3**).

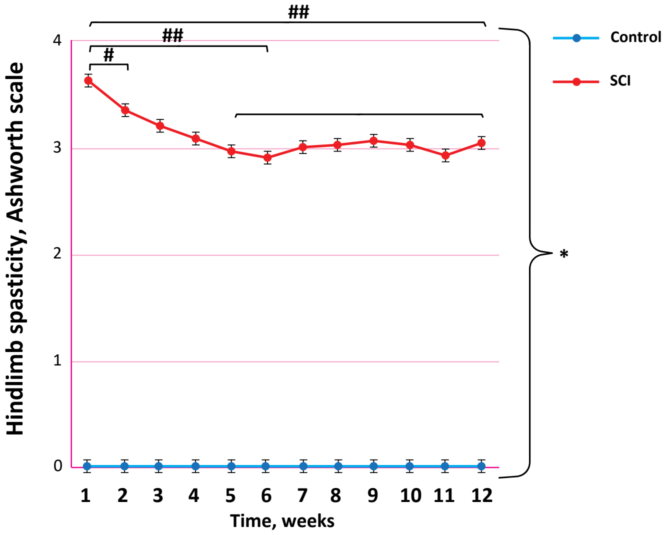

Fig. 3. The level of ipsilateral hindlimb spasticity in female FVB mice after spinal cord hemisection by the Ashworth scale
Notes: * – $p < 0.001$ compared to the control, 1st-12th weeks; # – $p < 0.01$, ## – $p < 0.00001$ compared to the 1st week in the experimental group.

At the 4th, 5th, and 6th weeks of the post-traumatic period in comparison with the 2nd week ($p < 0.01$) we continued to notice a slight decrease in the level of spasticity to 3.09 ± 0.38, 2.97 ± 0.36 and 2.91 ± 0.36, respectively, which at the functional level was characterized by increased muscle tone, free performance of passive movements or a slight increase in muscle tone, accompanied by catching, tension or relaxation with minimal resistance at the end of the movement (**Fig. 3**). Although at the 5th and 6th weeks there was no significant difference in the functional level compared to the 4th week. During the 7th, 8th, and 9th weeks after the hemisection modeling there also was not a significant change in the average spasticity in comparison with the 4th week: 3.0 ± 0.38, 3.02 ± 0.4 and 3.07 ± 0.39, respectively (**Fig. 3**). Minor fluctuations in this parameter were observed in the subsequent stages after spinal cord damage, namely at the 10th, 11th and 12th week: 3.03 ± 0.39, 2.93 ± 0.38 and 3.03 ± 0.39, respectively (**Fig. 3**).

In addition, the Ashworth scale is also used to establish the level of spasticity of the limb after modeling spinal cord contusion. Thus, in *C57BL/6N* mice after spinal cord contusion at the T9 level, the level of spasticity of the hindlimb was determined using the Ashworth scale and constituted 2.8 points [47].

It should also be noted that today there are several physiological mechanisms that determine the varying degrees of damage and the effectiveness of further spinal cord recovery in animals of different sexes. Thus, a significant role in the recovery of neural tissue after an injury is assigned to microglial cells, which are responsible for the local immune response of neural tissue and affect the formation of neural networks. In the transection model of SCI, it was found that in females, due to sex hormones that affect the activity of microglial cells, there is a more active recovery of the affected spinal cord tissue. The key hormone is considered to be the female sex hormone estrogen. Thus, estrogen promotes vascular relaxation in a NO-independent way by directly stimulating smooth muscle K-channels. Estrogen improves blood flow to the affected nervous tissue after injury by reducing the adhesion of leukocytes that occluded small vessels. Another possible neuroprotective mechanism of estrogen in female mice is due to its antioxidant properties, which in males leads to high levels of oxidative stress. Moreover, better recovery of nervous tissue in females may also be due to the effects of estrogen on anti-apoptotic proteins, in particular bcl-2, the level of which increases after SCI. *In vitro*, 17β-estradiol has been shown to reduce bcl-2 levels and enhance neuronal resistance to excitotoxicity [22].

After SCI modeling in female *FVB* mice in the 1st week, according to the BBB scale, low spontaneous motor activity and weak movements in one or two IH joints were observed. However, from the 3rd to the 6th week of the post-traumatic period, the scores on the BBB scale actively increased and remained almost at the same level until the 12th week. In particular, widespread movements in all IH joints, the frequent or constant plantar posture of the foot, coordination of the fore and hindlimbs, and holding body balance were observed (**Fig. 1**). On the B scale, there was no or weak motor activity of the IH knee on the 1st week of the post-traumatic period. However, from the 2nd to the 6th week the levels of IH locomotor activity dynamically improved and remained without significant changes until the 12th week (**Fig. 2**), which at the functional level corresponded to active movements of the knee joint, IH plantar posture, mostly synergistic work of posterior limbs and their parallel location during movement. According to the Ashworth scale, in the 1st week after the spinal cord hemisection modeling, significant violations of the IH muscle tone, difficulties in moving the animal's body, and performing passive movements were noted. Although, from the 2nd to the 4th week of the post-traumatic period, the level of IH spasticity gradually decreased and remained without significant changes until the 12th week (**Fig. 3**), which showed minor disorders of IH muscle tone, ease of movement, and catching, however, it remained at a high level throughout the study period.

CONCLUSION

In female FVB mice, after SCI modeling, locomotor activity was improved from the 3rd week (by the BBB scale) and from the 2nd week (by the B scale) and remained unchanged from the 6th week until the 12th week. In general, such levels of IH locomotor activity correspond to the phase of early recovery. The levels of IH spasticity were high during the whole research period, although from the 2nd to the 4th week there was a minor, however, significant decrease in this score in comparison with the 1st week of the posttraumatic period. From the 4th week to the 12th week no significant changes in spasticity were observed.

The results of this study can be used to further elucidate the features of post-traumatic physiological processes in spinal cord nervous tissue, as well as in the development and testing of new therapeutic approaches to relieving spinal cord injuries – the use of matrices of different types and stem cells of different origin or their combination.

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Показники локомоторної активності та спастичності кінцівки у самиць мишей з моделлю травми спинного мозку

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РЕЗЮМЕ

Травми спинного мозку призводять до масштабних фізичних, фізіологічних, психологічних та професійних втрат. Саме тому одним із актуальних на сьогодні завдань нейрофізіології є дослідження наслідків спінальної травми. Для комплексної та детальної оцінки функціонального посттравматичного відновлення спинного мозку розробляють різні варіанти моделі його половинного перетину, який є найбільш поширеним типом ураження.

***МЕТА ДОСЛІДЖЕННЯ**: проаналізувати рівень локомоторної активності та зміну показників спастичності задньої іпсилатеральної кінцівки (ЗІК) миші після моделювання половинного перетину спинного мозку.*

***МАТЕРІАЛИ ТА МЕТОДИ**: Травму лівобічного перетину половини спинного мозку моделювали на рівні нижніх грудних сегментів (T10-T11) у самиць мишей лінії FVB. За шкалами Basso-Beattie-Bresnahan (BBB), Basso (B) та шкалою Ashworth щотижня визначали локомоторну активність та спастичність ЗІК тварин протягом 1-12 тижнів посттравматичного періоду.*

***РЕЗУЛЬТАТИ**. В порівнянні з першими тижнями посттравматичного періоду, на більш пізні терміни (11-12 тижні) спостерігали помітне відновлення функції ЗІК: 4,39 ± 0,61 бали з 21 можливого за шкалою BBB та 2,22 ± 0,31 бали з 9 можливих за шкалою B. Проте, на всіх досліджуваних часових проміжках після травмування спинного мозку відзначали стабільно високий рівень спастичності кінцівки дослідних тварин, зокрема, на 12-й тиждень показник за шкалою Ashworth становив 3,03 ± 0,39 з 4 можливих.*

***ВИСНОВКИ**. Після моделювання травми спинного мозку методом половинного перетину спостерігали спонтанне відновлення локомоторної активності вже з 2-го тижня з показниками, які відповідали фазі раннього відновлення. В свою чергу, показники спастичності ЗІК миші були досить високими протягом всього періоду дослідження, хоча вже на 2-й тиждень спостерігали незначне зниження цього показника в порівнянні з 1-м тижнем посттравматичного періоду.*

***КЛЮЧОВІ СЛОВА**: травма спинного мозку; моделювання перетину половини спинного мозку; локомоторна активність; спастичність; шкала BBB; шкала B*