Batroxobin protects against spinal cord injury in rats by promoting the expression of vascular endothelial growth factor to reduce apoptosis

HUI YU, BIN LIN, YONGZHI HE, WENBIN ZHANG and YANG XU

Department of Orthopedics, The 175th Hospital of the PLA, Southeast Hospital of Xiamen University, Zhangzhou, Fujian 363000, P.R. China

Received July 5, 2014; Accepted March 3, 2015

DOI: 10.3892/etm.2015.2368

Abstract. The host response to spinal cord injury (SCI) can lead to an ischemic environment that can induce cell death. Therapeutic interventions using neurotrophic factors have focused on the prevention of such reactions in order to reduce this cell death. Vascular endothelial growth factor (VEGF) is a potent angiogenic and vascular permeability factor. We hypothesized in this study that batroxobin would exhibit protective effects following SCI by promoting the expression of VEGF to reduce the levels of apoptosis in a rat model of SCI. Ninety adult female Sprague Dawley rats were divided randomly into sham injury (group I), SCI (group II) and batroxobin treatment (group III) groups. The Basso-Bettie-Bresnahan (BBB) scores, number of apoptotic cells and expression of VEGF were assessed at 1, 3, 5, 7, 14 and 28 days post-injury. The BBB scores were significantly improved in group III compared with those in group II between days 5 and 28 post-injury (P<0.05). At each time-point subsequent to the injury, the number of apoptotic cells in group III was reduced compared with that in group II. Compared with group II, treatment with batroxobin significantly increased the expression of VEGF from day 3 until 2 weeks post-SCI (P<0.05), while no significant difference was observed at day 28. These data suggest that batroxobin has multiple beneficial effects on SCI, indicating a potential clinical application.

Introduction

Spinal cord injury (SCI) predominantly affects younger members of the population and is caused by traffic or sports-related accidents. The condition can result in severe neurological deficits, such as para- and quadriplegia (1). Since traumatic injury to the mammalian spinal cord exhibits a highly dynamic nature, characterized by a complex pattern of insidious, destructive biochemical and pathophysiological events, the potential for functional recovery from the condition is limited (2). Following SCI, substantial secondary damage within the tissue is caused by increased vascular permeability, infiltration of inflammatory cells and subsequent focal edema, which may induce apoptosis (3-5). Following SCI, cells at the site of injury may undergo cell death through post-traumatic necrosis or apoptosis, the latter of which can be demonstrated by nuclear DNA fragmentation and caspase activation. Apoptosis, in particular, is a prominent event in the spinal cord subsequent to SCI (6). Apoptosis has been shown to occur widely in the white matter, concurrently with Wallerian degeneration, and to affect neurons and oligodendrocytes. The apoptotic cell death of both neurons and oligodendrocytes may therefore be a causative factor contributing to the paralysis of patients with SCI (7,8). Therapeutic interventions using neurotrophic factors have focused on the prevention of such reactions to reduce cell death and promote tissue regeneration (9).

Vascular endothelial growth factor (VEGF) has long been known as a potent angiogenic factor that stimulates the proliferation and migration of endothelial cells and the in vivo formation of new blood vessels (10). The association of VEGF with the central nervous system (CNS) has been predominantly studied in models of ischemic stroke or brain tumor (11); however, VEGF has also been attracting attention as a neuroprotective and neurotrophic factor involved in nerve regeneration and the promotion of functional recovery (12,13). It has been found that VEGF enhances neurite outgrowth and neuroprotection, and reduces post-traumatic apoptosis following CNS injury (14-16). VEGF is crucial in a number of processes in the CNS, including vascularization, neuronal proliferation and the growth of coordinated vascular and neuronal networks (17). Accordingly, enhancing the expression of VEGF may have therapeutic potential for the treatment of SCI.

Batroxobin is a thrombin-like serine protease from the venom of the snake Bothrops moojeni that can decrease blood fibrinogen levels and promote blood flow (18). Batroxobin has been widely used clinically in various ischemic disorders, such as stroke, deep-vein thrombosis, myocardial infarction and

Correspondence to: Dr Bin Lin, Department of Orthopedics, The 175th Hospital of the PLA, Southeast Hospital of Xiamen University, 269 Zhanghua Road, Zhangzhou, Fujian 363000, P.R. China
E-mail: linbin813@163.com

Key words: batroxobin, vascular endothelial growth factor, apoptosis, spinal cord injury
were captured using a
registered trademark,
respectively, 1% acid
samples were
sections were
sections
sections
sections
sections
movement, weight support and stepping
rating scale
The rats were tested for locomotor deficits at 1 day before and
Basso-Beattie-Bresnahan (BBB) evaluation of locomotion.

Materials and methods

This protocol was evaluated and approved by the Governmental
Animal Care Committee of the Medical College of Xiamen
University (Zhangzhou, China) and was performed according
to the National Institutes of Health guidelines on the ethical
use of animals. Every effort was made to minimize animal
suffering and to reduce the number of animals used.

Animals and surgical procedures. Ninety adult female Sprague
Dawley rats (Experimental Animal Center of Xiamen University)
weighing 280-300 g were randomly assigned to the following
groups: Sham injury (group I, n=30), SCI (group II, n=30) and
batroxobin treatment (group III, n=30). Any animals that
died during the experiment were not included. Batroxobin was
obtained from TuKang Bio-Pharmaceutical, Inc. (Shenyang,
China). Prior to surgery, the animals were anesthetized by intra-
peritoneal injection of 400 mg/kg chloral hydrate (Beyotime
Institute of Biotechnology, Haimen, China). During the surgery,
the rats were placed in a prone position on a warming pad to
maintain a body temperature of 37.0±0.5°C. Upon completion
of the surgery, the rats were housed in individual cages with
access to food and water ad libitum, and administered an intra-
muscular injection of 200,000 U/day penicillin (175th Hospital
of the PLA, Zhangzhou, China) for 3 days.

All rats were injured at the thoracic level 12 (T12), using
an established weight-drop model described in a previous
study (22). Briefly, the skin and muscle overlying the spinal
column were incised and a laminectomy was performed at T12,
leaving the dura intact. A moderate-intensity weight-drop (10 g,
7.0 cm) was performed using an impactor with a diameter of
2.5 mm (Xiamen University) onto the exposed T12 cord. The
ratable group I were treated in an identical manner to the rats
in group I were treated in an identical manner to the rats
subjected to SCI with the omission of the weight-drop step.

Following the surgery, the bladders of the rats were
published twice daily until spontaneous voiding
occurred. The dosage of batroxobin (DF-521; Beijing Tobishi
Pharmaceutical Co., Ltd., Beijing, China) was selected
according to the manufacturer's instructions, which recom-
ended 10 batroxobin units (BU) as the regular initial dose
and 5 BU as the maintenance dose. Considering the differ-
ences between humans and rats, the rats in group III were
injected with batroxobin at a dosage of 5 BU/kg/day via the
tail vein within 8 h of SCI until 3 days post-injury. Instead of
batroxobin, the rats in groups I and II were administered saline
through pumps as a control treatment.

Basso-Beattie-Bresnahan (BBB) evaluation of locomotion.
The rats were tested for locomotor deficits at 1 day before and
1, 4 and 7 days after SCI with a standard open-field locomotor
test, developed by Basso et al (23). This BBB locomotor
rating scale evaluates the following criteria: Extent of joint
movement, weight support and stepping/walking behavior
of the hindlimbs. The rating scale ranges from 0 (no observ-
able hindlimb movement) to 21 (normal locomotion), and
scores were assigned for both hind limbs by two independent
observers blinded to the experiments. The main functional
outcome was calculated by the mean value.

Hematoxylin and eosin (HE) staining for the detection of patho-
logical changes. For the histological staining, 5-µm transverse
sections of injured spinal cord tissue from each group at 1, 3, 5, 7,
14 and 28 days post-injury were deparaffinized and placed into
fresh xylene for 15 min twice. The sections were re-hydrated in
100% alcohol for 5 min twice, and then 95 and 70% alcohol
once for 3 min, respectively. The sections were subsequently
washed briefly in double-distilled (dd)H2O and stained in Harris
hematoxylin (Beyotime Institute of Biotechnology) solution for
5 min. Following staining, the sections were washed in running
tap water for 8 min, subjected to differentiation with 1% acetic
alcohol for 30 sec and blued in 0.2% ammonia water for 30 sec.
The sections were then washed in running tap water for a further
5 min and rinsed in 95% alcohol for ~15 dips. The sections
were stained in Eosin-Phloxine solution (Beyotime Institute
of Biotechnology) for 1 min, prior to undergoing 95 and 100%
alcohol dehydration (5 min each) and clearing in two changes
of xylene (5 min each). Finally, the sections were mounted with
mounting medium (Beyotime Institute of Biotechnology). The
images were captured using an FV300 confocal microscope
(Olympus Corp., Tokyo, Japan).

Terminal deoxynucleotidyl-transferase-mediated dUTP nick
end labeling (TUNEL) test for apoptosis. For the detection of
apoptosis, TUNEL staining was performed using a TUNEL
detection kit according to the manufacturer's instructions
(ApopTag® horseradish peroxidase kit; DBA, Milan, Italy).
Briefly, sections of SCI tissue at 1, 3, 5, 7, 14 and 28 days
post-injury were immersed in xylene for 5 min twice at
room temperature, and in 100, 90, 80 and 70% ethanol for
5 min twice. The sections were then incubated in 15 µg/ml
Proteinase K solution for 20 min at room temperature and
washed with phosphate-buffered saline (PBS). Hydrogen
peroxide (3%), applied for 5 min at room temperature,
was utilized to terminate any endogenous peroxidase activity,
prior to the sections being washed with PBS. The sections
were then immersed in terminal deoxynucleotidyl transferase
(TdT) buffer containing TdT and biotinylated dUTP, incubated in a
humid atmosphere at 37°C for 90 min, and washed with PBS.
Subsequent to being washed, the sections were incubated at
room temperature for a further 30 min with anti-horseradish
peroxidase-conjugated antibody (GeneTex, San Antonio,
Texas, USA), and 3,3’-diaminobenzidine was used to visualize
the signals. The sections were then washed in ddH2O and
mounted. Images were captured using an FV300 confocal
microscope (Olympus Corp.).

Immunohistochemistry of VEGF. For immunohistochemical
staining, each specimen was embedded in paraffin and a
microtome was used to cut serial sections. VEGF immuno-
histochemical staining was performed using an avidin-biotin
peroxidase complex technique and a Histostain® SP kit
(Maixin-Bio, Inc., Fuzhou, China) in accordance with the
manufacturer's instructions. Mouse monoclonal antibody
Table I. BBB score of each group at different time-points.

<table>
<thead>
<tr>
<th>Days post-SCI</th>
<th>Group I (score)</th>
<th>Group II (score)</th>
<th>Group III (score)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>21.00±0.00</td>
<td>21.00±0.00</td>
<td>21.00±0.00</td>
</tr>
<tr>
<td>1</td>
<td>21.00±0.00</td>
<td>0.69±0.24</td>
<td>0.79±0.23</td>
</tr>
<tr>
<td>3</td>
<td>21.00±0.00</td>
<td>2.36±0.30</td>
<td>2.53±0.38</td>
</tr>
<tr>
<td>5</td>
<td>21.00±0.00</td>
<td>4.82±0.31</td>
<td>5.03±0.33</td>
</tr>
<tr>
<td>7</td>
<td>21.00±0.00</td>
<td>6.62±0.40</td>
<td>7.55±0.37</td>
</tr>
<tr>
<td>14</td>
<td>21.00±0.00</td>
<td>7.54±0.42</td>
<td>9.65±0.44</td>
</tr>
<tr>
<td>28</td>
<td>21.00±0.00</td>
<td>10.22±0.74</td>
<td>13.74±0.66</td>
</tr>
</tbody>
</table>

Compared with group II, *P<0.01 and †P<0.05. Results are presented as the mean ± standard deviation. BBB, Basso-Beattie-Bresnahan; Group I, sham injury; Group II, SCI; Group III, SCI with batroxobin treatment; SCI, spinal cord injury.

against VEGF (1:200; sc-30343; Santa Cruz Biotechnology, Inc., Santa Cruz, CA, USA) and rabbit polyclonal antibody against VEGF (1:100; sc-33547; Santa Cruz Biotechnology, Inc.) were used for this study. Two pathologists who were unaware of the experimental data were responsible for counting the number of VEGF-positive cells in 10 high-power fields (magnification, x400) in each specimen. The average number of VEGF-positive cells per specimen was then calculated. Images were captured using an FV300 confocal microscope (Olympus Corp.).

Statistical analysis. Statistical analysis was performed using SPSS version 13.0 for Windows (SPSS, Inc., Chicago, IL, USA). Data are presented as the mean ± standard deviation. The Mann-Whitney U-test and Spearman’s rank correlation were used for the statistical analyses. P<0.05 was considered to indicate a statistically significant difference.

Results

During surgery, the rectal temperature of the rats was maintained at 37±0.5°C. The mean body weight of the rats in the sham surgery group was 371.8±9.7 g (range, 362-385 g), while the mean body weights of the control and batroxobin group rats were 375.9±7.1 g (range, 367-388 g) and 369.0±11.3 g (range, 354-389 g), respectively. No significant differences in these physiological parameters existed between the groups.

Behavioral test. To evaluate the extent of motor function recovery, the BBB locomotor rating scale was used. The BBB scores were assessed for the three groups at different time-points following SCI. Table I shows the mean BBB scores of the rats in the three groups over the time-course of the experiment. Prior to surgery, the rats were all healthy (BBB score, 21±0.00; data not shown). In the group I rats, no significant difference was observed in the hind limb movement scores measured prior to and following SCI, and the rats exhibited normal movement throughout the observation period (BBB score, 21 points). In groups II and III, the rats showed improvements in motor function at day 5 post-SCI compared with the scores on the date of the SCI; however, the average BBB score was significantly higher in the group III rats than that in the group II rats between days 5 and 28 post-SCI. On day 28, the BBB score of the group III rats was 13.74±0.66 points, whereas the group II rats scored 10.22±0.74 points (P<0.05). A BBB score of 14 is indicative of consistent weight-supported plantar steps and front-hind limb coordination (23); neither of the scores in groups II or III exceeded the 14-point threshold (mean in group III = 13.74).

Histological assessments

Visual study. Following the initial injury, tissue edema appeared immediately in the dorsal region of the spinal cord in groups II and III, while no fresh bleeding spots were observed at day 3. Scar formation was observed in the region of the lesion and conglutination with the endorhachis was apparent at days 14 and 28 post-SCI. In addition, the spinal cord was atrophic with a reduction in the diameter. In group I, areas of scar formation were observed in the region of the lesion and conglutination with the endorhachis was apparent at days 14 and 28 post-SCI. This may have been a result of the trauma of the surgery; however, no obvious edema in the spinal cord was observed and the posterior central blood vessel and the structure of the spinal cord were clearly visible.

H&E staining. At 1 day post-SCI, H&E staining in the group II rats showed a large area of structural damage, multifocal hemorrhage and inflammatory cell infiltration. Notably, neuron pyknosis and chromatin condensation could be observed, which indicated cell apoptosis (Fig. 1). At 14 days post-SCI, H&E staining in the group II rats showed a small hemorrhagic focus in the gray and white matter of the spinal cord; evident destruction to the structure of the spinal cord was observed, and neurons were found to be dissolved and liquefied in the gray matter (Fig. 2). The resulting large, liquefied and necrotic area formed a cystic space. Numerous swollen axons and neovascularization were additionally observed in the white matter, and nerve fiber disorganization was apparent. Twenty-eight days after SCI, the hemorrhagic focus in the gray and white matter was almost entirely absorbed, and further destruction of the spinal cord was observed; the neurons that were dissolved and liquefied in the gray matter formed numerous vacuolar structures. Furthermore, a reduction in the inflammatory cell infiltration, and newborn disordered blood-vessels were observed (Fig. 3). Fourteen days after the SCI, H&E staining in group III revealed damage to the structure of the spinal cord, in addition to inflammatory cell infiltration, neuron dehydration and disintegration,
hyperplastic and hypertrophic gliocytes and the formation of cystic spaces; however, the damage was less severe and widespread compared with that in group II. Furthermore, fewer apoptotic cells were observed in group III than in group II. At 28 days post-SCI, it was observed that the inflammatory cell infiltration in the group III rats was reduced, and fewer apoptotic cells were present compared with the group II rats (Fig. 4). In summary, the spinal cord pathological changes that occurred following injury were significantly attenuated by batroxobin on the 5th, 7th, 14th and 28th days postoperatively.

**Effect of batroxobin on cellular apoptosis in the spinal cord.** SCI-induced cellular apoptosis could be detected using the TUNEL test. As shown in Fig. 5, apoptotic cells were barely detectable in group I at 1 day after SCI, as little apoptosis occurred in the absence of injury. In group II, an increased number of apoptotic cell bodies (indicated by arrows) were found at 1 day after SCI (Fig. 6), and this number continued to remain high from day 3 to day 5 (Fig. 7), prior to tapering until day 28. Compared with group II, however, a significant reduction in the number of apoptotic cell bodies (indicated by arrows) was detected in group III at 1 day after SCI (Fig. 8), and this number remained at a lower level from day 3 to day 5 (Fig. 9). Following treatment with batroxobin, the number of apoptotic cells was found to decrease significantly. This indicated that batroxobin inhibited cellular apoptosis subsequent to injury. The number of apoptotic cells in the field of view on the slides in each group was counted under a microscope and analyzed. The result of the TUNEL test indicated that the severity of tissue damage and neuronal loss was considerably milder in group III than that in group II (Table II).
Effect of batroxobin on VEGF expression in the spinal cord.
High-level constitutive expression of VEGF was observed in groups II and III; however, immunohistochemical study of VEGF in the spinal cord sections showed significant differences between the two groups. Compared with group II, batroxobin promoted the expression of VEGF between days 1 (Figs. 10 and 11) and 14 after injury. It was noted that the mean number of VEGF-positive cells per section was maximized at day 3 in groups II and III (Figs. 12 and 13). This indicated that batroxobin could promote the expression of VEGF, which played a central role in inducing angiogenesis (Table III).

Correlation of VEGF expression with the number of apoptotic cells. A significant correlation was found between the degree of VEGF expression and the number of apoptotic cells following injury ($r=-0.90052$, $P<0.05$). These data suggest...
that batroxobin may exert protective effects by promoting the expression of VEGF in order to reduce apoptosis in SCI in rats (Fig. 14).

Discussion

SCI is a serious and common CNS trauma, leading to irreversible damage to the sensory and motor functions. At present, treatment strategies for patients with SCI have been focusing increasingly on the surgical stabilization of the initial injury to prevent further loss of neurological function, without much attention being paid to nerve cell protection and a reduction of apoptosis in SCI in rats.

Table II. TUNEL test for apoptosis detection.

<table>
<thead>
<tr>
<th>Days post-SCI</th>
<th>Group I</th>
<th>Group II</th>
<th>Group III</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>15.30±0.32</td>
<td>65.48±4.03</td>
<td>63.65±1.58&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>3</td>
<td>15.08±0.63</td>
<td>49.14±2.86</td>
<td>39.64±1.33&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>5</td>
<td>15.03±0.40</td>
<td>32.80±2.34</td>
<td>28.53±1.29&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>7</td>
<td>15.11±0.42</td>
<td>25.04±0.83</td>
<td>23.05±0.62&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>14</td>
<td>15.08±0.42</td>
<td>21.92±0.65</td>
<td>21.53±0.47&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>28</td>
<td>15.13±0.59</td>
<td>21.39±0.59</td>
<td>20.98±0.35&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Compared with group II, <sup>a</sup>P<0.01 and <sup>b</sup>P<0.05. Results are presented as the mean ± standard deviation. TUNEL, terminal deoxynucleotidyl-transferase-mediated dUTP nick end labeling; Group I, sham injury; Group II, SCI; Group III, SCI with batroxobin treatment; SCI, spinal cord injury.

Table III. Number of VEGF-positive cells per section.

<table>
<thead>
<tr>
<th>Days post-SCI</th>
<th>Group I</th>
<th>Group II</th>
<th>Group III</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>3.66±0.03</td>
<td>53.89±1.87</td>
<td>55.02±1.44&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>3</td>
<td>3.66±0.01</td>
<td>61.35±1.89</td>
<td>69.63±4.69&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>5</td>
<td>3.66±0.04</td>
<td>67.11±3.03</td>
<td>79.10±4.61&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>7</td>
<td>3.66±0.08</td>
<td>59.75±1.30</td>
<td>61.37±2.90&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>14</td>
<td>3.66±0.01</td>
<td>30.51±0.85</td>
<td>40.50±1.97&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>28</td>
<td>3.66±0.02</td>
<td>20.08±0.35</td>
<td>20.41±0.72</td>
</tr>
</tbody>
</table>

Compared with group II, <sup>a</sup>P<0.01 and <sup>b</sup>P<0.05. Results are presented as the mean ± standard deviation. Group I, sham injury; Group II, SCI; Group III, SCI with batroxobin treatment; SCI, spinal cord injury; VEGF, vascular endothelial growth factor.

Figure 12. Three days after SCI, the expression of vascular endothelial growth factor (indicated by arrows) was maximized in the SCI group (magnification, x400). SCI, spinal cord injury.

Figure 13. Three days after SCI, the expression of VEGF (indicated by arrows) was maximized in the batroxobin-treated group. Compared with the SCI group, batroxobin significantly promoted the expression of VEGF (magnification, x400). SCI, spinal cord injury; VEGF, vascular endothelial growth factor.

Figure 14. A significant correlation was found between the degree of VEGF expression and the number of apoptotic cells following injury (r=-0.90052, P<0.05). VEGF, vascular endothelial growth factor.
cell death, as has been a focus for the treatment of stroke (24).

In a previous study, the potential of the nervous system to adapt to SCI from a functional (neuronal plasticity) and a structural (neuronal remodeling) perspective was demonstrated (25). However, following the primary SCI, a secondary injury expands continuously for ~4 weeks; understanding the mechanism and finding measures to control this secondary injury are of great importance. Following SCI, the response of the host can generate an ischemic environment that can lead to cell death. Furthermore, this ischemic environment limits cell transplantation approaches that could be used to promote spinal cord regeneration (26). It has been widely accepted that apoptosis is the most common form of cell death following SCI. The number of apoptotic cells is dependent on a number of factors, including external stimulation, injury severity, secondary edema and ischemia (27). By focusing on the regulation of apoptosis subsequent to SCI it has been found that the inhibition of this apoptosis could effectively protect the nerve cells (28).

A previous study has shown that enhancing spinal cord blood circulation reduces the secondary injury (29). The delivery of angiogenic factors, such as VEGF, from poly(lactide-co-glycolide) scaffolds formed by the gas foaming process can induce a local increase in blood vessel formation (30,31). VEGF signals are considered to act as neurotrophic factors (10,13,32). Previous studies have demonstrated the direct neurotrophic effects of VEGF on peripheral nerves (33) and reported increased neuron density and viability in mesencephalic explant cultures treated with VEGF (34). Successful neurotrophic or neuroprotective and tissue-sparing effects have also been observed following VEGF treatment in traumatic SCI (35,36). Tissue edema is one of the main causes of secondary damage subsequent to SCI (37,38). The application of VEGF following SCI can decrease vascular permeability and tissue edema in the spinal cord, and alleviate the deterioration of functional recovery (39,40).

The aim of the present study was to examine the effect of batroxobin, a drug widely used in various ischemic disorders (19,20), in reducing the secondary damage following SCI. One of the concerns for batroxobin administration in SCI is the possibility of inducing bleeding in the injured cord. It has been reported that a downstream product of batroxobin, fibrinopeptide-A, forms an unstable clot and even shortens the bleeding time in vivo (18,41). In the present study, batroxobin effectively increased the expression of VEGF and reduced the number of apoptotic cells, which suggests that the batroxobin has a positive effect on the secondary damage following SCI, promoting neuronal survival and improving locomotor recovery.

In conclusion, this study has underlined the potential of batroxobin for improving the functional outcome following SCI. Since batroxobin is clinically widely used, its beneficial effect in reducing SCI can be utilized in therapeutic strategies; however, future studies are required to detail the mechanisms underlying the batroxobin-induced decrease in apoptosis and the promotion of functional recovery following SCI.

References