Therapeutic Approaches Targeting Vascular Repair After Experimental Spinal Cord Injury: A Systematic Review of the Literature

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Traumatic spinal cord injury (SCI) disrupts the spinal cord vasculature resulting in ischemia, amplification of the secondary injury cascade and exacerbation of neural tissue loss. Restoring functional integrity of the microvasculature to prevent neural loss and to promote neural repair is an important challenge and opportunity in SCI research. Herein, we summarize the course of vascular injury and repair following SCI and give a comprehensive overview of current experimental therapeutic approaches targeting spinal cord microvasculature to diminish ischemia and thereby facilitate neural repair and regeneration. A systematic review of the published literature on therapeutic approaches to promote vascular repair after experimental SCI was performed using PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) standards. The MEDLINE databases PubMed, Embase, and OVID MEDLINE were searched using the keywords “spinal cord injury,” “angiogenesis,” “angiogenesis inducing agents,” “tissue engineering,” and “rodent subjects.” A total of 111 studies were identified through the search. Five main therapeutic approaches to diminish hypoxia-ischemia and promote vascular repair were identified as (1) the application of angiogenic factors, (2) genetic engineering, (3) physical stimulation, (4) cell transplantation, and (5) biomaterials carrying various factor delivery. There are different therapeutic approaches with the potential to diminish hypoxia-ischemia and promote vascular repair after experimental SCI. Of note, combinatorial approaches using implanted biomaterials and angiogenic factor delivery appear promising for clinical translation.

Keywords: Spinal cord injury, Blood-spinal cord barrier, Vascular injury, Spinal cord regeneration, Biocompatible materials, Therapeutics

INTRODUCTION

Traumatic spinal cord injury (SCI) is one of the leading causes of disability and a major burden for healthcare systems. Current therapeutic strategies are limited to acute medical and surgical management and rehabilitation, as there are no therapies available to promote spinal cord regeneration.1

SCI results from a primary mechanical injury which is amplified by a secondary injury cascade to result in loss or disruption of neural elements and vascular structures. The traumatic vascular disruption in the injured spinal cord plays a key role in modulating secondary injury development by promoting local blood-spinal cord barrier (BSCB) breakdown, inflammation, and neuronal cell death as well as creating a hypoxic-ischemic
environment which can spread to initially intact regions. The characterization of endogenous vascular regeneration following SCI-induced vascular injury has been the goal of some recent experimental studies, conducted mostly in rodents. Results show that vascular regeneration occurs spontaneously after SCI, following a definite time schedule – this regeneration process is however deficient and, due to an increased BSCB-permeability, even potentially detrimental to preserved neural tissue. But even if this new vasculature is only partially functional, it provides guidance for regenerating axons and is thereby crucial for neural regeneration. To find a way to improve vascular repair after SCI and restore a functional blood supply, thereby supporting neural regeneration, is one of the major challenges in experimental SCI research.

A multitude of experimental therapeutic approaches have been applied to restore functional vascularization, consisting of (1) the application of angiogenic factors, (2) genetic engineering therapy, (3) physical stimulation, (4) cell transplantation, and (5) biomaterials carrying various factor delivery.

The aim of this systematic review is to rigorously summarize and evaluate the present state of research in this emerging field. We first outline a focused overview of SCI pathophysiology with an emphasis on vascular injury and endogenous vascular repair and then shift to an examination of current experimental therapeutic approaches to promote vascular repair with the prospect of future translation of innovations into clinical patient care.

MATERIALS AND METHODS

1. Approach to the Systematic Literature Review

We performed a systematic, qualitative review of the literature, by screening the established MEDLINE databases PubMed, Embase, and OVID MEDLINE. The main search terms included “spinal cord injury,” angiogenesis,” “angiogenesis inducing agents,” and “tissue engineering,” in combination with “rodent subjects.” The search included no time limit. Articles in English were included, meeting the following criteria: experimental research, original studies, rodent subjects, therapeutic intervention, and focus on vascular injury and repair. The systematic literature search was conducted by one researcher from June 2021 to July 2021 and from May 2022 to June 2022. Data analysis was conducted according to the PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) guidelines. A total of 614 articles were selected for review, of which 475 were excluded after screening as a result of not meeting the inclusion criteria. An additional 30 articles were excluded due to not meeting the inclusion criteria after full-text assessment for eligibility. For qualitative synthesis 111 articles were assessed in detail and included in this publication (Fig. 1).

2. Relevant Experimental Therapeutic Approaches According to the Literature Search

According to the systematic literature search, 5 main categories of therapeutic approaches to promote vascular repair in experimental SCI research were identified: (1) angiogenic factors, (2) genetic engineering, (3) physical stimulation, (4) cell transplantation, and (5) biomaterials carrying various factor delivery (Table 1).

RESULTS

1. PART 1: Pathophysiology of Traumatic SCI With Emphasis on Vascular Injury And Repair

1) Pathophysiology and its progression following SCI

SCI is characterized as a sudden mechanical force impacting the spinal cord, e.g., following a fall, vehicle accident, or sports-related injury. It is commonly comprised of both contusion and compression injury causing direct cell death, axonal shearing, and disruption of the microvasculature with traumatic bleeding. This primary injury is not accessible to treatment and is immediately followed by a sustained secondary injury cascade. This secondary injury spreads to formerly unaffected regions, leading to further spinal cord damage with the consequences of demyelination and axonal dieback. The developing secondary injury can be divided into acute (first 48 hours), subacute (48 hours–14 days), intermediate (14 days to 6 months) and chronic phases (> 6 months).

2) Time course of BSCB disruption and restitution following SCI

The BSCB protects the spinal cord, as part of the central nervous system (CNS), from the periphery and maintains a homeostatic environment. The BSCB, formed by endothelial cells (ECs), astrocytic foot processes and pericytes as well as a basal lamina together with adjoining neurons comprise the neurovascular unit (NVU) (Fig. 2A). The unique cell-cell-contacts between ECs containing CNS-specific tight junctions (TJs) provide an immune privilege to the spinal cord compared to the periphery, restricting the passage of neurotoxic molecules and inflammatory cells.

Following the primary injury, a localized direct disruption of the NVU damages the microvasculature directly surrounding...
the injury site: Disruption of the protective cell-cell-contacts leads to altered cell permeabilization and consequent oedema formation, causing increased apoptotic and necrotic cell death.\textsuperscript{14,16,17} (Fig. 2B). These permeability changes not only lead to electrolyte influx and oedema, but to an influx of inflammatory cells (lymphocytes, macrophages, and neutrophils), as well as cytokines (tumor necrosis factor-\textalpha, interleukin [IL]-1\textbeta, IL-5) and vasoactive peptides. This immune response, commencing within the first minutes postinjury, leads to further oedema formation and thus tissue compression, resulting in further injury development spreading farther from the primary injury site.\textsuperscript{2,15,18}

These events further destabilize the BSCB and lead to a release of reactive oxygen species, resulting in the dysfunction of the NVU in formerly unharmed regions.\textsuperscript{2,14} Correlating with these mechanisms, the first peak in BSCB-permeability in the penumbra zone is reached 24-hour postinjury, followed by a smaller peak after 3–5 days.\textsuperscript{4,5}

Most of the BSCB function is believed to be reinstated after 14 days\textsuperscript{4,19} (Fig. 2C). However, experimental studies show that the permeability for small molecules is increased up to the chronic phase of SCI, for as long as 56-day postinjury.\textsuperscript{6}

3) Endogenous revascularization after SCI

The major form of vascular regeneration in the spinal cord is known to be angiogenesis, with a smaller part displayed by postnatal vasculogenesis. Angiogenesis is defined by the formation of blood vessels out of pre-existing vessels, whilst vasculogenesis describes the formation of new vessels.\textsuperscript{20} The hypoxic milieu after SCI results in the release of angiogenic factors from the injury site. Endogenous revascularization following SCI is
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Fig. 2. (A–C) Pathophysiology of spinal cord injury and the neurovascular unit in health and its disruption. SC, spinal cord; BSCB, blood – spinal cord barrier; EC, endothelial cells; AC, astrocytic foot processes; PC, pericyte; BL, basal lamina; TJ, tight junction; ROS, reactive oxygen species; h, hours; d, days; w, weeks; m, months.
expected to occur during the first week postinjury, and to disappear shortly afterwards. Importantly, the second, smaller peak in BSCB-permeability correlates with the peak of endogenous revascularization after SCI. As new vessels form due to angiogenesis or vasculogenesis, this young vasculature does not necessarily possess a functional BSCB. As some parts of this newly formed vasculature gain a functional barrier and promote a functional NVU during its genesis, other parts never do. This results in the increased extravasation of molecules and fluids causing edematous swelling, inflammation, and necrotic and apoptotic cell death and thus exacerbating the secondary injury \(^2\,^3\,^5\,^25\) (Fig. 2B).

4) Regenerative potential facilitated by vascular repair after SCI

As the spinal cord remains in a state of chronic hypoxia and ischemia postinjury, reinstatement of a steady tissue perfusion would be desirable. This was shown by previous research indicating that neovascularization is crucial for axon regeneration, as these were shown to grow along blood vessels. This suggests that the developing vasculature guides axonal sprouting after injury.\(^7\,^8\) However, the increased permeability for molecules otherwise excluded from the selective CNS environment might provide a unique chance for therapeutic intervention, with a potential window of opportunity displayed over the first days postinjury.\(^4\,^26\)

To address this multifaceted pathophysiology, several aspects of SCI must be taken into account to allow for tissue regeneration: Inhibition of the inflammatory response and sufficient perfusion might limit the secondary injury, while prevention of the glial scar formation could enable neural regeneration. Therefore, combinatorial approaches accounting for these mechanisms together in the adequate timespan might be promising for spinal cord repair.

2. PART 2: Experimental Therapeutic Approaches To Promote Vascular Repair

According to the systematic literature search, several experimental therapeutic approaches promising to diminish hypoxia and improve spinal cord microvasculature after SCI exist. In summary, 5 categories of experimental therapy were identified: (1) delivery of angiogenic factors, (2) genetic engineering, (3) physical stimulation, (4) cell transplantation, and (5) biomaterials carrying various factor delivery \(^9\) (Fig. 3). In the following section of this study, we aim to provide a comprehensive overview of the literature in this field, comparing and discussing the most promising approaches for clinical translation.

Fig. 3. Therapeutic approaches for vascular repair after experimental spinal cord injury: (1) angiogenic factors, (2) genetic engineering, (3) physical stimulation, (4) cell transplantation, and (5) biomaterials carrying various factor delivery.
1) Angiogenic factors

Many different therapeutic agents evoking an angiogenic answer, either directly or indirectly, have been investigated in experimental SCI models, often in combination with the implantation of biomaterials. In the following, we give an overview of different angiogenic factors and their previous experimental use in SCI research (Fig. 4A).

The therapeutic effects of vascular endothelial growth factor (VEGF) in SCI have been variable. VEGF-A is used in many studies and is a promising growth factor that binds to the tyrosine kinase receptors (VEGFRs) 1 and 2, resulting in the proliferation of ECs and angiogenesis. VEGF-A may also have neurotrophic, neuroprotective, and neuroproliferative effects. A common application of VEGF-A in experimental SCI is the direct injection into the lesion site which results in increased blood vessel density and blood vessel diameter. This effect has been confirmed in studies of controlled release of VEGF from biomaterials, such as gel foam. On the downside, increased BSCB-permeability was observed after application of VEGF-A indicating the growth of non- or only partially functional blood vessels. Changes in vessel architecture, resulting in tumor-like blood vessels, were described in human studies.

To improve the angiogenic and neurotrophic effect through the application of VEGF, a release from biomaterials in combination with other growth factors such as fibroblast growth factor-2 (FGF2), angiopoietin 1 and FGF2, or brain-derived...
neurotrophic factor was explored, also resulting in increased blood vessel density and associated localization of mature oligodendrocytes and axons.

FGF2 has also been frequently applied in experimental SCI. FGF2 was found to be highly overexpressed after SCI and FGF2 is known to play a role in proliferation, differentiation and BSCB-integrity. The application of FGF2 via biomaterials has been shown to increase the number of blood vessels, whereas no significant vascular changes were observed when FGF2 was infused at the injury site.

Further angiogenic factors that were previously tested for application in experimental SCI are hepatocyte growth factor (HGF) and the whole blood concentrates platelet-rich plasma (PRP) and platelet-derived wound healing formula (PDWHF). The number of blood vessels has been shown to increase after the application of activated PRP PDWHF alone, or PDWHF in combination with nerve growth factor and of HGF. Apart from these often-applied factors, other factors such as hormones, especially oestrogen, enzymes, and a multitude of anti-inflammatory substances listed in (Fig. 4A) also showed potential to induce angiogenesis or to increase BSCB-integrity following SCI.

2) Genetic engineering

Genetic engineering is not widely used in experimental SCI but was shown to hold the potential to increase vascular repair and functional outcome after SCI (Fig. 4B). Proofs-of-concept were brought forward in experiments using RNAs like micro-RNA-210, micro-RNA-126 contained in exosomes and X-inactive specific transcript-RNA. The endogenous overexpression of VEGF using viral vectors also increased the number of blood vessels at the injury site. The combination of VEGF165 and Ang-1 using an adeno-associated viral vector furthermore decreased the BSCB-permeability at the injury site. Therefore, genetic engineering could become a promising approach to support vascular repair after experimental SCI.

3) Physical stimulation

As the only noninvasive therapeutic approach, physical stimulation was also shown to increase angiogenesis and functional outcome after experimental SCI. Methods under investigation include water treadmill training, low-energy extracorporeal shock wave therapy, whole-body vibration, chronic mild hypoxia, and electric stimulation (Fig. 4C). Especially water treadmill training seems to improve the regeneration of functional blood vessels and improve functional outcome after SCI. Using low-energy extracorporeal shock wave therapy, not only increased vessel density but also enhanced functional outcomes suggesting that the regenerative potential is higher than the risk of further damage to the spinal cord. Electrical stimulation has made tremendous progress as experimental therapy in both preclinical and clinical studies, although only few studies focus on the evaluation of angiogenic effects. The application of transspinal direct current stimulation increases the blood temporarily and is able to stimulate cell proliferation and migration.

Another experimental therapeutic approach is the induction of chronic mild hypoxia after SCI. The induction of mild hypoxia results in vascular remodelling, with endothelial proliferation and vascular expansion being more pronounced in the white matter. Interestingly, newly formed blood vessels grow towards neurons and show the upregulation of TJ proteins, indicating the enhanced formation of functional blood vessels. These results are promising to increase angiogenic repair after SCI, but further research needs to address the effect of these approaches in the intermediate and chronic injury phase.

4) Cell transplantation

Cell transplantation is widely used after experimental SCI. Stem cells of different origins can result in a significant increase in angiogenesis and have previously been injected, infused, or implanted following SCI with the result of potently increasing the angiogenic response and improving functional outcome (Fig. 4D). Endothelial progenitor cells (EPCs), bone marrow mesenchymal stem cells, bone marrow-derived stem cells, human umbilical cord blood stem cells, neural stem cells (NSCs), amniotic mesenchymal stem cells, human-induced pluripotent stem cells, peripheral blood stem cells, lamina propria-derived olfactory ensheathing cells, olfactory bulb-derived ensheathing cells, and mesenchymal stem cells (MSCs) increase angiogenesis when applied after SCI by expressing a wide range of growth factors and modulating the microenvironment. The effect of these cell types is shown in Table 2.

As not all implanted cells survive the hypoxic environment at and around the injury site, cell-based therapies to date can only unfold a fraction of their potential. To overcome this limitation, other strategies rely on indirectly utilizing the therapeutic potential of stem cells. Especially the isolation of exosomes from different stem cells and their injection after experimental SCI showed similar results in terms of angiogenesis and functional outcome. Exosomes derived from many different cell types result in increased angiogenesis after injection. Amongst
these are NSC-derived exosomes, exosomes derived from human placenta-derived MSCs, and mesenchymal stromal cell-derived exosomes.

Another promising strategy is the combinatorial release of stem cells via biomaterials to enhance cell survival at the injury site by modulating the environment and by providing guidance for the regenerating tissue.

5) Biomaterials carrying various factor delivery

Biomaterials are materials engineered to interact with biological tissue and can be divided into natural materials sourced from plants or animals, synthetic materials and hybrid biomaterial combinations. Many different biomaterials improve angiogenesis when implanted in combination with stem cells or angiogenic factors or even without concomitant application of other therapeutics. The implantation of biomaterials alone already holds the potential to support the ingrowth of blood vessels into the lesion site. This angiogenic potential was shown using many different biomaterials, such as a reduced graphene oxide scaffold, an oxygen-generating hydrogel scaffold, a nanofiber-hydrogel composite, "NeuroGel", an aligned fibrin hydrogel, and a collagen type I scaffold (Fig. 4E). Biomaterials can be combined with other therapeutics at will, such as angiogenic factors and/or stem cells to potentiate their regenerative potential.

Both natural and synthetic biomaterials were previously used successfully in animal studies to promote vascular repair after SCI with different advantages and disadvantages. Natural biomaterials are often intrinsically biodegradable and therefore

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Table 2. Study overview of experimentally transplanted cells to promote vascular repair after experimental spinal cord injury (SCI)

<table>
<thead>
<tr>
<th>Species</th>
<th>SCI model</th>
<th>Method</th>
<th>Outcome</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rat</td>
<td>T10 Contusion</td>
<td>EPCs</td>
<td>Increased axonal and blood vessel regeneration with functional improvement.</td>
<td>Wang et al.37 (2018)</td>
</tr>
<tr>
<td>Rat</td>
<td>T10 Contusion</td>
<td>BMSCs overexpressing VEGF</td>
<td>Increased axonal and blood vessel regeneration with functional improvement. Increased VEGF protein expression.</td>
<td>Liu et al.58 (2020)</td>
</tr>
<tr>
<td>Rat</td>
<td>T8, 9 Compression</td>
<td>ADSCs</td>
<td>Increased blood vessel regeneration, vascular branching, and axonal regeneration. Increased formation of functional blood vessels, which are associated with blood vessels. Functional improvement.</td>
<td>Menezes et al.107 (2020)</td>
</tr>
<tr>
<td>Rat</td>
<td>T10 Contusion</td>
<td>HUCBCs</td>
<td>Increased blood vessel regeneration with functional improvement.</td>
<td>Ning et al.46 (2013)</td>
</tr>
<tr>
<td>Rat</td>
<td>T8 – 10 Contusion</td>
<td>NSCs</td>
<td>Increased blood vessel regeneration, VEGF protein expression, and remyelination with functional improvement.</td>
<td>Li et al.61 (2014)</td>
</tr>
<tr>
<td>Rat</td>
<td>T9 Contusion</td>
<td>AMSCs</td>
<td>Increased blood vessel regeneration, vascular branching, and VEGF protein expression, and functional improvement. The BSCB – disruption was reduced at 7 days indicating the regeneration of functional vessels.</td>
<td>Zhou et al.62,63 (2016, 2020)</td>
</tr>
<tr>
<td>Rat</td>
<td>T10 Contusion</td>
<td>hiPSCs</td>
<td>Increased blood vessel regeneration, myelination, and axonal regeneration with functional improvement. Transplanted cells differentiated into astrocytes, oligodendrocytes, and neurons which integrated with the host neural circuitry.</td>
<td>Nori et al.64 (2011)</td>
</tr>
<tr>
<td>Mice</td>
<td>T9 Contusion</td>
<td>PBSCs</td>
<td>Increased blood vessel regeneration, myelination, and axonal regeneration with functional improvement.</td>
<td>Takahashi et al.66 (2016)</td>
</tr>
<tr>
<td>Rat</td>
<td>C3, 4 Dorsolateral crush</td>
<td>LP and OB OECs</td>
<td>Increased axonal and blood vessel regeneration. The motor function was not assessed.</td>
<td>Richter et al.67 (2005)</td>
</tr>
<tr>
<td>Rat</td>
<td>T8 Contusion</td>
<td>MSCs</td>
<td>Increased axonal and blood vessel regeneration with no functional improvement.</td>
<td>Kumagai et al.68 (2013)</td>
</tr>
</tbody>
</table>

EPC, endothelial progenitor cell; BMSC, bone mesenchymal stem cell; VEGF, vascular endothelial growth factor; ADSC, adipose-derived stem cell; HUCBC, human umbilical cord blood stem cell; NSC, neural stem cell; AMSC, amniotic mesenchymal stem cell; hiPSC, human-induced pluripotent stem cell; PBSC, peripheral blood stem cell; LP OEC, lamina propria-derived olfactory ensheathing cell; OB OEC, olfactory bulb-derived ensheathing cell; MSC, mesenchymal stem cell.
useful to modify the delayed release of angiogenic factors or cells. Synthetic biomaterials, unless modified accordingly, are nondegradable and guarantee more structural and long-lasting stability as well as reduced batch-to-batch variance. For implantation and combination with factor delivery, they can be sterilized and chemically modified to allow for an optimal release profile. Due to their high modifiability, both natural and synthetic biomaterials can be adapted to provide optimal release kinetics of incorporated factors at a defined location. Release kinetics can be modified by using the intrinsic degradation rate of the biomaterial itself or noncovalent interactions between the incorporated therapeutics and the biomaterial.

After implantation or injection, biomaterials can occupy the space of a posttraumatic cavity and provide a cell-friendly environment with reduced infiltration of inflammatory cells. Therefore, biomaterials are often used in combination with stem cells, which highly depend on such an environment.

Exemplary, the number and density of blood vessels could be increased after implantation of an oligopolyethylene-glycol-fumarate-hydrogel combined with Rapamycin and Schwann-cells, of a prevascularized poly-L-lactic acid (PLLA)–polylactide-co-glycolic acid (PLGA) scaffold containing dental pulp stem cells, a fibrous porous silk scaffold containing human umbilical vein ECs, a hydrogel PLGA-scaffold containing NSCs and ECs, a gelatine sponge containing MSCs, and a PLGA-scaffold containing human MSCs. The implantation of a hydrogel containing NSCs and EPCs also increased the formation of blood vessels inside the scaffold. These findings show the great potential of combinatorial approaches using biomaterials containing angiogenic factors and/or stem cells of different types.

The application of biomaterials to promote vascular repair after SCI varies in time, location, and mode of application in the analysed studies (Table 3). In nearly all studies showing vascular changes, biomaterials are applied directly after induction of the injury with monitoring for up to 8 months. Only one study reports an increased blood vessel density after the implantation of a nanofiber-hydrogel composite 3 days after injury.
als are mostly applied at the injury site as scaffolds replacing the lesion after implantation or injection with only a few hydrogels being implanted on top of the injured spinal cord or injected systemically.}

**POTENTIAL FOR CLINICAL IMPLEMENTATION**

Taken together, all 5 experimental approaches discussed in this review show the potential to improve vascular repair and angiogenesis. The application of angiogenic factors (1) is a well-studied approach. Especially growth factors like VEGF have shown a promising potential to regenerate functional blood vessels. Genetic engineering (2) allows for direct activation of the endogenous expression of VEGF and other growth factors. Even if it is not applied as frequently as other approaches after SCI, it holds the potential to regenerate functional blood vessels. As physical stimulation techniques (3) also displayed the ability to induce vascular repair, they might be a useful supplementary therapy in a clinical setting as soon as intensive rehabilitation is possible. Cell transplantation (4) is another promising strategy to regenerate functional blood vessels after traumatic SCI, especially as they can be taken from a variety of sources. Their potential is inhibited by the toxic environment, which needs to be overcome to allow the cells to integrate at the injury site. The implantation of cells still poses a promising approach when combined with the implantation of biomaterials (5). These materials can be taken from many sources and are easily modifiable. Some have an inherent angiogenic potential and are biodegradable. As they can be modified to release incorporated therapeutics in the desired time frame at defined locations and provide guidance for regenerating axons, these combinatorial approaches seem promising for future research and potential clinical implementation. But before implementing promising biomaterials into clinical trials, several questions need to be addressed. For most biomaterials, especially when integrated in a combinatorial approach, the exact degradation time in SCI is neither known for humans nor for animals. As degradation byproducts might cause immunological reactions, biomaterials need to be actively characterized in preclinical and clinical studies. Furthermore, most of the biomaterials summarized are solid materials filling a transection or hemisection injury, but contusion SCI is most common clinically. This and the overall limited comparability from controlled animal models into the individual human SCI makes it difficult to translate the results of preclinical studies into the design of clinical trials and shows the need for using clinically relevant SCI animal models with contusion or compression injuries.

Before applying these approaches in a clinical setting, future research needs to evaluate the ideal combinations to accurately address the multifaceted pathophysiology and the time course of BSCB de- and regeneration to allow for the regeneration of functional blood vessels as a basis for neural regeneration.

**CONCLUSION**

SCI is characterized by the injury of both neural and vascular components and results in extensive tissue loss. The disruption of the vasculature and its insufficient regeneration with a prolonged state of BSCB-permeability exacerbate the primary injury by amplification of secondary injury mechanisms. To restore a functional vasculature to prevent neural loss and to promote neural regeneration is one of the most important challenges in SCI research. Several therapeutic approaches show an improved vascularization after SCI, like the application of angiogenic factors, genetic engineering, physical stimulation, cell transplantation, and biomaterials carrying various factor delivery. Combinatorial approaches, like implanted biomaterials with the ability to release angiogenic factors or therapeutic cells in a temporally and spatially controlled manner seem most promising to restore functional vasculature and to be translatable into clinical patient care in the future.

**NOTES**

**Supplementary Material:** Supplementary Table 1 can be found via https://doi.org/10.14245/ns.2244290.145.

**Supplementary Table 1.** Complete list of studies using biomaterials to promote vascular repair after spinal cord injury identified by the systematic literature search.

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**Author Contribution:** Conceptualization: LR, VH, MGF, PV; Data curation: LR, VH; Formal analysis: LR, VH; Project administration: VH, MGF; Visualization: LR; Writing - original draft: LR, VH; Writing - review & editing: VH, JS, LKS, MGF

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Supplementary Table 1. Complete list of studies using biomaterials to promote vascular repair after spinal cord injury identified by the systematic literature search

<table>
<thead>
<tr>
<th>Species</th>
<th>SCI model</th>
<th>Method</th>
<th>Time &amp; duration (day)</th>
<th>Outcome</th>
<th>References</th>
</tr>
</thead>
</table>
| Rat, Wistar, M | C6 Lateral hemisection (2 x 2 x 2 mm) | rGO scaffold | Acute (0) | Morphology: Functional blood vessels were observed in inner parts of the scaffold at 30 days; blood vessels were identified in close relation with regenerated axons inside the scaffold at 30 days; no significant alterations indicating toxicity 
Function: |
Survival: 10, 30 days | López-Dolado et al.19 (2016)
| Rat, Wistar, F | T8, 9 Transsection | Polyethylene glycol (PEG 600), mechanical microconnector system (mMS) | Acute (0) and chronic (5) | Morphology: After acute implantation of mMS blood vessels in close proximity with axonal structures could be detected in the mMS lumen at 5 wk; after scar resection and PEG 600 implantation the injury site was invaded by endothelial cells 
Function: Significant improvement at 10 and 30 days with mMS (BBB) and with PEG (mBBB) 
Survival: 30 days (mMS), 39 wk (PEG) | Brazda et al.38 (2016)
| Rat, Long Evans, F | T9, 10 Lateral hemisection (4 mm long) | VEGF165, FGF2 (in micropheros) PLGA multiple channel bridge | Acute (0) | Morphology: VEGF-levels at injury site were significantly (20-fold) greater than without VEGF-treatment at 1 week; significantly more blood vessels inside bridges with 2 μg VEGF and 1 μg FGF2 at 6 wk; the number of blood vessels was significantly increased; neurite growth was 1.7-fold greater at 6 wk in bridges with VEGF and FGF2 
Function: |
Survival: 1, 6 wk | De Laporite et al.52 (2011)
| Rat, Sprague-Dawley, F | T10 Transsection | DPSCs PLLA/PLGA-scaffold | Acute (0) | Morphology: Angiogenesis was significantly abundant at the injury site in rats treated with prevascularized scaffolds compared to DPSC-scaffolds, empty scaffolds and untreated rats; the mean diffusivity (indicator of molecular diffusion rate and demyelination) was significantly lower in prevascularized scaffolds at 8 wk, total vessel volume and vessel density in the lesion site were significantly higher in prevascularized scaffolds (increased vessel density in the rubrospinal tract, spinothalamic tract, dorsal column and spinocerebellar tract, but not in corticospinal tract) 
Function: Significant improvement at 2 and 4 wk (BBB) 
Survival: 8 wk | Guo et al.4* (2020)
| Rat, Wistar, F | T7 Transsection, (5 mm long) | FGF2 Hyaluronate collagen scaffold (CRS) | Acute (0) | Morphology: The number of blood vessels was significantly increased caudal, rostral and at the injury site in the FGF2-CRS and CRS groups compared to the control group at 12 wk as well as the FGF2-CRS group compared to the CRS group 
Function: Significant improvement after 4 wk (BBB) 
Survival: 12 wk | Shan et al.4* (2019)
| Mice, C57BL/6, M | T9 Hemisection | HUVECs Fibrous porous silk scaffold (FPSS) | Acute (0) | Morphology: Microvessel density and microvessel count in the FPSS-cells group were significantly higher at 28 days; significantly more regenerating axons formed along the blood vessels in the white matter at the injury site at 4 wk 
Function: Significant improvement after 4 wk (BBB) 
Survival: 4 wk | Zhong et al.46 (2020)
| Rat, Sprague-Dawley, F | T9, 10 Transsection (4 mm long) | Oxygen-generating scaffold (CPO/PLGA-microspheres in hydrogel) | Acute (0) | Morphology: Oxygen-generating scaffolds had a significantly oxygen level up to 21 days than hydrogel; blood vessels were observed in the scaffold and neovascularization was significantly greater than in control groups 
Function: Significant improvement after 2 wk (BBB) 
Survival: 12 wk | Liu et al.74 (2020)
| Rat, Fischer, F | T9 Transsection (2 mm long) | OPP+ hydrogel scaffold SCA, SCAs + RAPA (PLGA-microspheres) | Acute (0) | Morphology: Mean length density (Lv) of blood vessels was lowest in RAPA channels; blood vessel surface area was significantly larger in SC group; significantly larger mean vessel volumes in the SC group; mean diameter of blood vessels in SC channels was significantly greater; mean cross-sectional area per blood vessel in SC channels was significantly larger; number of axons regenerating had positive correlations to the surface and volume area densities of vessels and to the diameter of blood vessels; for SC channels significant negative correlations were shown between peak axonal density of total axon amplitudes and vessel cross-sectional area for total axons 
Function: |
Survival: 6 wk | Siddiqui et al.31 (2021)
| Rat, Sprague-Dawley, F | T8 Transsection (2 mm long) | Hydrogel scaffold with CBD-SDF1a + Taxol liposomes | Acute (0) | Morphology: The number of blood vessels was significantly higher in the full treatment group at 10 days; axonal fibers with a regenerative length longer than 1 mm at the lesion site were always close to regenerated blood vessels 
Function: Significant improvement after 3 wk (BBB) 
Survival: 5, 10 days, 6 wk | Liu et al.74 (2021)

(Continued)
### Supplementary Table 1. Complete list of studies using biomaterials to promote vascular repair after spinal cord injury identified by the systematic literature search (Continued)

<table>
<thead>
<tr>
<th>Species</th>
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<tbody>
<tr>
<td>Rat, Sprague-</td>
<td>T9,10 Lateral</td>
<td>VEGF/PDGF Hydrogel patch Mini-pump</td>
<td>Acute (0)</td>
<td>Morphology: The levels of VEGF and NT-3 were significantly increased at the injury site 1 and 4 wk; NT-3 + ASCS formation; &lt;br&gt;the blood vessel density was significantly higher; &lt;br&gt;more intensive blood vessels are &lt;br&gt;ordinarily accompanied with lower infiltration of macrophages and vice versa at the lesion site &lt;br&gt;Function: Significant improvement at all timepoints (BBB) &lt;br&gt;Survival: 1, 4, 8 wk</td>
<td>Xu et al. (2020)</td>
</tr>
<tr>
<td>Dawley, F</td>
<td>hemisection (4 mm long)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rat, Fischer</td>
<td>T9, 10 Lateral</td>
<td>NSCs, EPCs Hydrogel</td>
<td>Acute (0)</td>
<td>Morphology: Infiltration of native NSCs into lesion area and formation of blood vessels appeared in NSC/EPC hydrogel group, the acellular hydrogel group and in the control group with connective tissue formation &lt;br&gt;Function: significant improvement after 2 wk (BBB) &lt;br&gt;Survival: 4 wk</td>
<td>Marrotte et al. (2021)</td>
</tr>
<tr>
<td>F344, F</td>
<td>hemisection (4 mm long)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rat, Sprague-</td>
<td>T9,10 Lateral</td>
<td>FGF2, HAMC, PLGA-nanoparticles, (intrathecal injection)</td>
<td>Acute (0)</td>
<td>Morphology: The number of blood vessels was significantly higher; neovascularization was significantly higher at 4 and 8 wk; &lt;br&gt;— polarized M2 subtypes possibly secreted VEGF which promotes blood vessel formation &lt;br&gt;Function: No significant improvement after 3 wk (BBB) and 4 wk (Inclined Plane Test) &lt;br&gt;Survival: 4, 8 wk</td>
<td>Kang et al. (2013)</td>
</tr>
<tr>
<td>Dawley, F</td>
<td>Contusion injury</td>
<td>VEGF, Ang-1, FGF2 PLGA-microspheres Injection into injury site</td>
<td>Acute (0)</td>
<td>Morphology: The levels of VEGF, Ang-1 and FGF2 were significantly higher at the injury site at 2, 4, and 8 wk in animals treated with angiogenic microspheres; the numbers of blood vessels at the injury site at 4 and 8 wk were significantly higher; the numbers of cells positive for nestin or βIII-tubulin (marker of neural precursor recruitment) at the injury site were significantly higher and mostly associated with blood vessels; the density of neurofilament (NF)-positive fibers was significantly greater at the injury site at 8 wk in treated animals often aligned with blood vessels; serotonergic (5-HT) fibers were associated with blood vessels and significantly longer in treated rats; the numbers of MBP-positive mature oligodendrocytes were significantly higher in treated rats and aggregated around blood vessels in the white matter region; most axons in treated animals were myelinated and followed blood vessels at 12 wk &lt;br&gt;Function: Significant improvement after 14d (BBB) &lt;br&gt;Survival: 2, 4, 8, 12 wk</td>
<td>Yu et al. (2016)</td>
</tr>
<tr>
<td>Rat, Sprague-</td>
<td>T9, 10 Lateral</td>
<td>NPCs, ECs Hydrogel + PLGA-scaffold</td>
<td>Acute (0)</td>
<td>Morphology: Significantly more vessels in the implant + NPCs/ECs treated rats in the lesion epicenter at 8 wk; only implant + NPCs/ECs treated rats had EBA-positive vessels (marker of functional BSCB) at the injury epicenter; the vessel density was significantly increased at the injury epicenter &lt;br&gt;Function: / &lt;br&gt;Survival: 3, 8 wk</td>
<td>Rauch et al. (2009)</td>
</tr>
<tr>
<td>Dawley, F</td>
<td>Contusion injury</td>
<td>VEGF, Ang-1, FGF2 PLGA-microspheres Injection into injury site</td>
<td>Acute (0)</td>
<td>Morphology: The number of blood vessels was significantly higher; &lt;br&gt;— number of blood vessels at the injury site at 4 and 8 wk were significantly higher; the numbers of cells positive for nestin or βIII-tubulin (marker of neural precursor recruitment) at the injury site were significantly higher and mostly associated with blood vessels; the density of neurofilament (NF)-positive fibers was significantly greater at the injury site at 8 wk in treated animals often aligned with blood vessels; serotonergic (5-HT) fibers were associated with blood vessels and significantly longer in treated rats; the numbers of MBP-positive mature oligodendrocytes were significantly higher in treated rats and aggregated around blood vessels in the white matter region; most axons in treated animals were myelinated and followed blood vessels at 12 wk &lt;br&gt;Function: Significant improvement after 14d (BBB) &lt;br&gt;Survival: 2, 4, 8, 12 wk</td>
<td>Yu et al. (2016)</td>
</tr>
<tr>
<td>Rat, Sprague-</td>
<td>T10 Lateral</td>
<td>VEGF/NT-3, BMSCs PLGA-nanoparticles with acellular spinal cord scaffold (ASCs)</td>
<td>Acute (0)</td>
<td>Morphology: The blood vessel density 200 µm from the lesion cavity was not significantly affected by VEGF/PDGF treatment &lt;br&gt;Function: No significant improvement (BBB) &lt;br&gt;Survival: 1, 3 mo</td>
<td>Lutton et al. (2012)</td>
</tr>
<tr>
<td>Dawley, F</td>
<td>hemisection (3 mm long)</td>
<td></td>
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<td></td>
</tr>
<tr>
<td>Rat, Sprague-</td>
<td>T9 Lateral</td>
<td>Microsot electrospun oriented fiber scaffold pDNA/Liposome (NGF) + IL-4</td>
<td>Acute (0)</td>
<td>Morphology: The number of blood vessels was significantly higher; neovascularization was significantly higher at 4 and 8 wk; &lt;br&gt;— polarized M2 subtypes possibly secreted VEGF which promotes blood vessel formation &lt;br&gt;Function: Significant improvement after 3 wk (BBB) and 4 wk (Inclined Plane Test) &lt;br&gt;Survival: 4, 8 wk</td>
<td>Xi et al. (2020)</td>
</tr>
<tr>
<td>Rat, Sprague-</td>
<td>T9 Lateral</td>
<td>Nanofiber-hydrogel composite (NHC) (Injection into injury site)</td>
<td>Subacute (3)</td>
<td>Morphology: The blood vessel density increased in time in the injury with NHC or HA but decreased in the control group; the blood vessel density was significantly higher at 28 days &lt;br&gt;Function: No significant improvement (BBB) &lt;br&gt;Survival: 3, 7, 28, 56 days</td>
<td>Li et al. (2020)</td>
</tr>
<tr>
<td>Dawley, F</td>
<td>(175 kDyne)</td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Rat, Sprague-</td>
<td>T10 Lateral</td>
<td>VEGF/NT-3, BMSCs PLGA-nanoparticles with acellular spinal cord scaffold (ASCs)</td>
<td>Acute (0)</td>
<td>Morphology: The levels of VEGF and NT-3 were significantly increased at the injury site 1 and 4 wk in the VEGF/NT-3-ASCs- and BMSC-treatment groups; the blood vessel density was significantly higher; more intensive blood vessels are ordinarily accompanied with lower infiltration of macrophages and vice versa at the lesion site &lt;br&gt;Function: Significant improvement at all timepoints (BBB) &lt;br&gt;Survival: 1, 4, 8 wk</td>
<td>Xu et al. (2020)</td>
</tr>
<tr>
<td>Dawley, F</td>
<td>Contusion, (3 mm x 1.5 mm)</td>
<td>VEGF, BDNF PLGA-microspheres, HA-antiNgR -scaffold</td>
<td>Acute (0)</td>
<td>Morphology: The number of blood vessels and axonal fibers were significantly higher in HA + PLGA scaffolds treated rats at 8 wk; more myelinated axons were found in HA + PLGA scaffolds at 14 wk compared to HA scaffolds and they &lt;br&gt;often had contact with blood vessels &lt;br&gt;Function: Significant improvement after 2 wk (BBB, CatWalk) &lt;br&gt;Survival: 4, 8, 14 wk</td>
<td>Wen et al. (2016)</td>
</tr>
<tr>
<td>Rat, Sprague-</td>
<td>T9 Lateral</td>
<td>VEGF, BDNF PLGA-microspheres, HA-antiNgR -scaffold</td>
<td>Acute (0)</td>
<td>Morphology: The number of blood vessels and axonal fibers were significantly higher in HA + PLGA scaffolds treated rats at 8 wk; more myelinated axons were found in HA + PLGA scaffolds at 14 wk compared to HA scaffolds and they &lt;br&gt;often had contact with blood vessels &lt;br&gt;Function: Significant improvement after 2 wk (BBB, CatWalk) &lt;br&gt;Survival: 4, 8, 14 wk</td>
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<td>Contusion, (3 mm x 1.5 mm)</td>
<td>VEGF, BDNF PLGA-microspheres, HA-antiNgR -scaffold</td>
<td>Acute (0)</td>
<td>Morphology: The number of blood vessels and axonal fibers were significantly higher in HA + PLGA scaffolds treated rats at 8 wk; more myelinated axons were found in HA + PLGA scaffolds at 14 wk compared to HA scaffolds and they &lt;br&gt;often had contact with blood vessels &lt;br&gt;Function: Significant improvement after 2 wk (BBB, CatWalk) &lt;br&gt;Survival: 4, 8, 14 wk</td>
<td>Wen et al. (2016)</td>
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### Supplementary Table 1. Complete list of studies using biomaterials to promote vascular repair after spinal cord injury identified by the systematic literature search (Continued)

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<tbody>
<tr>
<td>Rat, Sprague-Dawley, F</td>
<td>T9, 10 Transection, (4 mm long)</td>
<td>NGF, ChABC Alginate beads, electrospun PDS scaffold</td>
<td>Acute (0)</td>
<td>Morphology: Many parallel aligned blood vessels in contact with regenerating axons were present in the implant; some endothelial cells were in close association with electrospun monofilaments at 7 days, after 2–3 wk blood cells were observed in this lumen &lt;br&gt; Function: Significant improvement at 21 days (BBB) &lt;br&gt; Survival: 7, 21 days</td>
<td>Colell et al.24 (2016)</td>
</tr>
<tr>
<td>Rat, Sprague-Dawley, F</td>
<td>T10 Transection, (1,5 mm long)</td>
<td>MSCs PLGA, GS-scaffold</td>
<td>Acute (0)</td>
<td>Morphology: The number of blood vessels was significantly increased in GS+MSCs treated rats at the injury site at 1 wk; only MSCs surrounding blood vessels expressed VEGF &lt;br&gt; Function: / &lt;br&gt; Survival: 1, 8 wk</td>
<td>Zeng et al.44 (2011)</td>
</tr>
<tr>
<td>Rat, Sprague-Dawley, M</td>
<td>T9 Clip compression hemisection, (3 mm long)</td>
<td>pSV-VEGF PLGA/DC-Chol-nanospheres, Injection into injury site</td>
<td>Acute (0)</td>
<td>Morphology: Arteriole density was significantly higher in VEGF-loaded PLGA/DC-Chol nanosphere treated rats at 4 wk &lt;br&gt; Function: Significant improvement after 2 wk (BBB) &lt;br&gt; Survival: 2, 4, 6 wk</td>
<td>Gwak et al.46 (2016)</td>
</tr>
<tr>
<td>Rat, Sprague-Dawley, F</td>
<td>T9, 10 Lateral hemisection, (4 mm long)</td>
<td>antiNgR HA-PLL hydrogel</td>
<td>Acute (0)</td>
<td>Acute (0)</td>
<td>Function: / &lt;br&gt; Survival: 2, 4, 8, 12 wk</td>
</tr>
<tr>
<td>Rat, Wistar, F</td>
<td>T12 Transection, (3 mm long)</td>
<td>PDWHF, NGF Collagen-I</td>
<td>Acute (0)</td>
<td>Morphology: Axonal regrowth and number of blood vessels were significantly greater in PDWHF and NGF groups; the number of vessels was significantly greater in PDWHF group compared to NGF group &lt;br&gt; Function: No significant improvement observed &lt;br&gt; Survival: 4, 8, 12 wk</td>
<td>Hiraizumi et al.45 (1996)</td>
</tr>
<tr>
<td>Rat, Sprague-Dawley, M</td>
<td>T9–11 Lateral hemisection, (3 mm long)</td>
<td>VEGF615 PLGA-nanospheres, ASCS</td>
<td>Acute (0)</td>
<td>Morphology: Vessel branches increased significantly at 1 wk in V-ASCS group compared with control and B-ASCS groups but in the following weeks the density of vessel branches decreased in B-and V-ASCS groups, vessel volume/tissue volume (VV)/(TV) were significantly increased in B- and V-ASCS groups at 1wk and VV/TV were significantly greater in V-ASCS compared with B-ASCS; VV/TV was not significantly different between V-ASCS and B-ASCS groups at 8 wk and data in V-ASCS group was significantly lower than in Sham group; vessel density (VDm) was significantly highest in V-ASCS group, higher in B-ASCS group and the lowest in control group at 1wk; VDm was significantly higher in V-ASCS group compared with B-ASCS at 8 wk; average vessel diameter was significantly greater in V-ASCS and B-ASCS groups compared with control group at 1wk but there was no significant difference at 4 and 8 wk, density of vessel branches (VBDn) was significantly higher in V-ASCS group compared with control group and B-ASCS groups at 1, 4 and 8 wk &lt;br&gt; Function: Significant improvement after 3d (BBB) &lt;br&gt; Survival: 4, 8, 8 wk</td>
<td>Xu et al.57 (2017)</td>
</tr>
<tr>
<td>Cat</td>
<td>L2 Incomplete cord injury (5 mm longitudinal insertion of Teflon catheters)</td>
<td>PDWHF Hydron (coated on Teflon catheter sheaths)</td>
<td>Acute (0)</td>
<td>Morphology: The number of vessels was significantly greater in PDWHF-treated animals and the number of vessels appeared significantly more 1 mm from the lesion site &lt;br&gt; Function: No significant improvement observed &lt;br&gt; Survival: 3 wk</td>
<td>Hiraizumi et al.56 (1993)</td>
</tr>
<tr>
<td>Rat, Sprague-Dawley, M</td>
<td>T9 Transection, (3 mm long)</td>
<td>VEGF Collagen scaffold (CS)</td>
<td>Acute (0)</td>
<td>Morphology: The microvessel density and microvessel count in the CS/VEGF group were significantly higher at 12 wk &lt;br&gt; Function: Significant improvement after 7 wk &lt;br&gt; Survival: 4, 10, 12 wk</td>
<td>Wang et al.208 (2018)</td>
</tr>
<tr>
<td>Rat, Long Evans, F</td>
<td>T9, 10 Lateral hemisection, (4 mm long)</td>
<td>VEGF164 Alginate/fibrinogen-hydrogel, chitosan-nanoparticles</td>
<td>Acute (0)</td>
<td>Morphology: Endothelial, III-III tubulin and growing neurites staining intensity were significantly greater in rats treated with VEGF-loaded hydrogels at 4 wk &lt;br&gt; Function: No significant improvement (CatWalk) &lt;br&gt; Survival: 4 wk</td>
<td>des Rieux et al.55 (2014)</td>
</tr>
<tr>
<td>Rat, Sprague-Dawley, F</td>
<td>T5, 10 Transection, (3 mm long)</td>
<td>NeuroGel (PCHIPMA)-hydrogel</td>
<td>Acute (0)</td>
<td>Morphology: Vascular response with proliferating capillary sprouts into the hydrogel at 7 days as well as glial cells; extensive ingrowth of sinusoidal capillaries &lt;br&gt; Function: Significant improvement observed after 2 wk &lt;br&gt; Survival: 2, 4 mo</td>
<td>Woorly et al.60 (2001)</td>
</tr>
<tr>
<td>Rat, Fischer, F</td>
<td>T9, 10 Transection, (4 mm long)</td>
<td>BDNE EGF1 PLA-foam scaffold, fibrin glue</td>
<td>Acute (0)</td>
<td>Morphology: The number of blood vessels was significantly higher in fibrin only group at 2, 4, and 8 wk and in BDNF + foam group at 8 wk &lt;br&gt; Function: No significant improvement (BBB) &lt;br&gt; Survival: 2, 4, 8 wk</td>
<td>Patist et al.64 (2004)</td>
</tr>
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</table>

www.e-neurospine.org https://doi.org/10.14245/ns.2244624.312

(Continued)
**Supplementary Table 1.** Complete list of studies using biomaterials to promote vascular repair after spinal cord injury identified by the systematic literature search (Continued)

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<tr>
<td>Cat</td>
<td>T6, 7</td>
<td>NeuroGel</td>
<td>Acute (0)</td>
<td>Function: At 17 months large capillaries crossed the injury site and a profuse network of blood vessels in proximity of the spinal stumps were seen.</td>
<td>Woerly et al. (2004)</td>
</tr>
<tr>
<td></td>
<td>Transaction, (3 mm long)</td>
<td></td>
<td></td>
<td>Function: Improvement observed (treadmill test) Survival: 6, 9, 17 mo</td>
<td></td>
</tr>
<tr>
<td>Rat, Sprague-Dawley, F</td>
<td>T9</td>
<td>Chitosan − graphene oxide (CS/GO) scaffold</td>
<td>Acute (0)</td>
<td>Function: Regenerating neurons and erythrocytes were seen inside the GS/GO scaffold.</td>
<td>Yang et al. (2021)</td>
</tr>
<tr>
<td></td>
<td>Transaction, (2 mm long)</td>
<td></td>
<td></td>
<td>Function: Significant improvement after 7 wk (BBB) and 10 wk (SSEP). Survival: 10 wk</td>
<td></td>
</tr>
<tr>
<td>Rat, Sprague-Dawley, F</td>
<td>T9</td>
<td>NeuroGel (PHPMA hydrogel)</td>
<td>Acute (0)</td>
<td>Function: Blood vessels, processes of astrocytes and myelinated and unmyelinated axons were observed arranged inside the porous hydrogel implant at 5 mo Function: / Survival: 5 months</td>
<td>Woerly et al. (1999)</td>
</tr>
<tr>
<td>Rat, Sprague-Dawley, F</td>
<td>T9, 10</td>
<td>AFG/ISAP hydrogel (aligned fibrin hydrogel/self-assembling peptide nanofiber hydrogel)</td>
<td>Acute (0)</td>
<td>Function: Blood vessels were found in the lesion site in AFG/ISAP-treated rats with larger diameter compared with AFG-treated rats at 12 wk; the microvessel density was significantly greater in AFG/ISAP-treated rats at the lesion site compared with AFG-treated rats and control group at 12 wk; regenerating blood vessels and axons showed colocalization Function: Significant improvement after 3 wk (BBB) and at 12 wk (Catwalk, MEP). Survival: 1, 8, 12 wk</td>
<td>Man et al. (2021)</td>
</tr>
<tr>
<td>Rat, Fischer, F</td>
<td>T9</td>
<td>SC, RAPA OPP+ hydrogel (MG), PLGA-microspheres (MS)</td>
<td>Acute (0)</td>
<td>Function: Vessel length and vessel surface area were significantly greater in SC + Empty-MS treated rats compared with MG + Empty-MS at 6 wk but not different to SC + Low or medium RAPA-MS; vessel length and surface area in SC-loaded scaffolds with high doses RAPA was not different from MG-only scaffolds without RAPA; the mean blood vessel diameter in SC + Empty-MS was greater than in MG-only scaffolds; the number of vessels with Pericytes (PC)/ Endothelial cells (EC) was greater in SC + Empty-MS treated rats; PC/EC ratio was highest in SC + Low RAPA-MS treated rats; surface area of functionals was significantly higher in RAPA treated rats compared to MG + Empty-MS group indicating an improved vascular connectivity to the systemic circulation</td>
<td>Hakim et al. (2019)</td>
</tr>
<tr>
<td>Rat, Sprague-Dawley, F</td>
<td>C3, 4</td>
<td>Collagen-1 scaffold</td>
<td>Acute (0)</td>
<td>Function: Blood vessels were seen inside the scaffold but most of them were not associated with ZO-1-immunoreactive tight junctions, ZO-1-immunoreactivity was intensive at the transition zones, density of blood vessels was significantly greater at the transition zone of the scaffold compared to the contralateral non-lesioned white matter but staining within the scaffold was not significantly greater than that of the contralateral white matter; the number of functional vessels was significantly lower within the scaffold at 10 wk than the total number of vessels and most functional vessels within the scaffold were not positive for ZO-1 tight junction → delayed or slowed maturation Function: / Survival: 10 wk</td>
<td>Altinova et al. (2020)</td>
</tr>
<tr>
<td>Rat, Sprague-Dawley, F</td>
<td>T9, 10</td>
<td>Aligned fibrin hydrogel (AFG)</td>
<td>Acute (0)</td>
<td>Function: The number of blood vessels was significantly greater in AFG-treated rats at 2 and 4 wk Function: Significant improvement after 2 wk (BBB) Survival: 1, 2, 4, 8 wk</td>
<td>Yao et al. (2018)</td>
</tr>
<tr>
<td>Canine, Beagle, F</td>
<td>T10</td>
<td>Gelatin sponge (GS), NT-3/fibroin particles (NF)</td>
<td>Acute (0)</td>
<td>Function: Blood vessels or capillaries were identified only in the NF-GS group at 4 wk Function: Significant improvement after 3 wk (Olby score test) and at 4 wk Survival: 4 wk</td>
<td>Li et al. (2018)</td>
</tr>
<tr>
<td>Rat, Sprague-Dawley, F</td>
<td>T9, 10</td>
<td>MSCs, PLGA-scaffold</td>
<td>Acute (0)</td>
<td>Function: The number of blood vessels was significantly increased around the lesion site at 6 wk in the transplant group Function: Significant improvement after 7d (BBB), 2 wk (inclined plane downward, righting reflex) and 4 wk (at level allodynia) Survival: 6 wk</td>
<td>Ropper et al. (2017)</td>
</tr>
<tr>
<td>Rat, Sprague-Dawley, F</td>
<td>T10</td>
<td>CORM-2-SLNs (Carbon monoxide-releasing molecule-2 solid lipid nanoparticle) (i.p. injection)</td>
<td>Acute (0) For 8 days</td>
<td>Function: The fluorescence intensity of Evan’s Blue dye was significantly lower in the treatment group at the injury site at 1d indicating reduced RSCB permeability; the number of blood vessels was significantly greater in the treatment group at 21 days Function: Significant improvement after 7d (BBB, withdrawal test) Survival: 1, 3, 14, 21 days</td>
<td>Joshi et al. (2018)</td>
</tr>
</tbody>
</table>

(Continued)
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<tbody>
<tr>
<td>Rat, Wistar, F</td>
<td>T8 Complete transection of CST</td>
<td>VEGF165, Ad.CMV. VEGF165 (Injection in lesion site, controlled release via Matrigel)</td>
<td>Acute (0) For 30 days</td>
<td>Morphology: The number of vessels in VEGF-treated rats was significantly higher (ca. 300%), retrograde axonal degeneration was significantly reduced in VEGF-treated rats, regenerating CST axons (HRP-labeled) where located mostly in the ventral gray matter. Function: / Survival: 0, 3, 7, 10, 16, 18, 30 days</td>
<td>Facchiano et al. (2002)</td>
</tr>
<tr>
<td>Rat, Sprague-Dawley, M</td>
<td>T7 Contusion injury</td>
<td>VEGF165 Gelfoam placed on injury site</td>
<td>Acute (0)</td>
<td>Morphology: Significant increase in BSCB permeability after VEGF-treatment in non-enhancing-areas (magnetic resonance imaging) in the epicenter in the subacute (7–14 days) and chronic (28–56 days) periods. Function: Significant improvement at 28 days, but not at 56 days (BBB). Survival: 56 days</td>
<td>Patel et al. (2009)</td>
</tr>
</tbody>
</table>

/ not assessed; AFG, aligned fibrin hydrogel; fSAP, functionalized self-assembling peptides; CORM-2, carbon monoxide-releasing molecule-2; rGO, reduced graphene oxide; DPSCs, dental pulp stem cells; PLGA, polylactide-co-glycolide acid; PLLA, poly-L-lactic acid; FGF2, fibroblast growth factor 2; DPSCs, dental pulp stem cells; CRS, hyaluronate collagen scaffold; HUVECs, human umbilical vein endothelial cells; FPSS, fibrous porous silk scaffold; CPO, calcium peroxide; OPE, oligopolyethylene glycol-fumarate; mMS, mechanical microconnector system; BBB, Basso, Beattie and Bresnahan score; VEGF, vascular endothelial growth factor; PGGF, fibroblast growth factor 2; DPSCs, dental pulp stem cells; CRB, corticospinal tract; Ad.CMV.VEGF, replication-defective adenovirus coding for VEGF.