ACS Chemical Neuroscience

REVIEW

pubs.acs.org/acschemicalneuroscience

Hydrogels in Spinal Cord Injury Repair Strategies

Giuseppe Perale,[†,‡] Filippo Rossi,[*,†,‡] Erik Sundstrom,[§] Sara Bacchiega,[∥] Maurizio Masi,[†] Gianluigi Forloni,[‡] and Pietro Veglianese[‡]

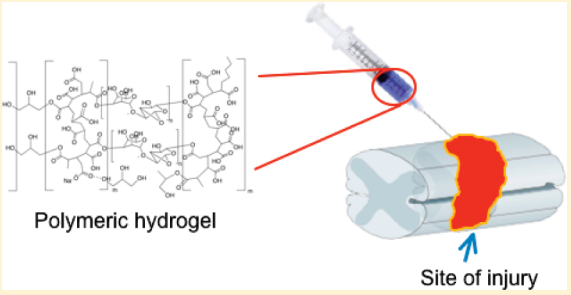
[†]Department of Chemistry, Materials and Chemical Engineering "Giulio Natta", Politecnico di Milano, via Mancinelli 7, 20131 Milan, Italy

[‡]Department of Neuroscience, Mario Negri Institute for Pharmacological Research, via La Masa 19, 20156 Milan, Italy

[§]Department of NeuroBiology, Karolinska Institutet, Novum 5, 14186 Stockholm, Sweden

[∥]Mi.To. Technology s.r.l., Licensing Department, Viale Vittorio Veneto 2/a, 20124 Milan, Italy

ABSTRACT: Nowadays there are at present no efficient therapies for spinal cord injury (SCI), and new approaches have to be proposed. Recently, a new regenerative medicine strategy has been suggested using smart biomaterials able to carry and deliver cells and/or drugs in the damaged spinal cord. Among the wide field of emerging materials, research has been focused on hydrogels, three-dimensional polymeric networks able to swell and absorb a large amount of water. The present paper intends to give an overview of a wide range of natural, synthetic, and composite hydrogels with particular efforts for the ones studied in the last five years. Here, different hydrogel applications are underlined, together with their different nature, in order to have a clearer view of what is happening in one of the most sparkling fields of regenerative medicine.



KEYWORDS: Hydrogels, polymers, scaffold, regenerative medicine, spinal cord injury, tissue engineering

Traumatic spinal cord injury (SCI) is an irreversible dramatic event that can incapacitate victims for life.[1−4] Although the incidence is relatively low, the often severe disability that follows and the fact that the victims are often young people, the consequences for the patient is severe and the impact on societal costs is significant. The injury is the result of a primary event due to contusive, compressive, or stretch injury,[1,2,5] followed by the so-called "secondary injury", commonly considered the main cause of the post-traumatic neural degeneration of the cord itself.[6−8] Functional deficits of SCI are caused by different temporal events: spinal cord compression and/or contusion lead to ischemic events that limit both oxygen and glucose contribution to the tissue, with concomitant neuronal cell death, axon damage, and demyelination.[5] Subsequently, glial activation, release of inflammatory factors and cytokines, and scar formation that impedes axons to regrow[8,9] aggravate the progression of the damage.

SCI research is following two principal paths.[6,9−11] The first one, already applied in human cases, is based on systemic pharmacological treatments in order to contain side effects (ischemia, free radical release, and inflammation) using neuroprotective drugs (such as corticosteroids)[12−14] and to promote self-regeneration using stimulating factors.[15] The second one relies on tissue engineering[16−18] approaches such as the direct injection of stem cells[19−21] and active agents (drugs, antibodies, and peptides) into the affected area with the aim to bridge the lesion, possibly after removal of the glial scar or reducing endogenous neurite-inhibitory molecules.[22,23] Direct injection of in vitro cultured cells or drugs is the most common choice, but keeping transplanted cells in the lesion area is often desired as transplanted cells readily leave the zone of injection if not confined by any support. To achieve this, a new potential approach is to combine material science with tissue engineering as has been proposed and developed.[16,24−26] In Figure 1 are presented classic tissue engineering approaches as the combination of scaffolds with cells and active agents in order to replace damaged parts of biological tissues.[17,18]

In the wide field of biomaterials, increased attention is given to polymers, not only to fabricate three-dimensional scaffolds but also to develop injectable systems for tissue engineering.[26−34] One of the most suitable classes of compounds for these purposes is surely represented by hydrogels.[16,28,31,35−39] These polymers are typically soft and elastic due to their thermodynamic compatibility with water.[16,33,36,40] They can be designed as temporary structures having desired geometry and physical, chemical, and mechanical properties adequate for implantation into chosen target tissue.[6,41−43]

The aim of this Review is to show the different types of hydrogels used as scaffolds for SCI repair strategies. We on purpose decided to focus our attention only on the past few years, in order to show the most promising and recent perspectives in this field. Indeed, the rapid expansion of nanotechnology during the last years has led to new perspectives and advances in biomedical research as well as in clinical practice.[7]

Received: March 24, 2011
Accepted: May 4, 2011
Published: May 04, 2011

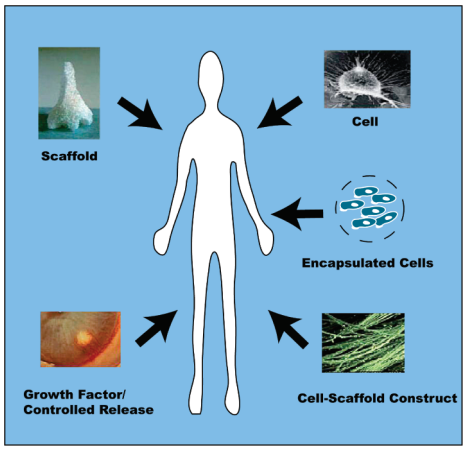


Figure 1. Tissue engineering approaches: the smart combination of cells and materials to replace damaged or missing parts of living tissues. Reproduced with permission from ref 16. Copyright Wiley-VCH Verlag GmbH & Co. KGaA.

HYDROGELS

The physical aspects of scaffold design, as with polymer choice, depend largely on the application. The scaffold is meant to provide the appropriate chemical, physical, and mechanical properties required for cell survival and tissue formation.[35,36,40] Essentially, the polymeric scaffold is designed to define the cellular microenvironment required for optimal function. Indeed, in the wide field of biopolymers,[20,44,45] one of the most suitable classes for these purposes is represented by hydrogels. They are three-dimensional (3D) networks of hydrophilic polymers held together by covalent bonds or other cohesive forces such as hydrogen or ionic bonds.[36,46−48]

They are glassy in the dry state and then, in the presence of solvents, able to swell while preserving their original shape to form elastic gels. Capable to retain a large amount of water in their structure (up to 95% of the total weight), they can either degrade in it by polymer chain degradation reactions (e.g., hydrolysis or proteolysis into smaller molecules) and are then called resorbable hydrogels, or they cannot and are then called stable hydrogels.[35,36] These scaffolds slowly degrade in the physiological environment, leading the growing tissue to replace the former filled site.[46] An important advantage is the possibility to minimize the risks of surgical procedures due to their injectability and ability to create a 3D network in situ, in the target tissue.[26,35]

In general, hydrogels may be classified as either synthetic or natural in origin. On one hand, synthetic polymers can be tuned in terms of composition, rate of degradation, and mechanical and chemical properties.[49,50] On the other hand, naturally derived polymers provide structures extremely similar to living tissues such as stimulating a specific cellular response, which sometimes supersedes the advantages of synthetic polymers. Moreover, owing to their similarity with the extracellular matrix (ECM), natural polymers may also reduce the stimulation of chronic inflammation or immunological reactions and toxicity, often detected with synthetic polymers.[51,52] However, this is not true for every natural-derived polymer; the ones from nonmammalian sources (e.g., seaweed and crustaceans) can induce immune reactions. Moreover, even mammalian hydrogels (e.g., those collagen-based), if raw materials are improperly harvested from some species, might induce immune reactions in humans.

Thus, different reasons make the above-mentioned biomaterials very attractive for improving tissue regeneration and central nervous system (CNS) repair:[45] (i) tissuelike mechanical abilities, conformable to the CNS tissue;[43,53] (ii) porous structure allowing cell infiltration, transplantation, and axon outgrowth;[53] (iii) ability to incorporate adhesion and/or growth-promoting molecules in the hydrogel to enhance cell attachment and tissue growth;[54] (iv) capacity of drug/gene vector incorporation and precise in situ delivery.[38,42,55,56]

In order to make a comprehensive overview of their use in spinal cord injury repair strategies we decided to classify hydrogels on the basis of the following.

Nature: natural, synthetic, or a combination of the two.[49,50]

Function: drug or cell carriers or a combination of the two.[24,39,57]

1. Natural Derived Hydrogels. In order to follow the similarities between the implanted materials and the living tissue, researchers studied the possibility to synthesize hydrogels starting from molecules present in living tissues. In particular, the most suitable are collagen, hyaluronic acid (HA), and polysaccharides (agarose, alginate, cellulose, gellan gum, scleroglucan, and xyloglucan). Although regenerative axonal growth occurs in a liquified spinal cord lesion cavity without obvious physical support,[58] regeneration is facilitated by a supporting scaffold equivalent to the endoneurium and perineurium in a peripheral nerve, that can act as a bridge in order to approximate the disconnected axonal groups for the damaged area.[43,53] The aim of using hydrogel is to replace the damaged area with a structural matrix.[24,51]

As explained before, naturally derived macromers and their use have increased in the past few years due to inherent biocompatibility and enzymatic degradation.[49] They are macroporous, soft materials able to allow cell adhesion and migration.[50] Moreover, they can be manipulated in order to obtain channels for nerve guidance or sustained drug delivery. Table 1 presents in detail the main natural polymers and highlights examples of their SCI application. For the sake of clarity, it is useful to briefly comment on Table 1. Following most promising regenerative medicine approaches toward other pathologies, also several recent studies in SCI repair are combining hydrogels with stem cells in order to provide in situ cell delivery. In these applications, hydrogels are used as 3D cell growth matrices and cell reservoirs. Hence, it has to be underlined that not only materials but also stem cell choice are key points in the regeneration strategies: indeed, researchers are mostly focusing their attention on pluripotent stem cells (embryonic)[59,60] or multipotent ones (mesenchymal or neural).[38,61−69] Natural hydrogels used for this purpose are either synthesized starting from polysaccharides such as alginate[59,66,69] and hyaluronic acid as homopolymer[62,65] or copolymerized with methylcellulose,[61] cellulose,[63] and xyloglucan.[60,67] Other materials are also being investigated to support cell therapies: the commercial Matrigel,[38,69] fibrin,[68] and gelatin.[64] A dedicated mention should be addressed to in vivo studies that already showed functional improvement in animal models after hydrogel implantation. In these studies, hydrogels, such as agarose[70,71] or alginate,[72] were used as scaffolds able to support oriented axonal regeneration. Moreover, with hydrogels being able to provide controlled drug delivery to improve axonal regrowth, they can be loaded with active substances such as chondroitinase ABC,[73] methylprednisolone,[74] or brain-derived neurotrophic factor (BDNF).[62,75]

Table 1. Naturally Derived Hydrogels Used for SCI Repair

material	description	acronym	application in SCI
agarose	polysaccharide		cell growth matrix[74]
			encapsulation and delivery of neurotrophic factors[14]
			controlled chondroitinase delivery[73]
			support for nanoparticle delivery[14,74]
			brain-derived neurotrophic factor (BDNF) controlled delivery[75,76]
			linear guidance (freeze-dried)[70,71]
			cell encapsulation for growthmatrix[37]
	co-methylcellulose	agarose/MC	nerve guidance[77]
alginate	polysaccharide		anisotropic scaffold for axonal regrowth[72]
			neural stem cell groth matrix[66,69,78]
			embryonic stem cell growth matrix[59]
cellulose	polysaccharide		mesenchymal stem cell growth matrix[63]
chitosan	polysaccharide		scaffold for cell adhesion and growth with polylysine[79]
			scaffold for neurite regrowth with hyaluronic acid[80]
collagen	polypeptide		polymeric channels[81]
			filament bridges as growth substances[82]
			cell growth matrix[83,84]
fibrin	linked proteins		neural stem cell growth matrix[68]
gelatin	hydrolyzed collagen		mesenchymal stem cell growth matrix[64]
gellan gum	polysaccharide		tubular, porous scaffold for axonal regrowth[85]
hyaluronic acid	polysaccharide	HA	controlled delivery of neurotrophic factors[62]
			scaffold for neurite regrowth[65,80]
			controlled peptide delivery[26,86,87]
	co-polylysine		Nogo 66 receptor antibody delivery system[88,89]
	co-methylcellulose	HAMC	intrathecal drug and growth factor delivery[90−95]
			neural stem cell carrier for cell therapies[61]
	co-collagen		cell growth matrix[96]
Matrigel	laminin, collagen IV, heparin		scaffold supporting cell adhesion and growth[38]
			neural stem cell carrier for cell therapies[69]
scleroglucan	polysaccharide		controlled drug delivery[97]
xyloglucan	polysaccharide		scaffold supporting cell adhesion and growth[60,67]

2. Synthetic Hydrogels. Synthetic hydrogels, such as those based on poly(hydroxyethyl methacrylate) (PHEMA), were some of the earliest biomaterials used as tissue engineering scaffolds.[43,98] This class of materials shows very important advantages in this field: easier large-scale production and highly tunable properties.[49] Both of them contributed to the large number of formulations. In contraposition with the advantages of the naturally derived hydrogels, synthetic polymers offer wider scope to design and control the characteristics of the material. Moreover, the possibility to reduce the allergenic risks using a completely artificial biocompatible material devoid of animal proteins is evident.[98,99]

The more recent use of hydrogels as cell carriers offers the possibility to provide precise temporal control of the donor and host cell interactions. The ability to carry cells in a matrix, initially impermeable to cells, could afford donor cells protection from potentially harmful substances such as cytotoxic cytokines immediately after transplantation, being a barrier for their diffusion as in the case of hydrogel based microcapsules.[100−102] At later time points, as the gel degrades and the overall mesh size of the gel increases, donor cells will be delivered. Additionally, the gel network can serve as a scaffold to support regeneration within the host environment until the material is ultimately resorbed by the tissue. The surface of the hydrogel could also be easily modified or charged in order to favor cell attachment, or differentiation. They can also be cross-linked with other polymers in a classic block copolymerization in order to design smart delivery systems. Emblematic is the case of cyclodextrin, able to carry insoluble drugs into water based systems. With respect to synthetic formulations, care must be taken to ensure that contaminant and unreacted reagents present during synthesis are completely removed due to their possible toxicity. Details of synthetic polymers used in SCI are presented in Table 2.

Briefly commenting on this second table, being that regenerative medicine is considered the future in life sciences, several studies were performed to develop synthetic polymeric gels showing full compatibility with stem cells. Stem cells are mainly chosen between multipotent cell lines (mesenchymal and neural)[99,103−107] and pluripotent ones (embryonic).[108] Synthetic materials that seem to be extremely suitable as 3D growth matrices are polymethacrylates, such as pHPMA and pHEMA, which were tested with mesenchymal stem cells[103] and also showed relevant improvement in chronic spinal cord injury.[103,109,110] In addition some studies, involving polymethacylates, underlined relevant functional improvements on animals after hydrogel implantation: pHEMA and pHPMA favor axonal ingrowth,[111] showing also good outcome in chronic cases as said before,[103] while pHEMA-MMA influences axonal regrowth.[112−114] Stem

Table 2. Synthetic Based Hydrogels Studied in SCI Research

material	description	acronym	application in SCI
Carbopol	branched poly(acrylic acid)		controlled drug delivery with cyclodextrin[115,116]
lysine-leucine	co-polypeptide	DCH	tunable vehicles for factor delivery[117]
polyacrylamide			scaffold for neurite outgrowth[118]
polyalkylimide	acrylates		scaffold supporting cell adhesion and growth[119]
poly-ε-caprolactone	polyester	PCL	nanofiber for axonal growth orientation[99]
poly(ethylene glycol)	polyether	PEG	3D cell growth matrix[31,104,120−122]
			microcapsules for cell growth[123]
			controlled drug delivery with cyclodextrin[124]
			microvascular networks for cell growth with PLGA[105,107,108]
			controlled delivery of methylprednisolone[125]
		PLA-*b*-PEG-*b*-PLA	delivery of neurotrophins[126−129]
		PNIPAA-PEG	cell adhesion and neurotrophins release[106]
polyethylene oxide		PEO	injectable scaffold for drug delivery with cyclodextrin[130]
poly(hydroxethyl methacrylate)	polyester	PHEMA	charged modified scaffold as bridges for axonal growth[98,111]
			guidance channels[111,131]
			fiber templated scaffold[132]
			bone marrow stem cell carrier for cell therapies[109,110]
	co-methylmetahcrylate	PHEMA-MMA	reinforced guidance channels for nerve regrowth[112−114]
			controlled drug delivery[133]
poly(hydroxypropyl methacrylate)	polyester	PHPMA	mesenchymal stem cell growth matrix[103]
poly(*N*-isopropylacrylamide)-*co*-polyvinylpyrrolidone	copolymer	PNIPAA-PVP	scaffold for controlled drug delivery[134]
Pluronic	polypropylene oxide + ethylene oxide	PF127	scaffold supporting cell adhesion and growth[38,135]
PuraMatrix	oligopeptides		scaffold supporting cell adhesion and growth[38]
polyvynilalchol	acetate	PVA	scaffold for controlled drug delivery[136]

cell studies were conducted also using poly(ethylene glycol) (PEG) scaffold with neural,[106,107] embryonic,[108] or mesenchymal stem cells[104,105] and poly-ε-caprolactone with neural stem cells.[99]

3. Synthetic−Natural Composite Hydrogels. The idea of using natural macromers such as fibronectin, laminin, or agarose in order to coat synthetic polymers to favor cell attachment and viability has been suggested in tissue engineering concepts from its very first description.[17] However, in order to overcome matters of only natural or only synthetic hydrogels, this suggestion was dismissed, but now it has came back on the scene as one of the novelties of the last three years where great importance has been given to composite (synthetic−natural) hydrogels for spinal cord injury repair strategies.[28] They could be the result of a block copolymerization between synthetic and natural macromers, or just an interpolymer complex bonded by physical interactions. The goal of this approach should be to combine the biocompatibility of natural gels with the possibility to tune mechanical and physical properties by the inclusion of synthetic ones.[137] For example, the adhesive properties could be increased by adding polylysine to PEG channels, or chitosan to methacrylamide. This strategy was also studied in order to overcome the disadvantages of the "classic" 3D growth matrices, increasing cell viability and biocompatibility as in the case of agarose−Carbopol or hyaluronan−PEG. In these studies, multipotent stem cell lines happear to be promising great therapeutical advantages. Agarose−Carbopol hydrogels were tested with mesenchymal stem cells,[138] while chitosan−methacrylamide and PEG−polylysine hydrogels were tested with neural ones.[139−141]The complete list of the composite hydrogels used is presented in Table 3.

4. Patented Hydrogels. The field of materials for supporting SCI repair strategies is not only scientifically very rich but also very promising from an industrial point of view. Social and economical impacts of impaired SCI patients are unluckily very well-known, and the possibility to develop therapies toward repair is definitely appealing for the industry. Indeed, patent trends can be used as good indicators reflecting the increasing business interests in a specific technological area. This is illustrated by the fact that about 80% of technical and scientific knowledge generated worldwide is only published as patents and not elsewhere.[151]

Looking into biomaterials for SCI repair strategies, the first patent claiming the use of a hydrogel in SCI repair strategies dates back no further than 1984,[152] while the oldest patent application on a hydrogel was filed on 1966.[153] An ever since cumulated snapshot, taken today, accounts for just 44 patents on hydrogels for SCI repair, out of about 4000 patents relating to hydrogels in general (search performed with QPat). Nevertheless, looking at the filing trend, the tremendous interest displayed in the scientific literature in the last 5 years is also reflected onto intellectual property rights: the same number of patent applications filed from 1984 to 2005 was filed only from 2006 up to today.

The same criteria applied in scientific literature was also used to categorize claims on hydrogels based on their main components: natural, synthetic, or both. As expected, the vast majority (71.5%)

Table 3. Synthetic−Natural Composite Hydrogels Studied in SCI Research

material	description	acronym	application in SCI
Carbopol + agarose	copolymer	AC	3D mesenchymal stem cell growth matrix[138]
			scaffold for controlled drug delivery[142]
Carbopol + chitosan	interpolymer complex	IPC	multiple drug delivery[143]
methacrylamide + chitosan	cross-linked polymer	MC	cell adhesion and neurite penetration[140,144]
			neural stem cell growth matrix[139]
polyglycolic acid + chitosan	interpolymer complex	chitosan/PGA	bridge for neurite regrowth[145]
poly(ethylene glycol) + hyaluronan	interpolymer complex	HA-DTPH-PEGDA	3D growth matrix[146]
poly(ethylene glycol)/polyacrylic acid/agarose	layer	PEG/PAA/agarose	multilayer scaffold for BDNF controlled drug delivery[147]
poly(ethylene glycol) + polylysine	copolymer	PEG/PLL	cell growth matrix[141]
poly(ethylene glycol) + polypeptides	copolymer	PEG/peptide	3D growth matrix[148]
polylactide-*co*-glycolic acid + dex-lactate	interpolymer complex	DP,DS	controlled protein release[149]
tetronic + lactide + heparin	copolymer	TL	bridge, with antiinflammatory agents, for axonal regeneration[150]

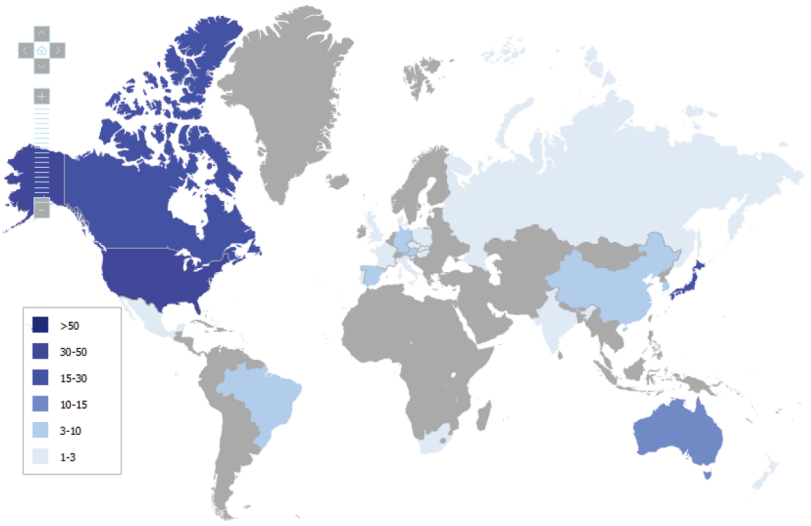


Figure 2. Geographical areas of intellectual property rights protection on hydrogels for SCi repair: darker colors illustrate a higher number of applications.

of applications claims the use of combined natural−synthetic hydrogels,[154−168] while natural[169−172] or synthetic[173,174] hydrogels only accounted for 19% and 9.5% of applications, respectively. The industrial attitude is indeed generally pointing toward the wider possible intellectual property protection, which in this specific field is represented by combined solutions.

Lastly, a statistical study was performed on all applications filed since 1984, to show the main geographic areas of protection. These data are shown in Figure 2 where the darker colors illustrate a higher number of applications.

CONCLUSIONS

It is increasingly recognized that cell or drug therapies alone will not be sufficient for successful tissue engineering in many CNS disorders and insults. For this reason, engineered scaffolds have gained greater interest in the last years. In particular, spinal cord injury for its neuropathological features (loss of neuronal tissue and presence of cavity) represents a good candidate to develop an engineered scaffold able to carry substances (drugs, antibodies, peptides, or other proteins) and/or cells. In this Review, we have given an overview of hydrogels used for experimental SCI repair, since this is an expanding field and most probably will be a useful applicable therapeutic tool in the near future. In this way, medicine and engineering work together to better define the promising therapies using this hybrid knowledge to design and engineer better tissue scaffolds.

AUTHOR INFORMATION

Corresponding Author

*Telephone: +39 02 2399 4743. Fax: +39 02 2399 3180. E-mail: filippo.rossi@mail.polimi.it.

Author Contributions

Dr. G. Perale, Dr. F. Rossi, and Dr. P. Veglianese equally wrote the paper. Dr. S. Bacchiega performed patent research. Prof. Dr. E. Sundstrom, Prof. Dr. M. Masi, and Dr. G. Forloni coordinated the research.

Funding Sources

Supported by Fondazione Cariplo, Grant No. 2010/0639.

■ ACKNOWLEDGMENT

Special thanks are due to Veronica Anceschi and Simone Montalbetti for their help in bibliographical research.

■ REFERENCES

(1) van den Berg, M. E. L., Castellote, J. M., de Pedro-Cuesta, J., and Mahillo-Fernandez, I. (2010) Survival after Spinal Cord Injury: A Systematic Review. *J. Neurotrauma 27*, 1517–1528.

(2) van den Berg, M. E. L., Castellote, J. M., Mahillo-Fernandez, I., and de Pedro-Cuesta, J. (2010) Incidence of Spinal Cord Injury Worldwide: A Systematic Review. *Neuroepidemiology 34*, 184–192.

(3) Abu-Rub, M., McMahon, S., Zeugolis, D. I., Windebank, A., and Pandit, A. (2010) Spinal cord injury in vitro: modelling axon growth inhibition. *Drug Discovery Today 15*, 436–443.

(4) Ditunno, J. F. (2010) Outcome measures: evolution in clinical trials of neurological/functional recovery in spinal cord injury. *Spinal Cord 48*, 674–684.

(5) Steward, O., Schauwecker, P. E., Guth, L., Zhang, Z. Y., Fujiki, M., Inman, D., Wrathall, J., Kempermann, G., Gage, F. H., Saatman, K. E., Raghupathi, R., and McIntosh, T. (1999) Genetic approaches to neurotrauma research: Opportunities and potential pitfalls of murine models. *Exp. Neurol. 157*, 19–42.

(6) Kubinova, S., and Sykova, E. (2010) Nanotechnology for treatment of stroke and spinal cord injury. *Nanomedicine 5*, 99–108.

(7) Kubinova, S., and Sykova, E. (2010) Nanotechnologies in regenerative medicine. *Minimally Invasive Ther. 19*, 144–156.

(8) Fawcett, J. W., and Asher, R. A. (1999) The glial scar and central nervous system repair. *Brain Res. Bull. 49*, 377–391.

(9) McDonald, J. W., Gottlieb, D. I., and Choi, D. W. (2000) What is a functional recovery after spinal cord injury? *Nat. Med. 6*, 358–358.

(10) Bradbury, E. J., and Carter, L. M. (2011) Manipulating the glial scar: Chondroitinase ABC as a therapy for spinal cord injury. *Brain Res. Bull. 84*, 306–316.

(11) Bradbury, E. J., and McMahon, S. B. (2006) Opinion - Spinal cord repair strategies: why do they work? *Nat. Rev. Neurosci. 7*, 644–653.

(12) Cao, K., Huang, L., Liu, J. W., An, H., Shu, Y., and Han, Z. M. (2010) Inhibitory effects of high-dose methylprednisolone on bacterial translocation from gut and endotoxin release following acute spinal cord injury-induced paraplegia in rats. *Neural Regener. Res. 5*, 456–460.

(13) Bracken, M. B., Shepard, M. J., Collins, W. F., Holford, T. R., Young, W., Piepmeier, J., Leosummers, L., Baskin, D. S., Eisenberg, H. M., Flamm, E., Marshall, L. F., Maroon, J., Wilberger, J., Perot, P. L., Sonntag, V. K. H., Wagner, F. C., and Winn, H. R. (1990) a Randomized, Controlled Trial of Methylprednisolone or Naloxone in the Treatment of Acute Spinal-Cord Injury. *New Engl. J. Med. 323*, 1209–1209.

(14) Kim, Y. T., Caldwell, J. M., and Bellamkonda, R. V. (2009) Nanoparticle-mediated local delivery of methylprednisolone after spinal cord injury. *Biomaterials 30*, 2582–2590.

(15) Green, P. S., and Simpkins, J. W. (2000) Neuroprotective effects of estrogens: potential mechanisms of action. *Int. J. Dev. Neurosci. 18*, 347–358.

(16) Langer, R. (2009) Perspectives and Challenges in Tissue Engineering and Regenerative Medicine. *Adv. Mater. 21*, 3235–3236.

(17) Langer, R., and Vacanti, J. P. (1993) Tissue Engineering. *Science 260*, 920–926.

(18) Peppas, N. A., and Langer, R. (1994) New Challenges in Biomaterials. *Science 263*, 1715–1720.

(19) Salewski, R. P. F., Eftekharpour, E., and Fehlings, M. G. (2010) Are Induced Pluripotent Stem Cells the Future of Cell-Based Regenerative Therapies for Spinal Cord Injury? *J. Cell. Physiol. 222*, 515–521.

(20) Stocum, D. L. (2005) Stem cells in CNS and cardiac regeneration. *Regener. Med. I: Theor., Models Methods 93*, 135–159.

(21) Taha, M. F. (2010) Cell based-gene delivery approaches for the treatment of spinal cord injury and neurodegenerative disorders. *Curr. Stem Cell. Res. Ther. 5*, 23–36.

(22) Sakurada, K., McDonald, F. M., and Shimada, F. (2008) Regenerative medicine and stem cell based drug discovery. *Angew. Chem., Int. Ed. 47*, 5718–5738.

(23) Sahni, V., and Kessler, J. A. (2010) Stem cell therapies for spinal cord injury. *Nat. Rev. Neurol. 6*, 363–372.

(24) Little, L., Healy, K. E., and Schaffer, D. (2008) Engineering biomaterials for synthetic neural stem cell microenvironments. *Chem. Rev. 108*, 1787–1796.

(25) Soria, J. M., Ramos, C. M., Sanchez, M. S., Benavent, V., Fernandez, A. C., Ribelles, J. L. G., Verdugo, J. M. G., Pradas, M. M., and Barcia, J. A. (2006) Survival and differentiation of embryonic neural explants on different biomaterials. *J. Biomed. Mater. Res., Part A 79A*, 495–502.

(26) Varghese, O. P., Sun, W. L., Hilborn, J., and Ossipov, D. A. (2009) In Situ Cross-Linkable High Molecular Weight Hyaluronan-Bisphosphonate Conjugate for Localized Delivery and Cell-Specific Targeting: A Hydrogel Linked Prodrug Approach. *J. Am. Chem. Soc. 131*, 8781–8784.

(27) Hellal, F., Hurtado, A., Ruschel, J., Flynn, K. C., Laskowski, C. J., Umlauf, M., Kapitein, L. C., Strikis, D., Lemmon, V., Bixby, J., Hoogenraad, C. C., and Bradke, F. (2011) Microtubule Stabilization Reduces Scarring and Causes Axon Regeneration After Spinal Cord Injury. *Science 331*, 928–931.

(28) Atala, R., Langer, R., Thomson, J., and Nerem, R. (2008) *Principles of regenerative medicine*, Academic Press, Burlington, MA.

(29) Mantsos, T., Chatzistavrou, X., Roether, J. A., Hupa, L., Arstila, H., and Boccaccini, A. R. (2009) Non-crystalline composite tissue engineering scaffolds using boron-containing bioactive glass and poly-(D,L-lactic acid) coatings. *Biomed. Mater. 4*, 055002.

(30) Rezwan, K., Chen, Q. Z., Blaker, J. J., and Boccaccini, A. R. (2006) Biodegradable and bioactive porous polymer/inorganic composite scaffolds for bone tissue engineering. *Biomaterials 27*, 3413–3431.

(31) Hu, B. H., Su, J., and Messersmith, P. B. (2009) Hydrogels Cross-Linked by Native Chemical Ligation. *Biomacromolecules 10*, 2194–2200.

(32) Blaker, J. J., Bismarck, A., Boccaccini, A. R., Young, A. M., and Nazhat, S. N. (2010) Premature degradation of poly(alpha-hydroxyesters) during thermal processing of Bioglass (R)-containing composites. *Acta Biomater. 6*, 756–762.

(33) Nakamatsu, J., Torres, F. G., Troncoso, O. P., Yuan, M. L., and Boccaccini, A. R. (2006) Processing and characterization of porous structures from chitosan and starch for tissue engineering scaffolds. *Biomacromolecules 7*, 3345–3355.

(34) Engel, E., Michiardi, A., Navarro, M., Lacroix, D., and Planell, J. A. (2008) Nanotechnology in regenerative medicine: the materials side. *Trends Biotechnol. 26*, 39–47.

(35) Yu, L., and Ding, J. D. (2008) Injectable hydrogels as unique biomedical materials. *Chem. Soc. Rev. 37*, 1473–1481.

(36) Slaughter, B. V., Khurshid, S. S., Fisher, O. Z., Khademhosseini, A., and Peppas, N. A. (2009) Hydrogels in Regenerative Medicine. *Adv. Mater. 21*, 3307–3329.

(37) Luo, Y., and Shoichet, M. S. (2004) A photolabile hydrogel for guided three-dimensional cell growth and migration. *Nat. Mater. 3*, 249–253.

(38) Thonhoff, J. R., Lou, D. I., Jordan, P. M., Zhao, X., and Wu, P. (2008) Compatibility of human fetal neural stem cells with hydrogel biomaterials in vitro. *Brain Res. 1187*, 42–51.

(39) Schmidt, J. J., Rowley, J., and Kong, H. J. (2008) Hydrogels used for cell-based drug delivery. *J. Biomed. Mater. Res., Part A 87A*, 1113–1122.

(40) Annabi, N., Nichol, J. W., Zhong, X., Ji, C. D., Koshy, S., Khademhosseini, A., and Dehghani, F. (2010) Controlling the Porosity and Microarchitecture of Hydrogels for Tissue Engineering. *Tissue Eng., Part B 16*, 371–383.

(41) Kwon, B. K., Sekhon, L. H., and Fehlings, M. G. (2010) Emerging Repair, Regeneration, and Translational Research Advances for Spinal Cord Injury. *Spine 35*, S263–S270.

(42) Perale, G., Bianco, F., Giordano, C., Matteoli, M., Masi, M., and Cigada, A. (2008) Engineering injured spinal cord with bone

marrow-derived stem cells and hydrogel-based matrices: a glance at the state of the art. *J. Appl. Biomater. Biomech. 6*, 1–8.

(43) Hejcl, A., Lesny, P., Pradny, M., Michalek, J., Jendelova, P., Stulik, J., and Sykova, E. (2008) Biocompatible Hydrogels in Spinal Cord Injury Repair. *Physiol. Res. 57*, S121–S132.

(44) Potter, W., Kalil, R. E., and Kao, W. J. (2008) Biomimetic material systems for neural progenitor cell-based therapy. *Front. Biosci. 13*, 806–821.

(45) Zhong, Y. H., and Bellamkonda, R. V. (2008) Biomaterials for the central nervous system. *J. R. Soc. Interface 5*, 957–975.

(46) Drury, J. L., and Mooney, D. J. (2003) Hydrogels for tissue engineering: scaffold design variables and applications. *Biomaterials 24*, 4337–4351.

(47) Griffith, L. G., and Naughton, G. (2002) Tissue engineering - Current challenges and expanding opportunities. *Science 295*, 1009–1014.

(48) Hoffman, A. S. (2002) Hydrogels for biomedical applications. *Adv. Drug Delivery Rev. 54*, 3–12.

(49) Shoichet, M. S. (2010) Polymer Scaffolds for Biomaterials Applications. *Macromolecules 43*, 581–591.

(50) Malafaya, P. B., Silva, G. A., and Reis, R. L. (2007) Natural-origin polymers as carriers and scaffolds for biomolecules and cell delivery in tissue engineering applications. *Adv. Drug Delivery Rev. 59*, 207–233.

(51) Tabata, Y. (2009) Biomaterial technology for tissue engineering applications. *J. R. Soc. Interface 6*, S311–S324.

(52) Mano, J. F., Silva, G. A., Azevedo, H. S., Malafaya, P. B., Sousa, R. A., Silva, S. S., Boesel, L. F., Oliveira, J. M., Santos, T. C., Marques, A. P., Neves, N. M., and Reis, R. L. (2007) Natural origin biodegradable systems in tissue engineering and regenerative medicine: present status and some moving trends. *J. R. Soc. Interface 4*, 999–1030.

(53) Nisbet, D. R., Crompton, K. E., Horne, M. K., Finkelstein, D. I., and Forsythe, J. S. (2008) Neural tissue engineering of the CNS using hydrogels. *J. Biomed. Mater. Res., Part B 87B*, 251–263.

(54) Hynd, M. R., Turner, J. N., and Shain, W. (2007) Applications of hydrogels for neural cell engineering. *J. Biomater. Sci., Polym. Ed. 18*, 1223–1244.

(55) Chung, H. J., and Park, T. G. (2007) Surface engineered and drug releasing pre-fabricated scaffolds for tissue engineering. *Adv. Drug Delivery Rev. 59*, 249–262.

(56) Willerth, S. M., and Sakiyama-Elbert, S. E. (2007) Approaches to neural tissue engineering using scaffolds for drug delivery. *Adv. Drug Delivery Rev. 59*, 325–338.

(57) Kretlow, J. D., Klouda, L., and Mikos, A. G. (2007) Injectable matrices and scaffolds for drug delivery in tissue engineering. *Adv. Drug Delivery Rev. 59*, 263–273.

(58) von Euler, M., Janson, A. M., Larsen, J. O., Seiger, A., Forno, L., Bunge, M. B., and Sundstrom, E. (2002) Spontaneous axonal regeneration in rodent spinal cord after ischemic injury. *J. Neuropathol. Exp. Neurol. 61*, 64–75.

(59) Shanbhag, M. S., Lathia, J. D., Mughal, M. R., Francis, N. L., Pashos, N., Mattson, M. P., and Wheatley, M. A. (2010) Neural Progenitor Cells Grown on Hydrogel Surfaces Respond to the Product of the Transgene of Encapsulated Genetically Engineered Fibroblasts. *Biomacromolecules 11*, 2936–2943.

(60) Nisbet, D. R., Rodda, A. E., Horne, M. K., Forsythe, J. S., and Finkelstein, D. I. (2010) Implantation of Functionalized Thermally Gelling Xyloglucan Hydrogel Within the Brain: Associated Neurite Infiltration and Inflammatory Response. *Tissue Eng., Part A 16*, 2833–2842.

(61) Hsieh, A., Zahir, T., Lapitsky, Y., Amsden, B., Wan, W. K., and Shoichet, M. S. (2010) Hydrogel/electrospun fiber composites influence neural stem/progenitor cell fate. *Soft Matter 6*, 2227–2237.

(62) Park, J., Lim, E., Back, S., Na, H., Park, Y., and Sun, K. (2010) Nerve regeneration following spinal cord injury using matrix metalloproteinase-sensitive, hyaluronic acid-based biomimetic hydrogel scaffold containing brain-derived neurotrophic factor. *J. Biomed. Mater. Res., Part A 93A*, 1091–1099.

(63) Gu, H. G., Yue, Z. L., Leong, W. S., Nugraha, B., and Tan, L. P. (2010) Control of in vitro neural differentiation of mesenchymal stem cells in 3D macroporous, cellulosic hydrogels. *Regener. Med. 5*, 245–253.

(64) Wang, L. S., Chung, J. E., Chan, P. P. Y., and Kurisawa, M. (2010) Injectable biodegradable hydrogels with tunable mechanical properties for the stimulation of neurogenesic differentiation of human mesenchymal stem cells in 3D culture. *Biomaterials 31*, 1148–1157.

(65) Pan, L. J., Ren, Y. J., Cui, F. Z., and Xu, Q. Y. (2009) Viability and Differentiation of Neural Precursors on Hyaluronic Acid Hydrogel Scaffold. *J. Neurosci. Res. 87*, 3207–3220.

(66) Banerjee, A., Arha, M., Choudhary, S., Ashton, R. S., Bhatia, S. R., Schaffer, D. V., and Kane, R. S. (2009) The influence of hydrogel modulus on the proliferation and differentiation of encapsulated neural stem cells. *Biomaterials 30*, 4695–4699.

(67) Nisbet, D. R., Moses, D., Gengenbach, T. R., Forsythe, J. S., Finkelstein, D. I., and Horne, M. K. (2009) Enhancing neurite outgrowth from primary neurones and neural stem cells using thermoresponsive hydrogel scaffolds for the repair of spinal cord injury. *J. Biomed. Mater. Res., Part A 89A*, 24–35.

(68) Willerth, S. M., Faxel, T. E., Gottlieb, D. I., and Sakiyama-Elbert, S. E. (2007) The effects of soluble growth factors on embryonic stem cell differentiation inside of fibrin scaffolds. *Stem Cells 25*, 2235–2244.

(69) Novikova, L. N., Mosahebi, A., Wiberg, M., Terenghi, G., Kellerth, J. O., and Novikov, L. N. (2006) Alginate hydrogel and matrigel as potential cell carriers for neurotransplantation. *J. Biomed. Mater. Res., Part A 77A*, 242–252.

(70) Stokols, S., Sakamoto, J., Breckon, C., Holt, T., Weiss, J., and Tuszynski, M. H. (2006) Templated agarose scaffolds support linear axonal regeneration. *Tissue Eng. 12*, 2777–2787.

(71) Stokols, S., and Tuszynski, M. H. (2006) Freeze-dried agarose scaffolds with uniaxial channels stimulate and guide linear axonal growth following spinal cord injury. *Biomaterials 27*, 443–451.

(72) Prang, P., Muller, R., Eljaouhari, A., Heckmann, K., Kunz, W., Weber, T., Faber, C., Vroemen, M., Bogdahn, U., and Weidner, N. (2006) The promotion of oriented axonal regrowth in the injured spinal cord by alginate-based anisotropic capillary hydrogels. *Biomaterials 27*, 3560–3569.

(73) Lee, H., McKeon, R. J., and Bellamkonda, R. V. (2010) Sustained delivery of thermostabilized chABC enhances axonal sprouting and functional recovery after spinal cord injury. *Proc. Natl. Acad. Sci. U.S.A. 107*, 3340–3345.

(74) Chvatal, S. A., Kim, Y. T., Bratt-Leal, A. M., Lee, H. J., and Bellamkonda, R. V. (2008) Spatial distribution and acute anti-inflammatory effects of Methylprednisolone after sustained local delivery to the contused spinal cord. *Biomaterials 29*, 1967–1975.

(75) Jain, A., McKeon, R. J., Brady-Kalnay, S. M., and Bellamkonda, R. V. (2011) Sustained Delivery of Activated Rho GTPases and BDNF Promotes Axon Growth in CSPG-Rich Regions Following Spinal Cord Injury. *PloS One 6*, 1–12.

(76) Jain, A., Kim, Y. T., McKeon, R. J., and Bellamkonda, R. V. (2006) In situ gelling hydrogels for conformal repair of spinal cord defects, and local delivery of BDNF after spinal cord injury. *Biomaterials 27*, 497–504.

(77) Zuidema, J. M., Pap, M. M., Jaroch, D. B., Morrison, F. A., and Gilbert, R. J. (2011) Fabrication and characterization of tunable polysaccharide hydrogel blends for neural repair. *Acta Biomater. 7*, 1634–1643.

(78) Ashton, R. S., Banerjee, A., Punyani, S., Schaffer, D. V., and Kane, R. S. (2007) Scaffolds based on degradable alginate hydrogels and poly(lactide-*co*-glycolide) microspheres for stem cell culture. *Biomaterials 28*, 5518–5525.

(79) Crompton, K. E., Goud, J. D., Bellamkonda, R. V., Gengenbach, T. R., Finkelstein, D. I., Horne, M. K., and Forsythe, J. S. (2007) Polylysine-functionalised thermoresponsive chitosan hydrogel for neural tissue engineering. *Biomaterials 28*, 441–449.

(80) Horn, E. M., Beaumont, M., Shu, X. Z., Harvey, A., Prestwich, G. D., Horn, K. M., Gibson, A. R., Preul, M. C., and Panitch, A. (2007) Influence of cross-linked hyaluronic acid hydrogels on neurite outgrowth and recovery from spinal cord injury. *J. Neurosurg.: Spine 6*, 133–140.

(81) Iwata, A., Browne, K. D., Pfister, B. J., Gruner, J. A., and Smith, D. H. (2006) Long-term survival and outgrowth of mechanically engineered nervous tissue constructs implanted into spinal cord lesions. *Tissue Eng. 12*, 101–110.

(82) Pfister, B. J., Iwata, A., Taylor, A. G., Wolf, J. A., Meaney, D. F., and Smith, D. H. (2006) Development of transplantable nervous tissue constructs comprised of stretch-grown axons. *J. Neurosci. Methods 153*, 95–103.

(83) Hiraoka, M., Kato, K., Nakaji-Hirabayashi, T., and Iwata, H. (2009) Enhanced Survival of Neural Cells Embedded in Hydrogels Composed of Collagen and Laminin-Derived Cell Adhesive Peptide. *Bioconjugate Chem. 20*, 976–983.

(84) Semino, C. E., Kasahara, J., Hayashi, Y., and Zhang, S. G. (2004) Entrapment of migrating hippocampal neural cells in three-dimensional peptide nanofiber scaffold. *Tissue Eng. 10*, 643–655.

(85) Silva, N. A., Salgado, A. J., Sousa, R. A., Oliveira, J. T., Pedro, A. J., Leite-Almeida, H., Cerqueira, R., Almeida, A., Mastronardi, F., Mano, J. F., Neves, N. M., Sousa, N., and Reis, R. L. (2010) Development and Characterization of a Novel Hybrid Tissue Engineering-Based Scaffold for Spinal Cord Injury Repair. *Tissue Eng., PartA 16*, 45–54.

(86) Wei, Y. T., He, Y., Xu, C. L., Wang, Y., Liu, B. F., Wang, X. M., Sun, X. D., Cui, F. Z., and Xu, Q. Y. (2010) Hyaluronic acid hydrogel modified with nogo-66 receptor antibody and poly-(L)-lysine to promote axon regrowth after spinal cord injury. *J. Biomed. Mater. Res., Part B 95B*, 110–117.

(87) Wei, Y. T., Tian, W. M., Yu, X., Cui, F. Z., Hou, S. P., Xu, Q. Y., and Lee, I. S. (2007) Hyaluronic acid hydrogels with IKVAV peptides for tissue repair and axonal regeneration in an injured rat brain. *Biomed. Mater. 2*, S142–S146.

(88) Tian, W. M., Zhang, C. L., Hou, S. P., Yu, X., Cui, F. Z., Xu, Q. Y., Sheng, S. L., Cui, H., and Li, H. D. (2005) Hyaluronic acid hydrogel as Nogo-66 receptor antibody delivery system for the repairing of injured rat brain: in vitro. *J. Controlled Release 102*, 13–22.

(89) Hou, S., Tian, W., Xu, Q., Cui, F., Zhang, J., Lu, Q., and Zhao, C. (2006) The enhancement of cell adherence and inducement of neurite outgrowth of dorsal root ganglia co-cultured with hyaluronic acid hydrogels modified with Nogo-66 receptor antagonist in vitro. *Neuroscience 137*, 519–529.

(90) Baumann, M. D., Kang, C. E., Tator, C. H., and Shoichet, M. S. (2010) Intrathecal delivery of a polymeric nanocomposite hydrogel after spinal cord injury. *Biomaterials 31*, 7631–7639.

(91) Wang, Y. F., Lapitsky, Y., Kang, C. E., and Shoichet, M. S. (2009) Accelerated release of a sparingly soluble drug from an injectable hyaluronan-methylcellulose hydrogel. *J. Controlled Release 140*, 218–223.

(92) Baumann, M. D., Kang, C. E., Stanwick, J. C., Wang, Y. F., Kim, H., Lapitsky, Y., and Shoichet, M. S. (2009) An injectable drug delivery platform for sustained combination therapy. *J. Controlled Release 138*, 205–213.

(93) Reynolds, L. F., Bren, M. C., Wilson, B. C., Gibson, G. D., Shoichet, M. S., and Murphy, R. J. L. (2008) Transplantation of porous tubes following spinal cord transection improves hindlimb function in the rat. *Spinal Cord 46*, 58–64.

(94) Shoichet, M. S., Tator, C. H., Poon, P., Kang, C., and Baumann, M. D. (2007) Intrathecal drug delivery strategy is safe and efficacious for localized delivery to the spinal cord. *Prog. Brain Res. 161*, 385–392.

(95) Kang, C. E., Poon, P. C., Tator, C. H., and Shoichet, M. S. (2009) A New Paradigm for Local and Sustained Release of Therapeutic Molecules to the Injured Spinal Cord for Neuroprotection and Tissue Repair. *Tissue Eng., Part A 15*, 595–604.

(96) Brannvall, K., Bergman, K., Wallenquist, U., Svahn, S., Bowden, T., Hilborn, J., and Forsberg-Nilsson, K. (2007) Enhanced neuronal differentiation in a three-dimensional collagen-hyaluronan matrix. *J. Neurosci. Res. 85*, 2138–2146.

(97) Coviello, T., Grassi, M., Palleschi, A., Bocchinfuso, G., Coluzzi, G., Banishoeib, F., and Alhaique, F. (2005) A new scleroglucan/borax hydrogel: swelling and drug release studies. *Int. J. Pharm. 289*, 97–107.

(98) Hejcl, A., Lesny, P., Pradny, M., Sedy, J., Zamecnik, J., Jendelova, P., Michalek, J., and Sykova, E. (2009) Macroporous hydrogels based on 2-hydroxyethyl methacrylate. Part 6: 3D hydrogels with positive and negative surface charges and polyelectrolyte complexes in spinal cord injury repair. *J. Mater. Sci.: Mater. Med. 20*, 1571–1577.

(99) Horne, M. K., Nisbet, D. R., Forsythe, J. S., and Parish, C. L. (2010) Three-Dimensional Nanofibrous Scaffolds Incorporating Immobilized BDNF Promote Proliferation and Differentiation of Cortical Neural Stem Cells. *Stem Cells Dev. 19*, 843–852.

(100) Mallett, A. G., and Korbutt, G. S. (2009) Alginate Modification Improves Long-Term Survival and Function of Transplanted Encapsulated Islets. *Tissue Eng., Part A 15*, 1301–1309.

(101) Wilson, J. T., and Chaikof, E. L. (2008) Challenges and emerging technologies in the immunoisolation of cells and tissues. *Adv. Drug Delivery Rev. 60*, 124–145.

(102) Kulseng, B., Thu, B., Espevik, T., and SkjakBraek, G. (1997) Alginate polylysine microcapsules as immune barrier: Permeability of cytokines and immunoglobulins over the capsule membrane. *Cell Transplant. 6*, 387–394.

(103) Hejcl, A., Sedy, J., Kapcalova, M., Toro, D. A., Amemori, T., Lesny, P., Likavcanova-Masinova, K., Krumbholcova, E., Pradny, M., Michalek, J., Burian, M., Hajek, M., Jendelova, P., and Sykova, E. (2010) HPMA-RGD Hydrogels Seeded with Mesenchymal Stem Cells Improve Functional Outcome in Chronic Spinal Cord Injury. *Stem Cells Dev. 19*, 1535–1546.

(104) Lampe, K. J., Mooney, R. G., Bjugstad, K. B., and Mahoney, M. J. (2010) Effect of macromer weight percent on neural cell growth in 2D and 3D nondegradable PEG hydrogel culture. *J. Biomed. Mater. Res., Part A 94A*, 1162–1171.

(105) Ali, O. A., Huebsch, N., Cao, L., Dranoff, G., and Mooney, D. J. (2009) Infection-mimicking materials to program dendritic cells in situ. *Nat. Mater. 8*, 151–158.

(106) Comolli, N., Neuhuber, B., Fischer, I., and Lowman, A. (2009) In vitro analysis of PNIPAAm-PEG, a novel, injectable scaffold for spinal cord repair. *Acta Biomater. 5*, 1046–1055.

(107) Rauch, M. F., Hynes, S. R., Bertram, J., Redmond, A., Robinson, R., Williams, C., Xu, H., Madri, J. A., and Lavik, E. B. (2009) Engineering angiogenesis following spinal cord injury: a coculture of neural progenitor and endothelial cells in a degradable polymer implant leads to an increase in vessel density and formation of the blood-spinal cord barrier. *Eur. J. Neurosci. 29*, 132–145.

(108) Rauch, M. F., Michaud, M., Xu, H., Madri, J. A., and Lavik, E. B. (2008) Co-culture of primary neural progenitor and endothelial cells in a macroporous gel promotes stable vascular networks in vivo. *J. Biomater. Sci., Polym. Ed. 19*, 1469–1485.

(109) Sykova, E., Jendelova, P., Hejcl, A., Kozubenko, N., and Amemori, T. (2010) Stem Cells and Hydrogel Bridges for the Treatment of Acute and Chronic Spinal Cord Injury. *Cell Transplant. 19*, 366–366.

(110) Sykova, E., Jendelova, P., Urdzikova, L., Lesny, P., and Hejcl, A. (2006) Bone marrow stem cells and polymer hydrogels-two strategies for spinal cord injury repair. *Cell. Mol. Neurobiol. 26*, 1113–1129.

(111) Hejcl, A., Urdzikova, L., Sedy, J., Lesny, P., Pradny, M., Michalek, J., Burian, M., Hajek, M., Zamecnik, J., Jendelova, P., and Sykova, E. (2008) Acute and delayed implantation of positively charged 2-hydroxyethyl methacrylate scaffolds in spinal cord injury in the rat. *J. Neurosurg.: Spine 8*, 67–73.

(112) Katayama, Y., Montenegro, R., Freier, T., Midha, R., Belkas, J. S., and Shoichet, M. S. (2006) Coil-reinforced hydrogel tubes promote nerve regeneration equivalent to that of nerve autografts. *Biomaterials 27*, 505–518.

(113) Nomura, H., Katayama, Y., Shoichet, M. S., and Tator, C. H. (2006) Complete spinal cord transection treated by implantation of a reinforced synthetic hydrogel channel results in syringomyelia and caudal migration of the rostral stump. *Neurosurgery 59*, 183–192.

(114) Tsai, E. C., Dalton, P. D., Shoichet, M. S., and Tator, C. H. (2006) Matrix inclusion within synthetic hydrogel guidance channels improves specific supraspinal and local axonal regeneration after complete spinal cord transection. *Biomaterials 27*, 519–533.

(115) Rodriguez-Tenreiro, C., Alvarez-Lorenzo, C., Rodriguez-Perez, A., Concheiro, A., and Torres-Labandeira, J. J. (2007) Estradiol sustained

release from high affinity cyclodextrin hydrogels. *Eur. J. Pharm. Biopharm. 66*, 55–62.

(116) Rodriguez-Tenreiro, C., Diez-Bueno, L., Concheiro, A., Torres-Labandeira, J. J., and Alvarez-Lorenzo, C. (2007) Cyclodextrin/Carbopol micro-scale interpenetrating networks (ms-IPNs) for drug delivery. *J. Controlled Release 123*, 56–66.

(117) Yang, C. Y., Song, B. B., Ao, Y., Nowak, A. P., Abelowitz, R. B., Korsak, R. A., Havton, L. A., Deming, T. J., and Sofroniew, M. V. (2009) Biocompatibility of amphiphilic diblock copolypeptide hydrogels in the central nervous system. *Biomaterials 30*, 2881–2898.

(118) Jiang, F. X., Yurke, B., Firestein, B. L., and Langrana, N. A. (2008) Neurite outgrowth on a DNA crosslinked hydrogel with tunable stiffnesses. *Ann. Biomed. Eng. 36*, 1565–1579.

(119) Ramires, P. A., Miccoli, M. A., Panzarini, E., Dini, L., and Protopapa, C. (2005) In vitro and in vivo biocompatibility evaluation of a polyalkylimide hydrogel for soft tissue augmentation. *J. Biomed. Mater. Res., Part B 72B*, 230–238.

(120) Skornia, S. L., Bledsoe, J. G., Kelso, B., and Kuntzwillits, R. (2007) Mechanical properties of layered poly (ethylene glycol) gels. *J. Appl. Biomater. Biomech. 5*, 176–183.

(121) Mahoney, M. J., and Anseth, K. S. (2006) Three-dimensional growth and function of neural tissue in degradable polyethylene glycol hydrogels. *Biomaterials 27*, 2265–2274.

(122) Krsko, P., McCann, T. E., Thach, T. T., Laabs, T. L., Geller, H. M., and Libera, M. R. (2009) Length-scale mediated adhesion and directed growth of neural cells by surface-patterned poly(ethylene glycol) hydrogels. *Biomaterials 30*, 721–729.

(123) Koh, W. G., Revzin, A., and Pishko, M. V. (2002) Poly-(ethylene glycol) hydrogel microstructures encapsulating living cells. *Langmuir 18*, 2459–2462.

(124) Salmaso, S., Sernenzato, A., Bersani, S., Matricardi, P., Rossi, F., and Caliceti, P. (2007) Cyclodextrin/PEG based hydrogels for multi-drug delivery. *Int. J. Pharm. 345*, 42–50.

(125) Pritchard, C. D., O'Shea, T. M., Siegwart, D. J., Calo, E., Anderson, D. G., Reynolds, F. M., Thomas, J. A., Slotkin, J. R., Woodard, E. J., and Langer, R. (2011) An injectable thiol-acrylate poly(ethylene glycol) hydrogel for sustained release of methylprednisolone sodium succinate. *Biomaterials 32*, 587–597.

(126) Burdick, J. A., Ward, M., Liang, E., Young, M. J., and Langer, R. (2006) Stimulation of neurite outgrowth by neurotrophins delivered from degradable hydrogels. *Biomaterials 27*, 452–459.

(127) Piantino, J., Burdick, J. A., Goldberg, D., Langer, R., and Benowitz, L. I. (2006) An injectable, biodegradable hydrogel for trophic factor delivery enhances axonal rewiring and improves performance after spinal cord injury. *Exp. Neurol. 201*, 359–367.

(128) Metters, A. T., Anseth, K. S., and Bowman, C. N. (2000) Fundamental studies of a novel, biodegradable PEG-b-PLA hydrogel. *Polymer 41*, 3993–4004.

(129) Metters, A. T., Bowman, C. N., and Anseth, K. S. (2001) Verification of scaling laws for degrading PLA-b-PEG-b-PLA hydrogels. *AIChE J. 47*, 1432–1437.

(130) Li, J., Ni, X. P., and Leong, K. W. (2003) Injectable drug-delivery systems based on supramolecular hydrogels formed by poly(ethylene oxide) and alpha-cyclodextrin. *J. Biomed. Mater. Res., Part A 65A*, 196–202.

(131) Bakshi, A., Fisher, O., Dagci, T., Himes, B. T., Fischer, I., and Lowman, A. (2004) Mechanically engineered hydrogel scaffolds for axonal growth and angiogenesis after transplantation in spinal cord injury. *J. Neurosurg.: Spine 1*, 322–329.

(132) Carone, T. W., and Hasenwinkel, J. M. (2006) Mechanical and morphological characterization of homogeneous and bilayered poly(2-hydroxyethyl methacrylate) scaffolds for use in CNS nerve regeneration. *J. Biomedical Mater. Res., Part B 78B*, 274–282.

(133) Piotrowicz, A., and Shoichet, M. S. (2006) Nerve guidance channels as drug delivery vehicles. *Biomaterials 27*, 2018–2027.

(134) Geever, L. M., Cooney, C. C., Lyons, J. G., Kennedy, J. E., Nugent, M. J. D., Devery, S., and Higginbotham, C. L. (2008) Characterisation and controlled drug release from novel drug-loaded hydrogels. *Eur. J. Pharm. Biopharm. 69*, 1147–1159.

(135) Strappe, P. M., Hampton, D. W., Cachon-Gonzalez, B., Fawcett, J. W., and Lever, A. (2005) Delivery of a lentiviral vector in a Pluronic F127 gel to cells of the central nervous system. *Eur. J. Pharm. Biopharm. 61*, 126–133.

(136) Gander, B., Gurny, R., Doelker, E., and Peppas, N. A. (1989) Effect of Polymeric Network Structure on Drug Release from Cross-Linked Polyvinyl-Alcohol) Micromatrices. *Pharm. Res. 6*, 578–584.

(137) Bhattarai, N., Li, Z. S., Gunn, J., Leung, M., Cooper, A., Edmondson, D., Veiseh, O., Chen, M. H., Zhang, Y., Ellenbogen, R. G., and Zhang, M. Q. (2009) Natural-Synthetic Polyblend Nanofibers for Biomedical Applications. *Adv. Mater. 21*, 2792–2797.

(138) Perale, G., Giordano, C., Bianco, F., Rossi, F., Tunesi, M., Daniele, F., Crivelli, F., Matteoli, M., and Masi, M. (2011) Hydrogel for Cell Housing in the Brain and in the Spinal Cord. *Int. J. Artif. Organs 34*, 295–303.

(139) Leipzig, N. D., and Shoichet, M. S. (2009) The effect of substrate stiffness on adult neural stem cell behavior. *Biomaterials 30*, 6867–6878.

(140) Leipzig, N. D., Wylie, R. G., Kim, H., and Shoichet, M. S. (2011) Differentiation of neural stem cells in three-dimensional growth factor-immobilized chitosan hydrogel scaffolds. *Biomaterials 32*, 57–64.

(141) Hynes, S. R., McGregor, L. M., Rauch, M. F., and Lavik, E. B. (2007) Photopolymerized poly(ethylene glycol)/poly(L-lysine) hydrogels for the delivery of neural progenitor cells. *J. Biomater. Sci., Polym. Ed. 18*, 1017–1030.

(142) Perale, G., Veglianese, P., Rossi, F., Peviani, M., Santoro, M., Llupi, D., Micotti, E., Forloni, G., and Masi, M. (2011) In situ agar - carbomer polycondensation: a chemical approach to regenerative medicine. *Mater. Lett. 65*, 1688–1692.

(143) Park, S. H., Chun, M. K., and Choi, H. K. (2008) Preparation of an extended-release matrix tablet using chitosan/Carbopol interpolymer complex. *Int. J. Pharm. 347*, 39–44.

(144) Yu, L. M. Y., Kazazian, K., and Shoichet, M. S. (2007) Peptide surface modification of methacrylamide chitosan for neural tissue engineering applications. *J. Biomed. Mater. Res., Part A 82A*, 243–255.

(145) Wang, X. D., Hu, W., Cao, Y., Yao, J., Wu, J., and Gu, X. S. (2005) Dog sciatic nerve regeneration across a 30-mm defect bridged by a chitosan/PGA artificial nerve graft. *Brain 128*, 1897–1910.

(146) Shu, X. Z., Liu, Y. C., Palumbo, F. S., Lu, Y., and Prestwich, G. D. (2004) In situ crosslinkable hyaluronan hydrogels for tissue engineering. *Biomaterials 25*, 1339–1348.

(147) Mehrotra, S., Lynam, D., Maloney, R., Pawelec, K. M., Tuszynski, M. H., Lee, I., Chan, C., and Sakamoto, J. (2010) Time Controlled Protein Release from Layer-by-Layer Assembled Multilayer Functionalized Agarose Hydrogels. *Adv. Funct. Mater. 20*, 247–258.

(148) DeForest, C. A., Sims, E. A., and Anseth, K. S. (2010) Peptide-Functionalized Click Hydrogels with Independently Tunable Mechanics and Chemical Functionality for 3D Cell Culture. *Chem. Mater. 22*, 4783–4790.

(149) de Jong, S. J., van Eerdenbrugh, B., van Nostrum, C. F., Kettenes-van de Bosch, J. J., and Hennink, W. E. (2001) Physically crosslinked dextran hydrogels by stereocomplex formation of lactic acid oligomers: degradation and protein release behavior. *J. Controlled Release 71*, 261–275.

(150) Kang, Y. M., Hwang, D. H., Kim, B. G., Go, D. H., and Park, K. D. (2010) Thermosensitive Polymer-based Hydrogel Mixed with the Anti-inflammatory Agent Minocycline Induces Axonal Regeneration in Hemisected Spinal Cord. *Macromol. Res. 18*, 399–403.

(151) Bregonje, M. (2005) Patents: A unique source for scientific technical information in chemistry related industry? In *World Patent Information* (Blackman, M., Ed.), Vol. 27, issue 4, pp 309−315, Elsevier, San Diego, CA.

(152) Eckenhoff, J. B., Theeuwes, F., and Deters, J. C. (1984) *Dispenser comprising inner and outer walls functioning as cooperative unit*, Alza Corporation. Patent US4663149.

(153) Kliment, K., Vacik, J., Ott, Z., Majkus, V., Stoy, V., Stol, M., and Wichterle, O. (1966) *Carrier for biologically active substances*, Czechoslovak Academy of Science. Patent CA836532.

(154) Giammona, G. and Mandracchia, D. (2004) *Anionic hydrogel matrices with pH dependent modified release as drug carriers*, Sigma Tau

Industrie Farmaceutiche Riunite e Sigma Tau Industry Farmaceutiche Riunite. Patent WO2005/094792.

(155) Deming, T. J., Sofroniew, M. V., Yang, C. Y., Song, B. B., and Ao, Y. (2009) *Synthetic diblock copolypeptide hydrogels for use in the central nervous system*, University of California. Patent WO2010/096572.

(156) Kim, M. S., Lee, H. B., Lee, J. Y., Ahn, H. H., Lee, J. H., and Kim, K. S. (2008) *Development of a tissue - engineered scaffold for nerve regeneration using a biocompatible and injectabe hydrogel*, Korea Research Institute of Chemical Technology. Patent US2010/040660.

(157) Daniele, F., Giordano, C., Masi, M., Perale, G., Rossi, F., and Tunesi, M. (2008) *Hydrogel capable of containing and conveying cells*, Politecnico di Milano. Patent WO2009/144569.

(158) Cederna, P. S., Egeland, B. M., Abidian, M. R., Peramo, A., Urbancheck, M. G., Kipke, D. A., Richardson-Burns, S., and Martin, D. C. (2008) *Hybrid bioelectrical interface device*, University of Michigan. Patent WO2010/011386.

(159) Hoke, A., Lim, S. H., Liu, X., and Mao, H. (2008) *Biodegradable nerve guides*, Johns Hopkins University. Patent WO2009/094225.

(160) Gormans, J. R. (2006) *Novel Regimens for Treating Diseases and Disorders*, Bioassets Development. Patent US2010/0047235.

(161) Sukuru, K. (2005) *Lipophilic vehicle-based dual controlled release matrix system as capsule fill*, Banner Pharmacaps. Patent WO2007/050724.

(162) Sukuru, K. (2005) *Hydrophilic vehicle-based dual controlled release matrix system as capsule fill*, Banner Pharmacaps. Patent WO2007/050975.

(163) Stein, S. and Sundaram, P. (2005) *Detoxification depot for Alzheimer's disease*, Senicure. Patent WO2007/047967.

(164) Lelkes, P. I., Li, M., Perets, A., Poblete, H., and Lazarovici, P. (2005) *Three-dimensional ccaffolds for tissue engineering made by processing complex extracts of natural extracellular matrices*, Drexel University. Patent WO2006/138718.

(165) Sinko, P. J., Stein, S., and Lalloo, A. (2004) *Controlled release hydrogels*, Rutgers the State University of New Jersey. Patent WO2006/069344.

(166) Tyagi, P., Chancellor, M. B., Li, Z., Chuang, Y. C., De Groat, W. C., Yoshimira, N., Fraser, M. O., and Huang, L. (2004) *Use of lipid and hydrogel vehicles for treatment and drug delivery*, University of Pittsburg. Patent WO2006/065234.

(167) Oray, S., Majewska, A. K., Sur, M., and Teng, Y. (2004) *Compositions and methods for enhancing structural and functional nervous system reorganization and recovery*, Brigham & Womens Hospital, Massachusetts Institute of Technology. Patent WO2006/023530.

(168) Romero-Ortega, M. I. and Galvan-Garcia, P. (2003) *Biomimetic synthetic nerve implant casting device*, Texas Scottish Rite Hospital. Patent US2007/100358.

(169) Kramer, B. C. and Herzberg, U. (2008) *Systemically and locally administered cells for neuropathic pain*, Ethicon. Patent WO2010/071862.

(170) Gilbert, R. J., Martin, B. C., Pap, M. M. and Minner, E. J. (2008) *Novel hydrogel compositions and methods of using*, Michigan Technological University. Patent US2010/112014.

(171) Font, P. J., Del Olmo, B. M., Castro, F. M. B., Infante, M. A., Alonso, V. A. I. and Palomares, C. T. (2008) *New biomaterial based on Wharton's jelly from the human umbilical cord*, Histocell. Patent WO2010/040865.

(172) Gupta, D., Shoichet, M. S., and Tator, C. H. (2005) *Blends of temperature sensitive and anionic polymers for drug delivery*. Patent US2006/280797.

(173) Pritchard, C. D., Langer, R., Reynolds, L. F., and Woodard, E. J. (2008) .*Spinal cord injury, inflammation, and immune disease: local controlled release of therapeutic agents*, Invivo Therapeutics. Patent WO2010/036961.

(174) Cauller, L. and Weiner, R. (2003) *Microtransponder array for implant microtransponder*, University of Texas. Patent US2009/198293.