Regenerative Therapy 3 (2016) 63–67

Contents lists available at ScienceDirect

Regenerative Therapy

journal homepage: http://www.elsevier.com/locate/reth

Review

Biomaterial-engineering and neurobiological approaches for regenerating the injured cerebral cortex

Itsuki Ajioka [a, b, *]

[a] Center for Brain Integration Research (CBIR), Tokyo Medical and Dental University (TMDU), Tokyo 113-8510, Japan
[b] The Japan Science and Technology Agency (JST), Precursory Research for Embryonic Science and Technology (PRESTO), 4-1-8 Honcho, Kawaguchi, Saitama 332-0012, Japan

ARTICLE INFO

Article history:
Received 26 November 2015
Received in revised form
8 January 2016
Accepted 12 February 2016

Keywords:
Cerebral cortex
Regeneration
Cell cycle
Biomaterials

ABSTRACT

The cerebral cortex is responsible for higher functions of the central nervous system (CNS), such as movement, sensation, and cognition. When the cerebral cortex is severely injured, these functions are irreversibly impaired. Although recent neurobiological studies reveal that the cortex has the potential for regeneration, therapies for functional recovery face some technological obstacles. Biomaterials have been used to evoke regenerative potential and promote regeneration in several tissues, including the CNS. This review presents a brief overview of new therapeutic strategies for cortical regeneration from the perspectives of neurobiology and biomaterial engineering, and discusses a promising technology for evoking the regenerative potential of the cerebral cortex.

Contents

1. Introduction

The cerebral cortex, one of the most highly evolved tissues in the body, is responsible for higher functions of the central nervous system (CNS), such as movement, sensation, and cognition. These higher functions are associated with complex but well-organized neuronal networks in the cerebral cortex, which is organized into layers and columns that receive inputs from and send outputs to the subcortical and other cortical areas. Over a century ago, neuroanatomists began investigating and describing the formation of

Abbreviations: CNS, central nervous system; ES, embryonic stem; MSC, mesenchymal stem cell; MZ, marginal zone; NSC, neural stem cell; OB, olfactory bulb; V–SVZ, ventricular–subventricular zone; VZ, ventricular zone.

* Center for Brain Integration Research, Tokyo Medical and Dental University, 1-5-45 Yushima, Tokyo 113-8510, Japan. Fax: +81 3 5803 4716.
E-mail address: iajioka@ams.kuramae.ne.jp.

Peer review under responsibility of the Japanese Society for Regenerative Medicine.

http://dx.doi.org/10.1016/j.reth.2016.02.002

the cerebral cortex. Ramón y Cajal, a pioneer in neuroanatomy research, believed that the regenerative property of the adult brain is extremely limited [1], and researchers since have assumed that the cerebral cortex and other CNS tissues do not regenerate after injury probably because of the following reasons. 1) Most of the neurons are generated from neuronal stem/progenitor cells during developmental stages except for the specific types of neurons generated from adult neural stem cells in the adult ventricular–subventricular zone (V–SVZ) of the lateral ventricle and the dentate gyrus of the hippocampus. 2) Many neural circuits are shaped during a critical period in early postnatal life and seem to largely lose plasticity after this period. Indeed, there are no clinically available therapies for regenerating the injured cerebral cortex. However, recent advances in neurobiology reveal that the cerebral cortex does have regenerative potential, and shed light on new therapeutic strategies for cortical regeneration. In this review, I will describe treatment strategies for regenerating the cerebral cortex after injury from the perspectives of neurobiology and biomaterial engineering, and identify technical limitations that these strategies must address.

2. Neurobiological approaches for cortical regeneration

2.1. Neural cell transplantation

During cortical development, cortical excitatory neurons and inhibitory neurons are generated from neural stem/progenitor cells in the cerebral cortex and the basal ganglia, respectively [2]. These immature neurons differentiate and form a 6-layered structure, based mostly on their network patterns. Excitatory neurons are generated from neural stem/progenitor cells in the cortical ventricular zone (VZ), which is quite far from the position where the neurons will eventually align and take their place in the cortical structure. Newly generated neurons migrate toward the brain surface, to the region beneath the marginal zone (MZ). Later-born neurons pass early-born neurons and reach their destination beneath the MZ first, to align at the upper layer. The upper-layer neurons extend their axons mainly into the intracortical area, while neurons in deeper layers extend mainly into the subcortical area. Cortical interneurons also align roughly according to their order of birth.

Since specific gene-expression patterns contribute to generating different types of neurons [3], it is theoretically possible to manipulate neural stem/progenitor cells *in vitro* to obtain specific types of neurons. Although the developmental process of the cerebral cortex is more complex than that of the retina, the retina provides a good model for studying the fundamental processes of CNS development. Photoreceptor cells, a type of retinal neuron, can be generated from pluripotent stem cells [4,5]. Since the degeneration of photoreceptor cells causes vision loss, transplanting photoreceptor cells into the retina is a promising strategy for retinal regeneration. Transplanted post-mitotic, immature photoreceptor cells integrate into the recipient retina more efficiently than do retinal progenitor cells [6]. When normal immature photoreceptor cells are transplanted into the *Gnat1−/−* mouse retina, in which photoreceptor cells degenerate, vision is recovered [7]. However, since there are no established techniques for generating specific types of cortical neurons, regenerative strategies for the cerebral cortex generally involve transplanting cortical progenitor cells rather than immature cortical neurons.

When neural stem/progenitor cells are transplanted into the developing cortex, their progeny neurons align in the correct layer [8]; this is also the case when progenitor cells are transplanted into the injured cortex [9]. Although it is not presently possible to generate specific types of cortical neurons from pluripotent stem cells, it is possible to generate transplantable neural stem/progenitor cells [10,11]. Transplanted neural stem/progenitor cells derived from human fetal brain tissue and induced pluripotent stem (iPS) cells can differentiate into neurons and survive in the ischemic brain [12,13]. The use of multi-potent mesenchymal stem cells (MSCs) is another attractive strategy, since MSCs are easily isolated from bone marrow [14]. Transplanted MSCs can migrate and differentiate into neurons in the recipient cerebral cortex [15]. Functional recovery has been observed after transplanting embryonic cortical cells [16], neuronal cell line cells [17], neural stem/progenitor cells derived from iPS cells [18], or MSCs [19] into the injured cortex. However, it is not certain whether this functional recovery was caused by new neuronal networks of transplanted cells themselves or by the trophic factors secreted from transplanted cells, which enhances new neuronal networks of existing neurons in the recipient cortex. Thus, we must identify the factors that specifically contribute to functional recovery. For example, abrogating the transplanted cells after functional recovery will reveal whether recipient mice require ongoing support from these cells. Techniques for generating specific types of cortical neurons must also be established to obtain more effective transplantable cells.

2.2. Endogenous adult neural stem cells (NSCs)

The discovery of neural stem cells in the adult V–SVZ of the lateral ventricle and the dentate gyrus of the hippocampus made it possible to consider regenerative strategies using endogenous NSCs [20–23]. In the normal adult, neuroblasts generated at the V–SVZ migrate into the olfactory bulb (OB), where they differentiate into mature neurons as part of the olfactory system [24]. When the cerebral cortex or striatum is injured, these neuroblasts migrate toward the injured area [25–30]. Therapies using endogenous NSCs do not require cell transplantation and thus have advantages with respect to ethical issues and the immune response. However, some technical issues need to be resolved before endogenous NSCs can be used effectively to promote functional recovery. For instance, it is not clear whether these neuroblasts have the potential to differentiate into cortical neurons, or whether NSCs and neuroblasts can generate enough neurons to promote functional recovery. Since self-recovery is not sufficient for functional brain regeneration, the effective use of endogenous NSCs to regenerate the cerebral cortex will require a breakthrough technique, such as a method for expanding NSCs and neuroblasts *in vivo*.

2.3. Cell cycle

Neurons are believed to be post-mitotic and non-dividing because of a fundamental principle in developmental biology: cell differentiation and proliferation are mutually exclusive [31]. In general, when differentiated cells proliferate, they de-differentiate before advancing their cell cycle. Cell differentiation and proliferation are tightly coupled in neurons. Although neurons can advance their cell cycle in pathological situations such as ischemia and Alzheimer's disease, they undergo cell death after re-entering the cell cycle [32]. Techniques to uncouple proliferation from neuronal differentiation might contribute to alternative cortical regeneration therapies, and recent findings in cell-cycle mechanics and neurobiology suggest new strategies for developing such techniques.

One cell becomes two cells after cell-cycle events such as DNA replication in the S phase and cell division in the M phase. The G1 and the G2 phases are gap phases that prepare the cell to advance into the S and the M phases, respectively. During the G1 phase, retinoblastoma protein (Rb) and its family members (p107 and

p130) function as a brake to prevent the cell from entering the S phase [33,34]. During G1 to S phase progression, cyclin D and cyclin-dependent kinase (Cdk) 4/6 complex phosphorylates Rb family members, which induce the dissociation of the Rb family from E2F transcriptional factors and the S phase-related gene to express. The loss of Rb in post-mitotic neurons induces cell-cycle reentry and causes cell death, probably because of the tight coupling of proliferation and neuronal differentiation [35].

Rb-family members also control the tight coupling of neuronal differentiation and proliferation in progenitor but not post-mitotic cells. In addition to directly regulating the transcription of cell-cycle-related genes, the Rb family regulates epigenetic modifications by associating with various epigenetic modifiers [31,33]. In human retinoblastoma, *RB1*-mediated epigenetic regulation contributes to cancer progression [36]. During cancer progression, retinoblastoma cells co-express genes that are normally expressed in only one retinal-cell type [37]; thus, the loss of the Rb family in retinal progenitors can uncouple proliferation from neuronal differentiation by epigenetic dysregulation. Indeed, when neurons lose the tight coordination between cell-cycle exit and neuronal differentiation by the loss of the Rb family in progenitor cells, neurons can divide in some cases. For example, retinal progenitor cells lacking Rb-family members other than one *p107* allele fully differentiate into horizontal interneurons [38]. However, p107-single ($Rb^{-/-}$; $p107^{+/-}$; $p130^{-/-}$) differentiated horizontal interneurons proliferate without de-differentiation and form tumors. In the mouse cerebral cortex, the loss of the Rb family or just *Rb* in progenitors causes immature neurons to divide [39–41]. However, the loss of the Rb family in post-mitotic neurons immediately after exiting the cell cycle does not cause cell division, even though the cells undergo S-phase progression [39]. Thus, once progenitor daughter cells exit their cell-cycle and initiate neuronal differentiation, they become tightly protected from undergoing cell division, and thereby maintain mitotic resistance even after acute Rb family inactivation. These findings lead us to speculate that neurons in the S phase may escape cell death and divide by overriding such mitotic safeguards. Importantly, in pathological situations, postmitotic neurons can advance their cell cycle, but then undergo cell death instead of cell division. For example, in pathological situations such as Alzheimer's disease (AD), Parkinson's disease (PD), and mouse stroke model, neurons advance their cell-cycle into the S phase and then undergo cell death [42–45]. Since the inhibition of cell-cycle reentry by Cdk inhibitor prevents pathological neuron death [46], cell-cycle reentry contributes to disease progression. Moreover, Rb phosphorylation is correlated with the cell-cycle reentry of pathological neurons [47,48] and the progression of AD [49,50]. Thus, the safeguard against cell division may prevent neurons from entering the M phase but instead to lead cell death subsequent to S-phase progression. Since pathological neurons undergo cell death after re-entering the cell cycle, the transient inactivation of such mitotic safeguards and later the inactivation of Cdk to prevent multiple cell cycle reentry is an attractive regenerative strategy to replace dying neurons with new neurons.

3. Biomaterial-engineering approaches to cortical regeneration

3.1. Biomaterials without cell transplantation

Although recent neurobiological studies show that the injured CNS has the potential to regenerate, self-recovery is not sufficient for functional regeneration and recovery. Thus, biomaterials engineered to accelerate regeneration are an important strategy for functional recovery. Potential regenerative strategies using endogenous NSCs face a technical obstacle: the migrating neuroblasts cannot align in the cortical lesion because they do not pass through the glial-scar barrier formed by reactive astrocytes in response to brain injury. Since migrating neuroblasts generated from endogenous NSCs use blood vessels as a scaffold to move toward a brain lesion [51–53], blood-vessel-like biomaterials can be used as a scaffold to allow migrating neuroblasts to pass through the glial-scar barrier. We recently developed a laminin-rich sponge material for the three-dimensional (3D) proliferation culture of *p107*-single horizontal interneurons [54]. Since laminin is a primary component of the extracellular matrix in the basement membrane on the surface of blood vessels, we transplanted the laminin-rich sponge material into the injured cerebral cortex [55]. As expected, neuroblasts migrated into the cortical lesion along the laminin-rich sponge material. Thus, blood-vessel-like biomaterials can support neuroblast migration, which is important for cortical regeneration. Other transplanted biomaterials are reported to enhance the regenerative process [56–58]. Furthermore, engineered biomaterials that release a bioactive factor to promote the formation of new blood vessels may promote neuronal migration and survival, and biomaterials that release a bioactive factor to enhance neuronal maturation may enhance neuronal connections to other areas of the brain.

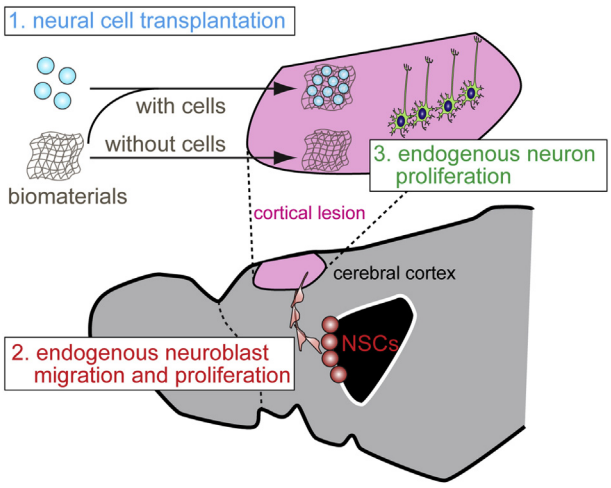

Fig. 1. Neurobiological and biomaterial-engineering approaches for regenerating the injured cerebral cortex Injured neurons are replaced by 1) neural cell transplantation, 2) endogenous neuroblast migration and proliferation, and/or 3) endogenous neuron proliferation. Transplanted biomaterials, either with or without living cells, can accelerate cortical regeneration..

3.2. Biomaterials with cell transplantation

Biomaterials were originally defined as nonviable materials designed to interact with biological systems when used in medical devices [59]. However, biomaterials now include transplantable hybrids containing both nonviable materials and living cells [60]. A combination of neural stem/progenitor cell transplantation and biomaterials can enhance the regenerative process and promote functional cortical recovery in some cases [61–64]. Again, to develop reliable regenerative therapies for functional recovery, we must determine which factors are essential during the regenerative process.

4. Future perspectives

In the past few years, remarkable advances have been made in culturing 3D organoids from pluripotent stem cells—retinal

organoids, for example [65]. This technology has great potential not only for determining the potency of pluripotent stem cells to generate self-organizing neural tissue, but also for retinal regeneration therapy. Transplants of 3D retinal organoids derived from pluripotent stem cells promote retinal regeneration in mice, even when advanced degeneration is present [66]. Cerebral organoids have also been generated from pluripotent stem cells [67,68], and the potential use of these organoids for regenerating the injured cerebral cortex merits further study.

Recent advances in neurobiology and in biomaterial engineering shed new light on the potential of cortical regeneration, and researchers have tried various methods to promote regeneration (see Fig. 1). Although functional recovery has occurred after neural stem/progenitor cell transplantation, careful analysis is required to understand how neuronal networks regenerate. There is no doubt that the cerebral cortex has the potential to regenerate after injury. Future studies combining neurobiology and biomaterial engineering will drive the development of therapeutic strategies to regenerate the injured cerebral cortex.

Acknowledgments

This work was supported by a Grant-in-Aid for Scientific Research from the Ministry of Education, Culture, Sports, Science and Technology (MEXT) (26106711) and by the Terumo Life Science Foundation.

References

[1] Cajal SR. Estudios sobre la degeneración y regeneración del sistema nervioso. Madrid: Imprenta de Hijos de Nicolás Moya; 1913.
[2] Kriegstein A, Alvarez-Buylla A. The glial nature of embryonic and adult neural stem cells. Annu Rev Neurosci 2009;32:149–84.
[3] Kohwi M, Doe CQ. Temporal fate specification and neural progenitor competence during development. Nat Rev Neurosci 2013;14:823–38.
[4] Meyer JS, Shearer RL, Capowski EE, Wright LS, Wallace KA, McMillan EL, et al. Modeling early retinal development with human embryonic and induced pluripotent stem cells. Proc Natl Acad Sci U S A 2009;106:16698–703.
[5] Osakada F, Ikeda H, Mandai M, Wataya T, Watanabe K, Yoshimura N, et al. Toward the generation of rod and cone photoreceptors from mouse, monkey and human embryonic stem cells. Nat Biotechnol 2008;26:215–24.
[6] MacLaren RE, Pearson RA, MacNeil A, Douglas RH, Salt TE, Akimoto M, et al. Retinal repair by transplantation of photoreceptor precursors. Nature 2006;444:203–7.
[7] Pearson RA, Barber AC, Rizzi M, Hippert C, Xue T, West EL, et al. Restoration of vision after transplantation of photoreceptors. Nature 2012;485:99–103.
[8] McConnell SK. Fates of visual cortical neurons in the ferret after isochronic and heterochronic transplantation. J Neurosci 1988;8:945–74.
[9] Macklis JD. Transplanted neocortical neurons migrate selectively into regions of neuronal degeneration produced by chromophore-targeted laser photolysis. J Neurosci 1993;13:3848–63.
[10] Reubinoff BE, Itsykson P, Turetsky T, Pera MF, Reinhartz E, Itzik A, et al. Neural progenitors from human embryonic stem cells. Nat Biotechnol 2001;19:1134–40.
[11] Zhang SC, Wernig M, Duncan ID, Brüstle O, Thomson JA. In vitro differentiation of transplantable neural precursors from human embryonic stem cells. Nat Biotechnol 2001;19:1129–33.
[12] Jensen MB, Yan H, Krishnaney-Davison R, Al Sawaf A, Zhang S-C. Survival and differentiation of transplanted neural stem cells derived from human induced pluripotent stem cells in a rat stroke model. J Stroke Cerebrovasc Dis 2013;22:304–8.
[13] Kelly S, Bliss TM, Shah AK, Sun GH, Ma M, Foo WC, et al. Transplanted human fetal neural stem cells survive, migrate, and differentiate in ischemic rat cerebral cortex. Proc Natl Acad Sci U S A 2004;101:11839–44.
[14] Pittenger MF, Mackay AM, Beck SC, Jaiswal RK, Douglas R, Mosca JD, et al. Multilineage potential of adult human mesenchymal stem cells 2011;143:143–8.
[15] Kopen GC, Prockop DJ, Phinney DG. Marrow stromal cells migrate throughout forebrain and cerebellum, and they differentiate into astrocytes after injection into neonatal mouse brains. Proc Natl Acad Sci U S A 1999;96:10711–6.
[16] Labbe R, Firl A, Mufson EJ, Stein DG. Fetal brain transplant: reduction of cognitive deficits in rats with frontal cortex lesions. Science 1983;221:470–2.
[17] Borlongan CV, Tajima Y, Trojanowski JQ, Lee VM, Sanberg PR. Transplantation of cryopreserved human embryonal carcinoma-derived neurons (NT2N cells) promotes functional recovery in ischemic rats. Exp Neurol 1998;149:310–21.
[18] Mohamad O, Drury-Stewart D, Song M, Faulkner B, Chen D, Yu SP, et al. Vector-free and transgene-free human iPS cells differentiate into functional neurons and enhance functional recovery after ischemic stroke in mice. PLoS One 2013;8:e64160.
[19] Chen J, Li Y, Wang L, Zhang Z, Lu D, Lu M, et al. Therapeutic benefit of intravenous administration of bone marrow stromal cells after cerebral ischemia in rats. Stroke 2001;32:1005–11.
[20] Eriksson PS, Perfilieva E, Björk-Eriksson T, Alborn AM, Nordborg C, Peterson DA, et al. Neurogenesis in the adult human hippocampus. Nat Med 1998;4:1313–7.
[21] Kuhn HG, Dickinson-Anson H, Gage FH. Neurogenesis in the dentate gyrus of the adult rat: age-related decrease of neuronal progenitor proliferation. J Neurosci 1996;16:2027–33.
[22] Reynolds BA, Weiss S. Generation of neurons and astrocytes from isolated cells of the adult mammalian central nervous system. Science 1992;255:1707–10.
[23] Doetsch F, Caillé I, Lim DA, García-Verdugo JM, Alvarez-Buylla A. Subventricular zone astrocytes are neural stem cells in the adult mammalian brain. Cell 1999;97:703–16.
[24] Lois C, Alvarez-Buylla A. Long-distance neuronal migration in the adult mammalian brain. Science 1994;264:1145–8.
[25] Saha B, Peron S, Murray K, Jaber M, Gaillard A. Cortical lesion stimulates adult subventricular zone neural progenitor cell proliferation and migration to the site of injury. Stem Cell Res 2013;11:965–77.
[26] Yamashita T, Ninomiya M, Hernandez Acosta P, Garcia-Verdugo JM, Sunabori T, Sakaguchi M, et al. Subventricular zone-derived neuroblasts migrate and differentiate into mature neurons in the post-stroke adult striatum. J Neurosci 2006;26:6627–36.
[27] Kreuzberg M, Kanov E, Timofeev O, Schwaninger M, Monyer H, Khodosevich K. Increased subventricular zone-derived cortical neurogenesis after ischemic lesion. Exp Neurol 2010;226:90–9.
[28] Jin K, Sun Y, Xie L, Peel A, Mao XO, Batteur S, et al. Directed migration of neuronal precursors into the ischemic cerebral cortex and striatum. Mol Cell Neurosci 2003;24:171–89.
[29] Faiz M, Acarin L, Villapol S, Schulz S, Castellano B, Gonzalez B. Substantial migration of SVZ cells to the cortex results in the generation of new neurons in the excitotoxically damaged immature rat brain. Mol Cell Neurosci 2008;38:170–82.
[30] Arvidsson A, Collin T, Kirik D, Kokaia Z, Lindvall O. Neuronal replacement from endogenous precursors in the adult brain after stroke. Nat Med 2002;8:963–70.
[31] Ajioka I. Coordination of proliferation and neuronal differentiation by the retinoblastoma protein family. Dev Growth Differ 2014;56:324–34.
[32] Herrup K, Yang Y. Cell cycle regulation in the postmitotic neuron: oxymoron or new biology? Nat Rev Neurosci 2007;8:368–78.
[33] Burkhart DL, Sage J. Cellular mechanisms of tumour suppression by the retinoblastoma gene. Nat Rev Cancer 2008;8:671–82.
[34] Weinberg RA. The retinoblastoma protein and cell cycle control. Cell 1995;81:323–30. http://dx.doi.org/10.1016/0092-8674(95)90385-2.
[35] Andrusiak MG, Vandenbosch R, Park DS, Slack RS. The retinoblastoma protein is essential for survival of postmitotic neurons. J Neurosci 2012;32:14809–14. http://dx.doi.org/10.1523/JNEUROSCI.1912-12.2012.
[36] Zhang J, Benavente CA, McEvoy J, Flores-Otero J, Ding L, Chen X, et al. A novel retinoblastoma therapy from genomic and epigenetic analyses. Nature 2012;481:329–34.
[37] McEvoy J, Flores-Otero J, Zhang J, Nemeth K, Brennan R, Bradley C, et al. Coexpression of normally incompatible developmental pathways in retinoblastoma genesis. Cancer Cell 2011;20:260–75.
[38] Ajioka I, Martins RAP, Bayazitov IT, Donovan S, Johnson DA, Frase S, et al. Differentiated horizontal interneurons clonally expand to form metastatic retinoblastoma in mice. Cell 2007;131:378–90.
[39] Oshikawa M, Okada K, Nakajima K, Ajioka I. Cortical excitatory neurons become protected from cell division during neurogenesis in an Rb family-dependent manner. Development 2013;140:2310–20.
[40] MacPherson D, Sage J, Crowley D, Trumpp A, Bronson RT, Jacks T. Conditional mutation of Rb causes cell cycle defects without apoptosis in the central nervous system. Mol Cell Biol 2003;23:1044–53.
[41] Ferguson KL, Vanderluit JL, Hébert JM, McIntosh WC, Tibbo E, MacLaurin JG, et al. Telencephalon-specific Rb knockouts reveal enhanced neurogenesis, survival and abnormal cortical development. EMBO J 2002;21:3337–46.
[42] Yang Y, Geldmacher DS, Herrup K. DNA replication precedes neuronal cell death in Alzheimer's disease. J Neurosci 2001;21:2661–8.
[43] Kuan C-Y, Schloemer AJ, Lu A, Burns KA, Weng W-L, Williams MT, et al. Hypoxia-ischemia induces DNA synthesis without cell proliferation in dying neurons in adult rodent brain. J Neurosci 2004;24:10763–72.
[44] Höglinger GU, Breunig JJ, Depboylu C, Rouaux C, Michel PP, Alvarez-Fischer D, et al. The pRb/E2F cell-cycle pathway mediates cell death in Parkinson's disease. Proc Natl Acad Sci U S A 2007;104:3585–90.
[45] El-Khodor BF, Oo TF, Kholodilov N, Burke RE. Ectopic expression of cell cycle markers in models of induced programmed cell death in dopamine neurons of the rat substantia nigra pars compacta. Exp Neurol 2003;179:17–27.
[46] Osuga H, Osuga S, Wang F, Fetni R, Hogan MJ, Slack RS, et al. Cyclin-dependent kinases as a therapeutic target for stroke. Proc Natl Acad Sci U S A 2000;97:10254–9.
[47] Yu Y, Ren QG, Zhang ZH, Zhou K, Yu ZY, Luo X, et al. Phospho-Rb mediating cell cycle reentry induces early apoptosis following oxygen–glucose deprivation in rat cortical neurons. Neurochem Res 2012;37:503–11.

[48] Nguyen MD, Boudreau M, Kriz J, Couillard-Després S, Kaplan DR, Julien J-P. Cell cycle regulators in the neuronal death pathway of amyotrophic lateral sclerosis caused by mutant superoxide dismutase 1. J Neurosci 2003;23:2131–40.
[49] Biswas SC, Shi Y, Vonsattel JG, Leung CL, Troy CM, Greene LA. Bim is elevated in Alzheimer's disease neurons and is required for beta-amyloid-induced neuronal apoptosis. J Neurosci 2007;27:893–900.
[50] Silva AR, Santos AC, Farfel JM, Grinberg LT, Ferretti RE, Campos AH, et al. Repair of oxidative DNA damage, cell-cycle regulation and neuronal death may influence the clinical manifestation of Alzheimer's disease. PLoS One 2014;9:e99897.
[51] Le Magueresse C, Alfonso J, Bark C, Eliava M, Khrulev S, Monyer H. Subventricular zone-derived neuroblasts use vasculature as a scaffold to migrate radially to the cortex in neonatal mice. Cereb Cortex 2012;22:2285–96.
[52] Thored P, Wood J, Arvidsson A, Cammenga J, Kokaia Z, Lindvall O. Long-term neuroblast migration along blood vessels in an area with transient angiogenesis and increased vascularization after stroke. Stroke 2007;38:3032–9.
[53] Kojima T, Hirota Y, Ema M, Takahashi S, Miyoshi I, Okano H, et al. Subventricular zone-derived neural progenitor cells migrate along a blood vessel scaffold toward the post-stroke striatum. Stem Cells 2010;28:545–54.
[54] Ajioka I, Ichinose S, Nakajima K, Mizusawa H. Basement membrane-like matrix sponge for the three-dimensional proliferation culture of differentiated retinal horizontal interneurons. Biomaterials 2011;32:5765–72.
[55] Ajioka I, Jinnou H, Okada K, Sawada M, Saitoh S, Sawamoto K. Enhancement of neuroblast migration into the injured cerebral cortex using laminin-containing porous sponge. Tissue Eng Part A 2015;21:193–201.
[56] Deguchi K, Tsuru K, Hayashi T, Takaishi M, Nagahara M, Nagotani S, et al. Implantation of a new porous gelatin–siloxane hybrid into a brain lesion as a potential scaffold for tissue regeneration. J Cereb Blood Flow Metab 2006;26:1263–73.
[57] Hou S, Xu Q, Tian W, Cui F, Cai Q, Ma J, et al. The repair of brain lesion by implantation of hyaluronic acid hydrogels modified with laminin. J Neurosci Methods 2005;148:60–70.
[58] Huang K-F, Hsu W-C, Chiu W-T, Wang J-Y. Functional improvement and neurogenesis after collagen–GAG matrix implantation into surgical brain trauma. Biomaterials 2012;33:2067–75.
[59] Williams D. Definitions in biomaterials. In: Proc. a consens. conf. eur. soc. biomater., vol. 4. Chester, England: Elsevier; 1986. p. 414.
[60] Ratner BD, Hoffman AS, Schoen FJ, Lemons JE. Biomaterials science: an introduction to materials in medicine. 3rd ed. Academic Press; 2012.
[61] Elias PZ, Spector M. Implantation of a collagen scaffold seeded with adult rat hippocampal progenitors in a rat model of penetrating brain injury. J Neurosci Methods 2012;209:199–211.
[62] Bible E, Chau DYS, Alexander MR, Price J, Shakesheff KM, Modo M. The support of neural stem cells transplanted into stroke-induced brain cavities by PLGA particles. Biomaterials 2009;30:2985–94.
[63] Cheng T-Y, Chen M-H, Chang W-H, Huang M-Y, Wang T-W. Neural stem cells encapsulated in a functionalized self-assembling peptide hydrogel for brain tissue engineering. Biomaterials 2013;34:2005–16.
[64] Jin K, Mao X, Xie L, Galvan V, Lai B, Wang Y, et al. Transplantation of human neural precursor cells in matrigel scaffolding improves outcome from focal cerebral ischemia after delayed postischemic treatment in rats. J Cereb Blood Flow Metab 2010;30:534–44.
[65] Eiraku M, Takata N, Ishibashi H, Kawada M, Sakakura E, Okuda S, et al. Self-organizing optic-cup morphogenesis in three-dimensional culture. Nature 2011;472:51–6.
[66] Assawachananont J, Mandai M, Okamoto S, Yamada C, Eiraku M, Yonemura S, et al. Transplantation of embryonic and induced pluripotent stem cell-derived 3D retinal sheets into retinal degenerative mice. Stem Cell Rep 2014;2:662–74.
[67] Eiraku M, Watanabe K, Matsuo-Takasaki M, Kawada M, Yonemura S, Matsumura M, et al. Self-organized formation of polarized cortical tissues from ESCs and its active manipulation by extrinsic signals. Cell Stem Cell 2008;3:519–32.
[68] Lancaster MA, Renner M, Martin C-A, Wenzel D, Bicknell LS, Hurles ME, et al. Cerebral organoids model human brain development and microcephaly. Nature 2013;501:373–9.