ACTA NEUROBIOLOGIAE EXPERIMENTALIS

Review Acta Neurobiol Exp 2009, 69: 1–10

Challenges and possibilities of intravascular cell therapy in stroke

Anna Hicks and Jukka Jolkkonen*

Department of Neurology, University of Kuopio, Kuopio, Finland, *Email: jukka.jolkkonen@uku.fi

Stroke is the third leading cause of death in Western countries and more importantly a leading cause of adult disability. The recovery process of stroke patients might be enhanced by intensive rehabilitation, which acts through brain plasticity mechanisms. Restorative approaches such as cell-based therapies are clinically appealing as it might be possible to help patients even when treatment is initiated days or weeks after the ischemic insult. An extensive number of experimental transplantation studies have been conducted with cells of different origins (e.g., embryonic stem, fetal neural stem, human umbilical cord blood) with promising results. Noninvasive intravascular administration of cells, which provides a broad distribution of cells to the close proximity of ischemic tissue, has perhaps the most immediate access to clinical applications. However, surprisingly little is known about whole body biodistribution of intravascularly administered cells and mechanisms leading to improved functional recovery. This review examines the recent literature concerning intravascular cell-based therapies in experimental stroke.

Key words: biodistribution, experimental cerebral ischemia, functional recovery, intravascular administration, mechanism

INTRODUCTION

Stroke affects approximately 6 million people in the EU, with 1.1 million new cases every year. Despite spontaneous recovery in some patients, more than 50% of stroke patients have residual impairment placing a huge burden on the patients, their relatives, and society. Therapy of cerebrovascular diseases includes prevention, acute care and neuroprotection, and rehabilitation of which preventive pharmacotherapies together with lifestyle modifications are effective in decreasing stroke mortality and incidence (Sacco et al. 2006). In contrast, neuroprotective approaches have proven to be more difficult as shown by a recent disappointing multi centre clinical SAINT II trial (Shuaib et al. 2007). Tissue plasminogen activator (tPA) remains a treatment for only a fraction of stroke victims, due to the narrow therapeutic time window (Barber et al. 2001). Thus, much hope is placed on cell-based therapies as a promising treatment option or treatment addition for stroke patients.

Stem cells of different origins, i.e., embryonic stem (ES), fetal neural stem (NS), human umbilical cord blood (HUCB), and bone marrow stromal (BMS) cells, have been tested in experimental stroke models to promote functional recovery (Vora et al. 2006, Bacigaluppi et al. 2008, Guzman et al. 2008b). Since cell types, administration routes, stroke models, and behavioral tests used as outcome measures have varied significantly (Table I), it is difficult to compare the studies of which most indicate a promising regenerative potential. Safety and feasibility of cell-based therapy have also been reproduced in small studies with stroke patients (Kondziolka et al. 2000, 2005, Bang et al. 2005).

Two main routes have been used for the stem cell delivery. The first is a stereotaxic transplantation of cells into the brain. Given that stroke often produces large ischemic damage, it is not known whether such a targeted approach can provide efficient and wide cell engraftment, even with the aid of anatomical and functional imaging to explore location of cell transplantation.

Correspondence should be addressed to J. Jolkkonen, Email: jukka.jolkkonen@uku.fi

Received 09 September 2008, accepted 27 November 2008

Table I

Factors affecting homing of cells after intravascular administration
Type of stem cells (adult or embryonic)
The origin of adult stem cells (bone marrow, skin, or adipose tissue)
Species (allogenic, autogenic, xenografting)
Dose of cells (e.g., microvascular occlusions with high doses)
Size of cells (e.g., accumulation in lungs)
Time of treatment (acute, subacute, or chronic stage of stroke)
Administration route (intravenous, intra-arterial, or intracerebral)
Others (e.g., immunosuppression, cell modifications, co-medication)

Another concern is the invasive nature of intracerebral transplantation. The second approach is intravascular administration, which does not necessary rely on the cellular replacement but rather on the activation of the brain's own repair mechanisms. Interestingly, entry of intravenously injected cells into the central nervous system is not required for therapeutic effects, indicating that peripheral mechanisms may play a role as well (Borlongan et al. 2004). Intravenous delivery may actually be more effective than intraparenchymal injection of cells (Willing et al. 2003). In this review we will discuss the challenges and possibilities of cell-based therapies in animal models of stroke with a special emphasis on intravascular administration of cells.

SPECIAL CHALLENGES OF CELL-BASED THERAPY IN STROKE

Cell-based therapy after a massive ischemic damage of particular brain structures in stroke patients can be challenging for example compared to diabetes or Parkinson's disease, where restricted population of cells are lost. Not only neurons, but glial cells and blood vessels need to be repaired. Another distinction is that stroke is an acute injury with little or no degenerative process. The issue concerning timing of cell transplantation is twofold. While early cell transplantation may provide neuroprotection, the hostile environment endangers the long-term survival of transplanted cells. Transplantation at later time points targeted against the secondary neurodegeneration and enhancement of the brain's own repair mechanisms allows a better cell survival, although scar formation and lack of functional vasculature may limit the therapeutic benefit. Also, reduced expression of homing signals secreted by ischemic tissue such as stromal-derived factor-1 (SDF-1) and macrophage inflammatory protein alpha (MIP-1α) may lessen the efficacy particularly if cells are administered intravenously (Newman et al. 2005, Jiang et al. 2008). Shen and co-workers (2007a) showed, however, an increased SDF-1 expression up to 4 months after middle cerebral artery occlusion (MCAO) suggesting a much longer time window for cell-based therapy than it was previously believed. Finally, cell transplantation should be combined with other rehabilitative treatments to ensure the maximal therapeutic benefit for stroke patients.

BIODISTRIBUTION OF CELLS AFTER INTRAVASCULAR ADMINISTRATION

The most effective transplantation route to deliver cells into the brain following cerebral ischemia remains undetermined. Jin and co-workers (2005) concluded that a variety of routes (e.g., intrastriatal, intraventricular, and intravenous injections) in MCAO rats can deliver cells to the ischemic brain regions. Noninvasive intravenous administration has perhaps the most immediate access to clinical applications and it has been used in experimental studies with variable success (see Guzman et al. 2008b). However, not much is known about the proportional distribution of cells administered by intravenous route at the whole body level (Table II). Chen and co-workers (2001a) estimated that after intravenous administration of HUCB cells in MCAO rats, only 1% of injected cells were detected in the brain. Radioactively labeled rat mesenchymal cells have been shown to localize in the internal organs and primarily in the lungs after intravenous and intra-arterial infusions in naive rats (Gao et al. 2001). In addition, tracking of ^{111}In–oxine labeled HUCB cells and human ES cell-derived neural progenitors by small animal

Table II

Biodistribution of cells after intravascular administration in experimental animals

Cell type	Model	Administration route, time	Cell tracking, time	Distribution pattern	Comments	Reference
rat BMS cells	naive rats	femoral vein or artery	SPECT/CT, ^{111}In-oxine immediately and 48 h after infusion	liver<lung <kidney<spleen	large size of BMCs is suggested to explain homing in lung	Gao et al. 2001
human EP cells	immunodeficient rats, myocardial infarction	tail vein, 24 h after operation	gamma camera, ^{111}In-oxine, 24 and 96 h	liver<spleen <kidney<lung		Aicher et al. 2003
human UCB cells	rat tMCAO	femoral vein, 24 h	PCR, immunohisto-chemistry	spleen, ipsilateral hemisphere	no cells in spleen or brain in control rats	Vendrame et al. 2004
human UCB cells	rat tMCAO	femoral vein, 24 h	SPECT/CT, ^{111}In-oxine immediately and 24 h	lung<liver <spleen	no signal in brain	Mäkinen et al. 2006
male rat BMS cells	rat tMCAO	internal carotid artery, 24 h	*in situ* hybridization for Y chromosome at one-year	very few cells in heart, lung, liver, spleen or kidney	cells do not survive in internal organs	Shen et al. 2007b
human ES cells, rat hippocampal cells	rat tMCAO	internal carotid artery or femoral vein, 24 h	SPECT/CT, ^{111}In-oxine immediately and 24 h	liver<spleen <kidney	minor signal in ischemic brain after intra-arterial administration	Lappalainen et al. 2008
rat BMS cells	rat tMCAO	internal carotid artery or femoral vein, 30 min	MRI, 2–24 h	signal in ischemic brain only after intra-arterial infusion	microvascular occlusions as a complication	Walczak et al. 2008
human AMS cells	immunocom-promised mice	tail vein	G-Luc, bioluminence imaging up to 32 weeks	liver the preferred target		Vilalta et al. 2008

(AMS) adipose tissue-derived mesenchymal stem; (BMS) bone marrow stromal; (EP) endothelial progenitor; (ES) embryonic stem; (tMCAO) transient middle cerebral artery occlusion; (SPECT) single photon emission computed tomography; (UCB) umbilical cord blood

Table III

Summary of studies using intravascular administration of cells in experimental stroke models with behavioral follow-up

Cell type	Injury model	Route, time after stroke	Survival time	Surviving cells in the brain tissue	Differentiation	Effect on infarct volume	Behavioral outcome	Reference
human UCB cells	rat tMCAO	tail vein, 24 h or 7 days	14 days and 35 days	3.2% and 2.7%	neurons 2%, astrocytes 7%	no effect	improvements in rotarod and NSS in 24 h group and improvement in 7 day group in NSS	Chen et al. 2001a
rat BMS cells	rat tMCAO	intravenous, 24 h	14 days and 35 days	3.2% and 1.6%	neurons 1–2%, astrocytes 5%	no effect	higher dose (3×10^6) led to improvements in adhesive tape removal and NSS, no effect in rotarod test	Chen et al. 2001b
human UCB cells	rat tMCAO	intravenous, during occlusion	3 days	no cells found (despite BBB opening)	no cells found	reduced cerebral infarcts	improvements in body swing and passive avoidance test.	Borlongan et al. 2004
human NS cells	rat tMCAO	tail vein, 24 h	1 day – 540 days	<1 %	neurons 20%, astrocytes 50%	at day 56 less hemispheric atrophy	improvements in rotarod, modified limb placement, turning in an alley tests up to 28 days	Chu et al. 2004
rat BMS cells	rat tMCAO (45 min)	intravenous, 3–72 h	2 weeks	ND	some cells expressed neuronal and glial markers	early intervention neuro-protective	improved performance in treadmill and Morris water-maze	Iihoshi et al. 2004
human UCB cells	rat tMCAO	femoral vein, 24 h	4 weeks	ND	ND	reduced hemispheric infarct volume	improved behavior in spontaneous activity, elevated body swing and step tests	Vendrame et al. 2004
human BMS cells transfected with BDNF	rat pMCAO	intravenous, 6 h	7 days	458.6 cells/mm^2 around lesion in hMSCs + BDNF group	NeuN-positive 7.90%, GFAP-positive 7.65% in hMSCs + BDNF group	neuro-protection hMSCs <MSCs + BDN	improved performance in treadmill controls <hMSCs<MSCs + BDNF	Nomura et al. 2005

Table III

human UCB cells	rat tMCAO	femoral vein, 24 h	25 days	only few cells	ND	no effect	no effects on water maze, limb-placement, beam walking or cylinder tests	Mäkinen et al. 2006
human BMS cells	rat tMCAO	femoral vein, 12 h	1 week	ND	NeuN-positive 9.31%, GFAF-positive 7.98%	significant reduction in infarct size	improved performance in treadmill and Morris water-maze	Honma et al. 2006
human BMS cells transfected with GDNF	rat pMCAO	intravenous, 3 h	31 days	422.5 cells/mm^2 in the penumbra	ND	protection 1–7 days after MCAO	improvement in treadmill test control <hMSC <GDNF-hMSC	Horita et al. 2006
rat BMS cells	rat tMCAO	internal carotid artery, 24 h	28 days			no effect	improvements in mNSS, adhesive-removal, and corner tests	Shen et al. 2006
rat BMS cells	rat tMCAO	internal carotid artery, 24 h	up to one year	38 cells per section	neurons 16.8%, astrocytes 22.3%, microglia 5.5%, endothelial cells <1%	no effect	improvements in NSS and adhesive tape removal test	Shen et al. 2007b
CD49d-positive mice NS cells	mice hypoxia/ ischemia	internal carotid artery, 48 h	17 days	up to 1300 cells/mm^2	numerous β-tubulin III and DCX positive cells	ND	improvement in rotarod	Guzman et al. 2008a
rat MS cells +EPO	rat tMCAO	tail vein, 24 h	51 days	11.8 cells/cm^2 ipsilateral hemisphere	NeuN-positive 36%, GFAP-positive 17%	no differences between groups	improved performance in passive avoidance test after EPO and MSC + EPO	Esneault et al. 2008

(BDNF) brain derived neurotrophic factor; (BMS) mesenchymal bone marrow stem; (EPO) erythropoietin; (DCX) doublecortin; (GFAP) glial fibrillarary acidic protein; (pMCAO) permanent middle cerebral artery occlusion; (tMCAO) transient middle cerebral artery occlusion; (NeuN) a neuronal specific nuclear protein; (NS) neural stem; (ND) not determined; (NSS) neurological severity score; (UCB) umbilical cord blood

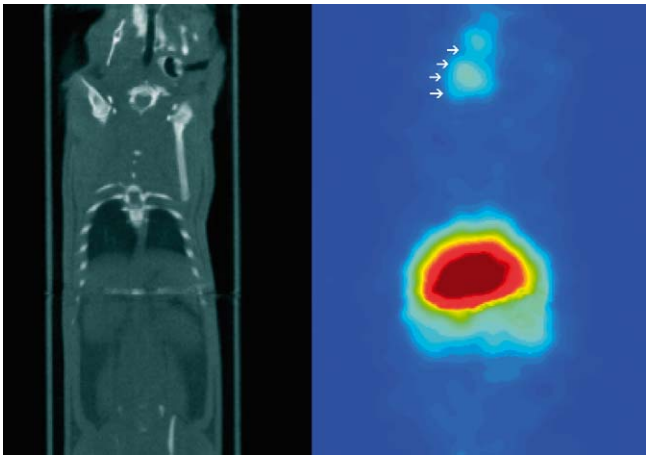

Fig. 1. The accumulation of ^{111}In-oxine labeled human embryonic stem (ES) cell-derived neural cells after intra-arterial infusion in rats subjected to middle cerebral artery occlusion (MCAO). Arrows indicate minor accumulation in the ischemic hemisphere. A CT image on the left is provided for an anatomical reference (from Lappalainen et al. 2008*).

single-photon emission computed tomography (SPECT) showed localization of cells in the internal organs after intravenous injection in MCAO rats (Mäkinen et al. 2006, Lappalainen et al. 2008). Undesirable biodistribution is most likely caused by the accumulation of cells in trapping and filtering organs such as lung, liver, and spleen, rather than due to the cell type injected or timing of administration. Internal organs are also characterized by very high concentration of SDF-1, which may regulate migration and circulation of cells (Kucia et al. 2005). Cell size, especially in the case of mesenchymal cells, may contribute to accumulation into the lungs (Gao et al. 2001). Recently, Walczak and others (2008) showed by using combined laser Doppler blood flow monitoring and magnetic resonance imaging (MRI) of iron labeled cells, that intra-arterial but not intravenous injections of mesenchymal stem cells provided a successful but variable cerebral engraftment. This was, however, associated with microvascular occlusions. Minor accumulation of neural progenitor cells in the ischemic hemisphere after intra-arterial administration was also shown by SPECT (Fig. 1) (Lappalainen et al. 2008). Thus, intra-arterial administration of cells may be a method of choice in future stroke related studies.

* Reprinted from Neuroscience Letters, Vol. 440, Lappalainen RS, Narkilahti S, Huhtala T, Liimatainen T, Suuronen T, Närvänen A, Suuronen R, Hovatta O, Jolkkonen J, The SPECT imaging shows the accumulation of neural progenitor cells into internal organs after systemic administration in middle cerebral artery occlusion rats, p. 246–250, Copyright (c) 2008, with permission from Elsevier.

EXPERIMENTAL STROKE STUDIES USING INTRAVASCULAR CELL THERAPY

Human umbilical cord blood, which is rich in adult stem and progenitor cells (both hematopoietic and non-hematopoietic), is one potential source for cells used for transplantations (Table III). The use of HUCB cells do not raise ethical issues, they have weak immunogenicity, and are proven to be safe treatment of pediatric diseases (Sanberg et al. 2005). When stem cells isolated from human umbilical cord blood were introduced to MCAO rats intravenously 24 hours after the insult (Chen et al. 2001a, Borlongan et al. 2004, Vendrame et al. 2004), improved behavioral recovery was observed in spontaneous activity, elevated body swing, step test, passive avoidance, rotarod, and neurological severity score (NSS). In contrast, our laboratory showed that intravenous delivery of unselected HUCB cells in MCAO rats provided no benefits in a battery of more demanding and perhaps more predictive tests of functional recovery such as tapered beam-walking, cylinder, and Morris water-maze tasks (Mäkinen et al. 2006). It may be that only a selected fraction of HUCB cells, such as CD34$^+$ or CD49d$^+$, provides significant treatment effects (Nystedt et al. 2006, Guzman et al. 2008a).

Bone marrow is another source of non-hematopoietic cells. A series of experiments conducted with BMS cells showed some functional recovery in rats after transient MCAO (Table III). After intravenous administration of BMS cells in MCAO rats 24 h after the insult, improved behavioral recovery was detected in rotarod and modified NSS (Chen et al. 2001b). Therapeutic benefit of BMS cells was also observed when cells were administered 1 month after stroke (Shen et al. 2006) and it persisted for at least 1 year (Shen et al. 2007b). Early intervention with BMS cells was neuroprotective up to 72 h in a transient MCAO model (Iihoshi et al. 2004).

Various attempts have been made to further improve the therapeutic effects of intravascular cell therapies (Table III). For example, intravenous administration of human mesenchymal stem cells engineered for production of brain-derived neurotrophic factor (BDNF) (Nomura et al. 2005) or glial cell line-derived neurotrophic factor (GDNF) (Horita et al. 2006) protected against injury in a cerebral ischemia model in the adult rat. A combination of nitric oxide donors (Chen et al. 2004, Cui et al. 2008) or erythropoietin (Esneault et al.

2008) and mesencymal stem cell transplantations in MCAO rats was superior when compared to either alone. Eventually, in clinical practice cell-based therapies will be combined with other rehabilitative approaches (e.g., constraint induced movement therapy). In an experimental setting this can be realized by housing rats in an enriched environment and providing them with an additional daily rehabilitative training after cell transplantation. There is some evidence that neural stem/progenitor (NP) cells improved behavioral recovery but only when combined with housing in the enriched environment (Grabowski et al. 1995). Also, a targeted homing of intravenously given cells by pharmacological manipulations should be possible in future.

MECHANISMS OF NEUROPROTECTION AND NEURAL REPAIR

Neuroprotection against ischemic damage is challenging due to the narrow therapeutic time window. Early delivery of cells has, however, provided a dose-dependent neuroprotection (Borlongan et al. 2004). This is suggested to be mediated by trophic factors without cell entrance into brain parenchyma. To support this idea only few, if any, transplanted cells were detected in the brain after the intravascular administration after MCAO, not even when coinfused with mannitol, a blood-brain barrier permeabilizer (Borlongan et al. 2004). The trophic factors responsible for neuroprotection include BDNF, NGF, and vascular endothelial growth factor (VEGF) (Chen et al. 2002). Consistent with this, BDNF and GDNF modified cells provide additive neuroprotection (Nomura et al. 2005, Horita et al. 2006). One should note that neuroprotection seems to be more pronounced in MCAO rats when short occlusion time (45 min) was applied, which produces mainly striatal damage (Iihoshi et al. 2004, Honma et al. 2006). Surprisingly HUCB cells were shown to be effective in decreasing infarct size even when administered 48 h after permanent MCAO possibly by preventing penumbral apoptosis (Newcomb et al. 2006). Also, inflammation plays an important role in neuroprotection by HUCB cells (Vendrame et al. 2005).

The cell delivery is thought to also target the brain's own repair mechanisms including angiogenesis and neurogenesis, attenuation of scar formation, or most likely a combination of them (Chen et al. 2004). For example, BMS cells secrete several growth factors involved in vascular formation (Chen et al. 2002) and increase endogenous production of vascular growth factors (Chen et al. 2003) promoting angiogenesis in the vulnerable ischemic border zone in need of oxygen. On the other hand, there is evidence that the VEGF dose necessary to promote angiogenesis is not neuroprotective and is actually harmful to recovering neurons (Manoonkitiwongsa et al. 2004). Cerebral ischemia induces endogenous neurogenesis, which seems to be further enhanced by cell therapy (Chen et al. 2004, Esneault et al. 2008). However, the long-term fate of new endogenously born neurons needs to be determined. BMS cells can also increase restorative changes in the corpus callosum (Shen et al. 2006). The structural changes induced by BMS cells seem to persist at least 1 year (Shen et al. 2007b).

CONCLUSIONS

Cell-based therapies alone or as part of a combination therapy are a promising approach to improve functional recovery following ischemic insults. Intravascular administration of cells has perhaps the most immediate access to clinical applications. However, best cell type and timing of treatment are the major factors which need to be determined in the near future. We also would like to emphasize the importance of using multiple, sensitive behavioral tests as functional outcome measures and translational imaging modalities. In addition, we should understand the underlying restorative mechanisms in more detail to ensure safe and effective translation of experimental results into clinics.

ACKNOWLEDGEMENT

We thank Dr. Susanna Narkilahti (Regea, Finland) and Jared Clarke for critically reading this manuscript.

REFERENCES

Aicher A, Brenner W, Zuhayra M, Badorff C, Massoudi S, Assmus B, Eckey T, Henze E, Zeiher AM, Dimmeler S (2003) Assessment of the tissue distribution of transplanted human endothelial progenitor cells by radioactive labeling. Circulation 107: 2134–2139.

Bacigaluppi M, Pluchino S, Martino G, Kilic E, Hermann DM (2008) Neural stem/precursor cells for the treatment of ischemic stroke. J Neurol Sci 265: 73–77.

Bang OY, Lee JS, Lee PH, Lee G (2005) Autologous mesenchymal stem cell transplantation in stroke patients. Ann Neurol 57: 874–882.

Barber PA, Zhang J, Demchuk AM, Hill MD, Buchan AM (2001) Why are stroke patients excluded from TPA therapy? An analysis of patient eligibility. Neurology 56: 1015–1020.

Borlongan CV, Hadman M, Sanberg CD, Sanberg PR (2004) Central nervous system entry of peripherally injected umbilical cord blood cells is not required for neuroprotection in stroke. Stroke 35: 2385–2389.

Chen J, Sanberg PR, Li Y, Wang L, Lu M, Willing AE, Sanchez-Ramos J, Chopp M (2001a) Intravenous administration of human umbilical cord blood reduces behavioral deficits after stroke in rats. Stroke 32: 2682–2688.

Chen J, Li Y, Wang L, Zhang Z, Lu D, Lu M, Chopp M (2001b) Therapeutic benefit of intravenous administration of bone marrow stromal cells after cerebral ischemia in rats. Stroke 32: 1005–1011.

Chen J, Zhang ZG, Li Y, Wang L, Xu YX, Gautam SC, Lu M, Zhu Z, Chopp M (2003) Intravenous administration of human bone marrow stromal cells induces angiogenesis in the ischemic boundary zone after stroke in rats. Circ Res 92: 692–699.

Chen J, Li Y, Zhang R, Katakowski M, Gautam SC, Xu Y, Lu M, Zhang Z, Chopp M (2004) Combination therapy of stroke in rats with a nitric oxide donor and human bone marrow stromal cells enhances angiogenesis and neurogenesis. Brain Res 1005: 21–28.

Chen X, Li Y, Wang L, Katakowski M, Zhang L, Chen J, Xu Y, Gautam SC, Chopp M (2002) Ischemic rat brain extracts induce human marrow stromal cell growth factor production. Neuropathology 22: 275–279.

Chu K, Kim M, Park KI, Jeong SW, Park HK, Jung KH, Lee ST, Kang L, Lee K, Park DK, Kim SU, Roh JK (2004) Human neural stem cells improve sensorimotor deficits in the adult rat brain with experimental focal ischemia. Brain Res 1016: 145–153.

Cui X, Chen J, Zacharek A, Roberts C, Savant-Bhonsale S, Chopp M (2008) Treatment of stroke with (Z)-1-[N-(2-aminoethyl)-N-(2-ammonioethyl) amino] diazen-1-ium-1, 2-diolate and bone marrow stromal cells upregulates angiopoietin-1/Tie2 and enhances neovascularization. Neuroscience 156: 155–164.

Esneault E, Pacary E, Eddi D, Freret T, Tixier E, Toutain J, Touzani O, Schumann-Bard P, Petit E, Roussel S, Bernaudin M (2008) Combined therapeutic strategy using erythropoietin and mesenchymal stem cells potentiates neurogenesis after transient focal cerebral ischemia in rats. J Cereb Blood Flow Metab 28: 1552–1563.

Gao J, Dennis JE, Muzic RF, Lundberg M, Caplan AI (2001) The dynamic in vivo distribution of bone marrow-derived mesenchymal stem cells after infusion. Cell Tiss Org 169: 12–20.

Grabowski M, Sørensen JC, Mattsson B, Zimmer J, Johansson BB (1995) Influence of an enriched environment and cortical grafting on functional outcome in brain infarcts of adult rats. Exp Neurol 133: 96–102.

Guzman R, De Los Angeles A, Cheshier S, Choi R, Hoang S, Liauw J, Schaar B, Steiberg G (2008a) Intracarotid injection of fluorescence activated cell-sorted CD49d-positive neural stem cells improves targeted cell delivery and behavior after stroke in a mouse stroke model. Stroke 39: 1300–1306.

Guzman R, Choi R, Gera A, De Los Angeles A, Andres RH, Steinberg GK (2008b) Intravascular cell replacement therapy for stroke. Neurosurg Focus 24: E15.

Honma T, Honmou O, Iihoshi S, Harada K, Houkin K, Hamada H, Kocsis JD (2006) Intravenous infusion of immortalized human mesenchymal stem cells protects against injury in a cerebral ischemia model in adult rat. Exp Neurol 199: 56–66.

Horita Y, Honmou O, Harada K, Houkin K, Hamada H, Kocsis JD (2006) Intravenous administration of glial cell line-derived neurotrophic factor gene-modified human mesenchymal stem cells protects against injury in a cerebral ischemia model in the adult rat. J Neurosci Res 84: 1495–1504.

Iihoshi S, Honmou O, Houkin K, Hashi K, Kocsis JD (2004) A therapeutic window for intravenous administration of autologous bone marrow after cerebral ischemia in adult rats. Brain Res 1007: 1–9.

Jiang L, Newman M, Saporta S, Chen N, Sanberg PR, Willing AE (2008) MIP-1alpha and MCP-1 induce migration of human umbilical cord blood cells in models of stroke. Curr Neurovasc Res 5: 118–124.

Jin K, Sun Y, Xie L, Mao XO, Childs J, Peel A, Logvinova A, Banwait S, Greenberg DA (2005) Comparison of ischemia-directed migration of neural precursor cells after intrastriatal, intraventricular, or intravenous transplantation in the rat. Neurobiol Dis 18: 366–374.

Kondziolka D, Wechsler L, Goldstein S, Meltzer C, Thulborn KR, Gebel J, Jannetta P, DeCesare S, Elder EM, McGrogan M, Reitman MA, Bynum L (2000) Transplantation of cultured human neuronal cells for patients with stroke. Neurology 55: 565–569.

Kondziolka D, Steinberg GK, Wechsler L, Meltzer CC, Elder E, Gebel J, Decesare S, Jovin T, Zafonte R, Lebowitz J,

Flickinger JC, Tong D, Marks MP, Jamieson C, Luu D, Bell-Stephens T, Teraoka J (2005) Neurotransplantation for patients with subcortical motor stroke: a phase 2 randomized trial. J Neurosurg 103: 38–45.

Kucia M, Reca R, Miekus K, Wanzeck J, Wojakowski W, Janowska-Wieczorek A, Ratajczak J, Ratajczak MZ (2005) Trafficking of normal stem cells and metastasis of cancer stem cells involve similar mechanisms: pivotal role of the SDF-1-CXCR4 axis. Stem Cells 23: 879–894.

Lappalainen RS, Narkilahti S, Huhtala T, Liimatainen T, Suuronen T, Närvänen A, Suuronen R, Hovatta O, Jolkkonen J (2008) The SPECT imaging shows the accumulation of neural progenitor cells into internal organs after systemic administration in middle cerebral artery occlusion rats. Neurosci Lett 440: 246–250.

Manoonkitiwongsa PS, Schultz RL, McCreery DB, Whitter EF, Lyden PD (2004) Neuroprotection of ischemic brain by vascular endothelial growth factor is critically dependent on proper dosage and may be compromised by angiogenesis. J Cereb Blood Flow Metab 24: 693–702.

Mäkinen S, Kekarainen T, Nysted J, Liimatainen T, Huhtala T, Närvänen A, Laine J, Jolkkonen J (2006) Human umbilical cord blood cells do not improve sensorimotor or cognitive outcome following transient middle cerebral artery occlusion in rats. Brain Res 1123: 207–217.

Newcomb JD, Ajmo CT Jr, Sanberg CD, Sanberg PR, Pennypacker KR, Willing AE (2006) Timing of cord blood treatment after experimental stroke determines therapeutic efficacy. Cell Transplant 15: 213–223.

Newman MB, Willing AE, Manresa JJ, Davis-Sanberg C, Sanberg PR (2005) Stroke-induced migration of human umbilical cord blood cells: time course and cytokines. Stem Cells Dev 14: 576–586.

Nomura T, Honmou O, Harada K, Houkin K, Hamada H, Kocsis JD (2005) I.V. infusion of brain-derived neurotrophic factor gene-modified human mesenchymal stem cells protects against injury in a cerebral ischemia model in adult rat. Neuroscience 36: 161–169.

Nystedt J, Mäkinen S, Laine J, Jolkkonen J (2006) Human cord blood CD34+ cells and behavioral recovery following focal cerebral ischemia in rats. Acta Neurobiol Exp (Wars) 66: 293–300.

Sacco RL, Adams R, Albers G, Alberts MJ, Benavente O, Furie K, Goldstein LB, Gorelick P, Halperin J, Harbaugh R, Johnston SC, Katzan I, Kelly-Hayes M, Kenton EJ, Marks M, Schwamm LH, Tomsick T; American Heart Association/American Stroke Association Council on Stroke; Council on Cardiovascular Radiology and Intervention; American Academy of Neurology (2006) Guidelines for prevention of stroke in patients with ischemic stroke or transient ischemic attack: a statement for healthcare professionals from the American Heart Association/American Stroke Association Council on Stroke: co-sponsored by the Council on Cardiovascular Radiology and Intervention: the American Academy of Neurology affirms the value of this guideline. Circulation 113: e409–449.

Sanberg PR, Willing AE, Garbuzova-Davis S, Saporta S, Liu G, Sanberg CD, Bickford PC, Klasko SK, El-Badri NS (2005) Umbilical cord blood-derived stem cells and brain repair. Ann N Y Acad Sci 1049: 67–83.

Shen LH, Li Y, Chen J, Zhang J, Vanguri P, Borneman J, Chopp M (2006) Intracarotid transplantation of bone marrow stromal cells increases axon-myelin remodeling after stroke. Neuroscience 137: 393–399.

Shen LH, Li Y, Chen J, Zacharek A, Gao Q, Kapke A, Lu M, Raginski K, Vanguri P, Smith A, Chopp M (2007a) Therapeutic benefit of bone marrow stromal cells administered 1 month after stroke. J Cereb Blood Flow Metab 27: 6–3.

Shen LH, Li Y, Chen J, Cui Y, Zhang C, Kapke A, Lu M, Savant-Bhonsale S, Chopp M (2007b) One-year follow-up after bone marrow stromal cell treatment in middle-aged female rats with stroke. Stroke 38: 2150–2156.

Shuaib A, Lees KR, Lyden P, Grotta J, Davalos A, Davis SM, Diener HC, Ashwood T, Wasiewski WW, Emeribe U; SAINT II Trial Investigators (2007) NXY-059 for the treatment of acute ischemic stroke. N Engl J Med 357: 562–571.

Vendrame M, Cassady J, Newcomb J, Butler T, Pennypacker KR, Zigova T, Sanberg CD, Sanberg PR, Willing AE (2004) Infusion of human umbilical cord blood cells in a rat model of stroke dose-dependently rescues behavioral deficits and reduces infarct volume. Stroke 35: 2390–2395.

Vendrame M, Gemma C, de Mesquita D, Collier L, Bickford PC, Sanberg CD, Sanberg PR, Pennypacker KR, Willing AE (2005) Anti-inflammatory effects of human cord blood cells in a rat model of stroke. Stem Cells Dev 14: 595–604.

Vilalta M, Dégano IR, Bagó J, Gould D, Santos M, García-Arranz M, Ayats R, Fuster C, Chernajovsky Y, García-Olmo D, Rubio N, Blanco J (2008) Biodistribution, long-term survival, and safety of human adipose tissue-derived mesenchymal stem cells transplanted in nude mice by high sensitivity non-invasive bioluminescence imaging. Stem Cells Dev 17: 993–1003.

Vora N, Jovin T, Kondziolka D (2006) Cell transplantation for ischemic stroke, Neurodegener Dis 3: 101–105.

Walczak P, Zhang J, Gilad AA, Kedziorek DA, Ruiz-Cabelo J, Young RG, Pittenger MF, van Zijl PCM, Huang J, Bulte JWM (2008) Dual-modality monitoring of targeted intraarterial delivery of mesenchymal stem cells after transient ischemia. Stroke 39: 1569–1574.

Willing AE, Lixian J, Milliken M, Poulos S, Zigova T, Song S, Hart C, Sanchez-Ramos J, Sanberg PR (2003) Intravenous versus intrastriatal cord blood administration in a rodent model of stroke. J Neurosci Res 73: 296–307.