HHS Public Access

Author manuscript

Expert Rev Neurother. Author manuscript; available in PMC 2016 April 01.

Published in final edited form as:

Expert Rev Neurother. 2015 April ; 15(4): 381–393. doi:10.1586/14737175.2015.1021787.

Development in intracerebral stem cell grafts

Stephanny Reyes, **Naoki Tajiri**, and **Cesar V. Borlongan**[1]

Abstract

The field of stem cell therapy has emerged as a promising research area for brain repair. Optimizing the safety and efficacy of the therapy for clinical trials will require revisiting transplantation protocols. The cell delivery route stands as a key translational item that warrants careful consideration in facilitating the success of stem cell therapy in the clinic. Intracerebral administration, compared to peripheral route, requires an invasive procedure to directly implant stem cells into injured brain. Although invasive, intracerebral transplantation circumvents the prohibitive blood brain barrier in allowing grafted cells when delivered peripherally to penetrate the brain and reach the discreet damaged brain tissues. This review will highlight milestone discoveries in cell therapy for neurological disorders, with emphasis on intracerebral transplantation in relevant animal models and provide insights necessary to optimize the safety and efficacy of cell therapy for the treatment of Parkinson's disease, Huntington's disease, stroke, and traumatic brain injury.

Keywords

stem cell therapy; neurodegeneration; neurological disorders; cell delivery; translational research

Current Status On Treatment Options For Neurological Disorders

Disorders of the brain constitute a major economic burden due to the cost of treatment and the reduced ability of patients and their caregivers to bring forth income [1]. Most brain disorders are chronic and incurable conditions that leave patients with debilitating outcomes for years, which also places an emotional and practical burden on the family due to having to care after a person for such long period of time [1]. The incidence of neurodegenerative diseases is even projected to affect more than 12 million Americans 30 years from now due to these diseases occurring not only at a later stage in life but also at middle age [2]. As a result, these disorders are thought to have an even greater disease burden when compared to its mortality statistics [2].

[1]Corresponding Author: Professor and Director, Center of Excellence for Aging and Brain Repair, Department of Neurosurgery and Brain Repair, University of South Florida Morsani College of Medicine, MDC 78, 12901 Bruce B. Downs Blvd, Tampa, FL 33612, Phone Number: 813-974-3154; Fax Number: 813-974-3078; cborlong@health.usf.edu.

Financial & Competing Interests Disclosure

CV Borlongan is supported by NIH NINDS 1R01NS071956-01, NIH NINDS 1 R21 NS089851-01, James and Esther King Foundation for Biomedical Research Program 1KG01-33966, SanBio Inc., Celgene Cellular Therapeutics, KMPHC and NeuralStem Inc. None of these funders had a role in the preparation of this manuscript. CV Borlongan holds patents and has pending patent applications in stem cell biology and therapeutic applications. The authors have no other relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter or materials discussed in the manuscript apart from those disclosed.

The impetus for scientists to focus on the discovery of efficacious treatments and potential cures for neurodegenerative diseases has been a gradual successful process. Current laboratory work on stem cells shows a promising therapeutic approach for neurodegenerative diseases, however, there is paucity in translating this to large-scale trials in the clinic until solid evidence is demonstrated on safety and efficacy of transplanted stem cells. Stem cells are of special interest due to their ability to develop into different cell types and aid in the repair of the injured brain. The underlying mechanisms of action behind the ability of stem cells to respond and repair the area of injury is thought to encompass the combination of the replacement of injured cells, trophic support to enhance cell survival, and/or regulation of the inflammatory response [3]. If stem cells were to be used on patients, it is expected for the cells to behave as such without causing disruption in the integrity of surrounding areas or unaffected cells. Because of the robust proliferative property of stem cells, which is an advantage when contemplating of generating ample supply of cells, this feature presents also as a potential risk in that uncontrolled proliferation may lead to tumorgenesis. Such possibility of adverse outcome has alarmed some groups in the medical community in pursuing the use of stem cells in the clinic. However, in light that current pharmaceuticals only treat symptoms and slows the progression, disease models with significant animal data displaying a feasible and promising outcome in a disease should encourage the pursuit towards such therapeutic implementation. This review focuses on discoveries in cell therapy for neurological disorders, with emphasis on intracerebral transplantation in relevant animal models of neurological disorders, providing insights into optimizing the safety and efficacy of cell therapy for the treatment of Parkinson’s disease, Huntington’s disease, stroke, traumatic brain injury, amyotrophic lateral sclerosis, multiple sclerosis, and multiple system atrophy.

Candidate Neurological Disorders for Stem Cell Therapy

The ability to regenerate specific cell phenotypes *in vitro* has allowed cell therapy to be tailored to particular central nervous system (CNS) diseases. Animal models of brain disorders have also been created and standardized to assess the safety and efficacy of stem cell therapy. A wide variety of brain disorders have been the target of stem cell therapy, including acute injury and chronic neurodegenerative diseases because of the substantial debilitating effects of these disorders without any current cure or therapeutic treatment that halts the progression of the disease. Neurodegenerative diseases such Parkinson’s disease (PD) [4], Huntington’s disease (HD) [4], amyotrophic lateral sclerosis (ALS) [4], multiple sclerosis (MS), multiple system atrophy, and acute insults (but recently recognized as accompanied by secondary cell death processes) to the brain such as stroke [5] and traumatic brain injury (TBI) [6] have been and are currently under extensive investigation for cell therapy. However, the optimal route of stem cell administration for specific diseases remains to be fully determined. The delivery of stem cells intravenously is a less invasive strategy but it raises concerns about microemboli formation and may not fully distribute cells to the specific area of the injured brain [7]. Compared to intravenous, an intra-arterial approach is preferred due to the deviation of first pass effect which results in better crossing of cells into the brain while intracerebral transplantation is more invasive but facilitates graft survival in the area [7]. Thus, while invasive, this direct intracerebral approach would accelerate the

neurorestoration of grafted cells. The drawbacks of each method has placed the development of an effective strategy with good safety outcomes for cell transplantation an on-going clinical challenge in cell therapy [8]. Because clinical trials of stem cell therapy have reached certain disease indications, further discussion will place an emphasis on PD, HD, stroke, TBI, ALS, MS, and multiple system atrophy, with a focus on intracerebral grafts versus other routes of administration.

Parkinson's disease

While patients with PD start a therapeutic regime to control symptoms, it has also been reported that in the later course of the disease certain motor features in patients tend to become unresponsive to dopaminergic (DA) treatment [9] even when the patient responded well to available treatment from the beginning [3]. To this end, it is proposed that to enhance the quality of life and effectively slow the progression of the disease, stem cell therapy should be considered at the point when patients have the greatest response to their treatment therapy [9]. A more prompt decision should be considered for those patients who present themselves at a higher risk of developing worsening disabilities even quicker [9]. Intracerebral stem cell grafts are expected to integrate into areas deficient of dopaminergic neurons and restore the dopaminergic neurons that are no longer functional through the release of neurotrophic factors and differentiation, respectively [3]. This may merit the implantation of grafts at an early stage of the disease or as a simultaneous therapy along with DA treatment.

PD models used for experimental studies utilize mostly rodents and monkeys that have been subjected to 6-hydroxydopamine or 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) [10]. In any disease models, the ideal route of administration is one that allows minimal invasiveness [11] mostly being intravenous and intraarterially [12]. However, the peripheral delivery of differentiated cells (i.e., DA neurons for PD) remains sub-optimal, in that differentiated cells display low migratory capacity [13]. In addition, the blood brain barrier (BBB), while partially compromised in PD, may not be conducive for entry of peripherally administered cells to reach the brain target areas. PD models traditionally often used intracerebral grafts of DA neurons from the ventral mesencephalon of developing embryos that have resulted in the regain of some functional recovery but not all [14]. An improved intracerebral microtransplantation technique was introduced to lessen the traumatic effects of such invasive procedure [15]. However, due to the ethical issues surrounding the use of human fetal tissue, other sources of stem cells have been extensively studied [14]. Sources such as induced pluripotent stem cells (iPSCs) could yield an ample supply of DA neurons from which transplantation therapy could benefit from [14] due to the advances in cell reprogramming which has allowed precursor cells to be induced into a specific stem cell lineage through exposure of small molecule activators [16] or fate-determining factors [10]. Subjecting PSCs to activators of sonic hedgehog and canonical WNT, for example, produces midbrain DA neurons [16]. The engraftment of these neurons into mice and rat PD models result in the survival of the neurons and the complete restoration of amphetamine-induced rotation behavior [16]. To validate these results in a bigger species and use a closer estimate of the number of cells that would be needed for a human, MPTP-lesioned rhesus monkeys underwent the grafting of 5×10^7 precursor DA neurons in which survival of the cell grafts

were effectively observed a month after [16]. Similarly, transplantation of neural progenitors into the striatum of adult rats just 10 days after induced neural cell differentiation resulted in excellent cell survival with differentiation and graft position maintained in the host brain [17]. Moreover, morphology of the brain did not show any tumors, as this is a huge barrier to the efficacy of stem cells, or necrosis after 6 weeks, revealing that engraftment is tolerated [17]. Further studies have focused on reconstructing the nigrostriatal pathway by transplanting cells directly into the substantia nigra while other studies have focused on 'bridging' the substantia nigra to the striatum through the use of trophic factors so that the host environment can be favorable for continual directed neural growth and eventually, motor improvement [14]. Unfortunately, results from these studies are insignificant to translate to the clinic [14].

Since the first successful clinical cell transplantation in 1987, hundreds of PD patients have been enrolled in limited clinical trials [18]. After a couple of clinical studies in the U.S. using aborted human fetal ventral midbrain tissue reported unsuccessful outcomes, however, no longer were fetal cells used [18]. It is noteworthy that patients who were seen years after the transplantation revealed motor symptom improvements [10]. Whether the lack of success was due to challenges incurred in the procedure or from graft integration into host tissue, transplanted stem cells were just as prone to degenerate as host neurons, a reality that dampened the potential of stem cell therapy [18]. Currently, there are open clinical trials assessing the use of stem cells for PD. Outside of the U.S., researchers are using autologous adipose-derived stem cells and delivering the cells to the patient via catheter into the vertebral artery and intravenously [19]. The intravenous treatment of stromal vascular fraction (SVF) is still being evaluated as another alternative due to the stem cells [20] and growth factors [21] SVF has been known to carry that can be of benefit for PD patients. A current transplantation study, better known as the TRANSEURO project, in the U.K. is grafting allogenic dopaminergic neuroblasts obtained from fetal ventral mesencephalic tissue into the brain of PD patients in the hopes of replacing lost dopaminergic cells [22]. The main scope of this project was to devise a safe dissection methodology that would be implemented in clinical setting to provide maximum benefits to patients with minimum side effects [23]. This dissection methodology is now in use throughout various centers in the U.K [23]. The transplantation of healthy cells intracerebrally provides a direct approach to replacing the dead or degenerating neurons in the nigrostriatal pathway and consequently to restoring neural function [24], which has remained elusive to date. This approach, while being invasive as noted earlier as, circumvents the need for long-distance migration if delivered peripherally, thereby avoiding for the grafted cells to migrate throughout the circulatory system before reaching the brain [24]. Thus, the potential benefit of such direct transplantation compared to peripheral injections is the possibility of reconstructing the neuronal circuitry, which may allow halting the disease process, rather than merely providing palliative effects [24]. To this date, the use of intracerebral transplants of stem cells remains the widely accepted approach for PD, but neural circuitry reconstruction has been a challenge.

Huntington's disease

The genetic inheritance of HD allows predictive genetic testing, even in utero [25]; an aspect that is unique compared to other neurodegenerative diseases. Experimental therapies have been aimed at conserving the structural and functional integrity of the striatum in HD models to prevent the onset and potentially slow down the functional decline of HD patients in the future [25]. Most HD models undergo graft transplantation of choroid plexus (CP) after quinolinic acid (QA) lesions due to the cells' ability to exert influence in homeostatic processes of the brain and release trophic factors [25], but olfactory ensheathing cells (OECs) [26] and iPSCs [27] have also been studied. Engraftment of CP into QA lesioned rat striatums have demonstrated less motor impairment, decreased lesion volumes, and protection of specific neuronal populations from excitotoxic damage [28], as the mutant huntingtin is thought to affect the production of chemokines and neurotrophic factors from glial cells [27]. Even in HD primate models, CP has been shown to exert the same neuroprotective effects when transplanted into the caudate and putamen, with an extraordinary 5-fold difference of lesion size compared to control primates [25]. Recently, the usage of OEC's delivered to the brain after differentiation into glia by somatic cell nuclear transfer has been considered a promising potential as this would allow the differentiated glia to contain healthy mitochondria without any fear of immune rejection [26]. This "transcribrial route" would deliver the OEC's via the cribriform plate of ethmoid bone in which physical neurosurgical procedures would be bypassed to attain a successful stem cell graft [26]. iPSCs are not left behind in current HD rodent models as they too survive, migrate into injured striatum, differentiate into neurons and glial cells after transplantation [27]. These iPSCs decrease striatal atrophy, improve functional recovery, and may provide a beneficial environment that attracts glial cell activation and proliferation, slowing the excitotoxic damage to γ-aminobutyric acid (GABAergic) neurons in the striatum [27].

Transplantation of cell grafts in HD patients has demonstrated mixed results. Intrastriatal grafts of human fetal striatal tissue in three out of five patients with mild to moderate HD resulted in improved motor and cognitive functions when daily-life activities were assessed [29]. Brain images even showed high metabolic activity of the grafts, indicating functionality, compared to untreated HD patients [29]. Similarly in another study, pathologic assessment of two patients with HD who died 74 and 79 months after fetal neural transplantation showed neuronal grafts without pathologic characteristics of HD [30]. These results are promising, however, highly restricted integration between graft and host seems to be more of an obstacle to this therapeutic approach than would have been predicted from animal models and likely was the reason for the limited clinical benefit in these patients [30]. In another assessment, surviving grafts in three HD patients who underwent neural transplantation in the striatum a decade earlier did not show encouraging results [31]. These grafts had unhealthy neuron morphology with no survival of grafts in the caudate [31]. These results support the known conclusion that the caudate displays the greatest neuronal degeneration and astrogliosis in HD, which places a limit in the potential regions of cell grafting for this disease [31]. As these results differ significantly from 18-month and 6-year posttransplantation cases where healthy graft survival is reported, future trials of fetal-cell transplantation is deemed unjustifiable [31].

The conflicting data demonstrate that neuronal transplantation in HD provides a window of opportunity of several years with improvement and stability, but not a permanent cure for the disease [32]. Similar to PD, the use of fetal cell grafts for HD creates ethical and logistical issues. A minimally invasive procedure is likely more feasible and practical for HD in the clinic, but just like PD, the migration of grafted cells from periphery to the brain may be problematic since differentiated cells (in this case, GABAergic for HD) in general have low migratory potential [13]. A similar not-so-leaky BBB in HD [14] may present as an obstacle against peripherally transplanted cells to penetrate the brain and migrate towards the degenerated striatum. As discussed previously in PD, the use of intracerebral grafts, though invasive, remains the viable option for HD and the use of microtransplantation initially employed in PD may be extended to HD in order to reduce traumatic side effects of such intracerebral route of cell delivery. The microtransplantation approach allows the modification of the distribution of graft sites and the number of cells deposited per site, which can then influence the yield of functional neurons that arise within the transplant [33]. Multiple deposits produce an increased yield of striatal-like neurons, probably due to a greater graft–host border that exposes the cells to host trophic factors, compared to a single-tract microtransplantation [33]. This occurrence has an important value when fine-tuning cell based transplantation protocols for HD [33].

Current stem cell clinical trials for HD are scarce. Researchers have been focusing on measuring the changes over time in movement, thinking, behavior, brain imaging, blood and spinal fluid markers in early HD [34]. These recruited patients will then become candidates for a larger clinical study involving the intrastriatal delivery of mesenchymal stem cells (MSCs) in the hopes of advancing the current status of clinical trials for HD [34]. Analogous to intracerebral transplants of dopaminergic cells for PD, the repair of the neuronal circuitry by intracerebral transplantation of GABAergic cells for HD would bypass the unnecessary systemic migration of stem cells to other organs if administered peripherally, and may pose as an effective strategy to restore the functionality of the damaged striatal region of the HD brain [35]. Similar to PD, the intracerebral route of delivery of stem cells stands as the most efficacious route of stem cell delivery for HD.

Stroke

As stroke continues to be a leading cause of death in the U.S., the use of animal models to develop treatments is ever growing [36]. While the current treatment for ischemic stroke is dependent on a therapeutic time-window, it is vital to introduce novel treatments for the brain damage and physical disabilities that are often caused by it [36]. Moreover, treatment for hemorrhagic strokes has yet to be identified [36], prompting the extensive quest of researchers for a potential therapeutic regime. The most widely used method for inducing ischemia in animal models is thrombosis by middle cerebral artery occlusion (MCAO) [37]. This approach closely mimics human infarcts in terms of size and the structures affected [37]. Techniques other than MCAO in animals such as injection of endothelin 1 and different sizes of emboli also produces ischemic lesions and are used depending on protocol [37].

Off-the-shelf cryopreservable stem cell sources have a logistical and ethical advantage in future therapy over other freshly harvested stem cells for the treatment of cerebral ischemia primarily because of the abrupt onset and highly debilitative nature of the disease [38]. Cryopreserved stem cells that can be thawed and transplanted immediately allow a therapeutic window, which would significantly reduce the lapsed time between stroke and medical treatment [39]. In addition, because of the massive immune and inflammatory response associated with stroke, the identification of cells that do not further elicit such host response is likely to be more beneficial. To this end, the use of autologous cells seems appropriate. Bone marrow, peripheral blood, adipose tissue, and menstrual blood-derived cells have been studied in stroke models due to their ability to provide a source of autologous stem cells without ethical limitations, their expression of stem cell markers, and their ability to ameliorate stroke-induced behavioral and histological deficits [38, 40, 41]. Grafting cells that are known to produce vital trophic factors that protect against ischemic injury such as glial cell line derived neurotrophic factor (GDNF) or osteogenic protein-1 has also been proposed as potential treatment [42]. Indeed, transplantation of fetal kidney tissue reduced ischemic volume and behavioral deficits compared to adult kidney tissue, indicating that such fetal tissue grafts could serve as a cellular reservoir for important trophic factors to the injured brain [42]. Intrastriatal transplantation of mouse bone marrow stem cells (BMSCs) in MCAo models has also shown to dose-dependently restore cerebral blood flow and BBB permeability to near normal levels [43] while the grafting of CP isolated from adult rats and encapsulated within alginate microcapsules have resulted in a reduction of infarct volume and motor deficits, demonstrating the therapeutic potential of such cells for stroke [44].

The transfection of teratocarcinoma-derived Ntera2/D1 neuron-like cells (NT2N cells) with the transcription factor Nurr1 has provided a different source of cells for stroke [45]. The NT2N.Nurr1 cell line displays an expedited neuronal commitment, secretes a high level of GDNF [45], and when transplanted into the rodent stroke brain, significant attenuation of behavioral impairments after stroke are seen [46]. The postulated mechanism by which this efficacious cell line mediates observed benefits is through neurotrophic factor secretion [46] along with its highly potent neuronal differentiation commitment [45]. The intravenous transplantation of amniotic fluid-derived stem cells (AFSCs) in MCAo models has also attenuated stroke-induced behavioral and histological deficits, possibly via enhancement of endogenous repair mechanisms as increased cell proliferation and neuronal differentiation in the two neurogenic sites, subventricular zone (SVZ) and dentate gyrus (DG), have been reported [47]. Different dosages of intracerebral human neural stem cell (NSC) transplants in the striatum reveal a dose-dependent recovery from both motor and neurological deficits in MCAo rats [48]. Even though reduction in infarct volume was not observed in this study, transplantation did produce behavioral improvements, supporting the use of NSC lines for stroke therapy [48]. Despite the observed functional recovery in stroke models, the mechanism of action underlying stem cell therapy remains not well understood [49]. Although, robust vasculogenesis and neurogenesis in endothelial cell-transplanted stroke animals suggests that targeting vascular repair sets in motion a regenerative process in experimental stroke possibly via the vascular endothelial growth factor (VEGF) pathway [49].

Although extensive animal data show the potential of stem cell grafts in stroke, transplantation procedures for specific cells need to be optimized. As a case in point, the effects of timing and routes of transplantation on survival and functional benefits of human bone-marrow-derived CD133+ cells in stroke rats can serve as a guideline to tailoring transplantation procedures [50]. In the study, both immediate and delayed intracerebral transplantation resulted in a localized graft survival with a reduction of motor deficits while graft survival was only detected in delayed intravenous transplantation and behavioral improvement only apparent in immediate intravenous transplantation [50]. These results therefore highlight the need to modulate transplantation procedures in cell therapy so that they can be efficaciously applied to humans [50].

The FDA approved clinical trial of NT2N cell transplantation in 1998 on 12 stroke patients showed for the first time that cell transplantation for stroke is a potentially feasible and generally safe approach [51]. In a postmortem brain assessment of a phase I clinical stroke trial patient implanted with NT2N neurons adjacent to a lacunar infarct 27 months after surgery showed neurons in the graft site with no evidence of a neoplasm [52]. These findings indicate that the grafts survive for >2 years without deleterious effects [52] but because of the cells' high proliferative property due to its cancer origin, the major limiting factor for initiating a large clinical trial is the concern that these cells may revert to a neoplastic state over time after transplantation [45]. Because of this, safer stem and progenitor cells have been explored as alternative graft sources since then [51]. Human neuronal cell transplantation has been transplanted in patients with basal ganglia stroke [53] and in patients with motor deficits due to stroke [54]. Positron emission tomography (PET) scans at 6 months showed improved fluorodeoxyglucose uptake at the implant site [53] and improved activities of daily living [54]. Although there was no evidence of significant benefit in motor function, neuronal transplantation proved to be safe and feasible in patients with stroke [54].

Currently, ongoing clinical trials have been focused on evaluating the efficacy of the intracerebral injection of autologous bone marrow mesenchymal stem cells in patients with chronic stroke [55], a single treatment administration of autologous cultured bone marrow stromal cells and endothelial progenitor cells [56], allogeneic stem cells from adipose tissue in acute ischemic stroke patients [57], and the therapy composed of cells derived from a patients' own adipose tissue that are isolated within approximately 1 hour and immediately delivered back to the patient using a catheter delivery system [58]. For acute stroke, the preferred route of administration is intravenous transplantation, while for the chronic stroke, the indicated route of cell administration is via intracerebral route. For acute stroke, the chemoattractant cues in the host ischemic brain are upregulated, thus transplanted cell from periphery are guided to hone to the site of injury. Because, as discussed above, stroke is associated with massive inflammation and an immune response early on, intracerebral transplantation in the early stage may not be indicated since such traumatic and invasive procedure is likely to exacerbate the already inflamed brain. Therefore, a minimally invasive procedure is more appropriate in the early stage. This is not the case in the chronic stroke whereby the chemoattractant signals wane over time, thus peripherally administered cells will not find their way to the brain. When the patient is more stabilized in the chronic stage, then the intracerebral approach seems to be more advantageous to deliver the cells to deep

pockets of the brain. However, there are a few studies demonstrating the successful migration of stem cells to the ischemic brain when delivered with long delay post-insult [59]. Optimizing the timing of instrastriatal transplantation in stroke may benefits from lessons learned in HD and PD, which also targeted the striatum for intracerebral transplantation. Moreover, as discussed in previous sections, the grafting of cells to serve as a vehicle to deliver growth factors within and around the ischemic penumbra may offer an option to further enhance cell transplantation-mediated therapeutic benefits in stroke.

Traumatic Brain Injury

The number of TBIs that contribute to deaths and permanent disability increases every year [60]. An estimated 5.3 million Americans are living with a TBI-related disability, which affects all aspects of an individual's life [60]. Having to rely on early management and primary interventions [60] to help limit the impact of TBI has long interested researchers to find alternatives for the treatment of TBI. Animal models of TBI have focused on the fluid percussion and controlled cortical impact (CCI) injury models [61]. The CCI model produces injury to the brain similar to clinical TBI in humans, such as cell death, edema, ischemia, excitotoxicity, and altered gene expression [61]. Thus, cell grafts will have the greatest potential for success when administered soon after an injury [62].

Intrastriatal transplantation of bone marrow stromal cells have produced functional benefits associated with the restoration of cerebral blood flow and BBB permeability near normal levels in rat models of TBI [63]. Acute intracerebral transplantation of neural stem cells in moderate TBI rat models has also proven to be beneficial in improving functional recovery while delayed transplantation in various studies has shown mixed results with some showing motor but not cognitive recovery or vice versa [64]. The intravenous administration of human adipose-derived stem cells (hADSCs) in mild models of TBI using young (6 months) and aged (20 months) rats show an age effect where significant amelioration of motor and cognitive functions occur in young, but not aged, groups [65]. Significant reduction in cortical damage and hippocampal cell loss was observed in young rats, whereas less neuroprotection was detected in the aged rats [65]. Fluorescent imaging revealed labeled hADSCs in peripheral organs and brain after TBI, most notably reduced migration to the aged spleen [65]. hADSCs therefore are promising therapeutic cells for TBI as the spleen is known to be necessary for the neuroprotective action of MSCs after TBI pathology even with the reduced efficacy in aged rats, which may in part result from decreased homing of the cells to the spleen [65].

The stimulation and mobilization of endogenous stem/progenitor cells from the bone marrow through granulocyte colony stimulating factor (G-CSF) poses as an attractive therapeutic intervention for chronic TBI [66]. The combined therapy of human umbilical cord blood cells (hUCBCs) and G-CSF at the acute stage of TBI displays dampened neuroinflammation while enhancing endogenous neurogenesis and reducing hippocampal cell loss [66]. Vigorous and long-lasting recovery of motor function accompanied the combined therapy, which was short-lived in the monotherapy of hUCBCs or G-CSF conditions [66]. Possible mechanisms yielding the beneficial outcomes of the combined therapy include G-CSF producing a conducive microenvironment for the transplanted

hUCBCs to integrate with the host tissue, G-CSF might have directed the migration of endogenous stem cells mobilized from the bone marrow to the site of injury, and/or the grafted hUCBCs might have released growth factors secreted by hUCB grafts [67]. This synergistic combinatorial effect allows a much more beneficial functional outcome than a single, stand-alone treatment [67].

A unique mechanism of action exerted by stem cells in the repair of TBI involves their ability to harness a biobridge between the neurogenic SVZ niche and the injured brain site or cortex [68]. The intracerebral transplantation of cultured notch-induced human bone MSCs (referred to as SB623) after 7 days in a TBI animal model facilitated migration of endogenous cells via a biobridge, which expressed high levels of extracellular matrix metalloproteinases (MMPs) and characterized initially by a stream of graft cells [68]. In a matter of time, only few to non-detectable grafts overgrown by newly formed host cells is observed, implicating a novel property of stem cells [68]. MMPs then facilitate the movement of the newly formed cells from the neurogenic niche into the injured tissue as the grafts manifest themselves as pathways for trafficking the migration of host neurogenic cells that disappear once this biobridge is formed [68]. These novel observations expand our current knowledge of stem cell biology since the two major schools of discipline in stem cell repair mechanism include the concept of “cell replacement” and bystander effects of “trophic factor secretion” [68]. Based on the resulting observations, similar biobridges to facilitate the migration of cells across tissues remains to be explored [68]. However, stem cells such as umbilical cord blood, peripheral blood, and adult brain have demonstrated to alter levels and functions of MMPs and extracellular matrix metalloproteinases (ECMs), which would suggest their potential to similarly serve as biobridges as seen with SB623 [68]. Like any other stem cell, a limiting factor for endogenous repair is the successful migration of the newly formed host cells to reach the injured brain area so future studies will need to focus on assessing the efficacy and safety of SB623 in chronic TBI to further optimize transplantation protocol in clinical trials [68].

Stem cell therapy may offer a better functional outcome when employed in the acute stage of TBI, but such stem cell-based regenerative medicine may still target TBI at the chronic stage. Similar to the diseases discussed, the microenvironment in the mild and severe stages of TBI differs substantially. In essence, a mild TBI microenvironment is more favorable for stem cell survival compared to severe TBI [64] due to the significant increase in neurotrophic factors in the acute stage versus their absence in severe TBI [69]. Intravenous route of administration, which circumvents the trauma associated with the invasive intracerebral transplantation route, will be more appropriate for the acute stage of TBI. In contrast, when chemoattractant signals have waned during the chronic stage of TBI and when the patient is more stabilized, the intracerebral route of transplantation may seem to be more conducive for brain repair.

Current clinical trials involve cell transplantation in TBI patients by intrathecal and intravenous infusion for acute and chronic TBI of autologous bone marrow progenitor cells (BMPC) [70, 71, 72, 73] and treatment with stem cell mobilization agents, such as erythropoietin [74] and G-CSF [66], to increase endothelial progenitor cells (EPCs). As noted above, TBI may also extrapolate findings from the intracerebral transplantation

experience from PD, HD and stroke clinical trials. To this end, stereotaxic transplantation of the stem cells into the peri-impact area may allow the rescue of dying neural cells during the secondary cell death associatd with TBI, via a mechanism that similarly involves trophic factor secretion from the transplanted cells as shown in PD, HD, and stroke.

Amyotrophic Lateral Sclerosis

The pathology of ALS is characterized by the progressive degeneration of upper and lower motor neurons [75]. Treatments currently available for ALS have minimal effects on the disease progression and are limited to riluzole, noninvasive positive pressure ventilation, and nutritional support [76]. The genetic anomaly of the superoxide dismutase 1 (SOD1) gene found in the inherited form of ALS has allowed the creation of the glycine 93 changed to alanine (G93A) SOD1 transgenic mouse and rat experimental models [75]. Indeed, intraspinal injections of bone marrow mononuclear cells have been able to ameliorate the course of ALS in these models due to the cells acting as pumps of trophic factors that keep motor neurons functional [77]. Furthermore, these stem cells have dose-dependent effects on SOD1 mice via intrathecal injection [78]. Human spinal cord-derived stem cells (hSSCs) are also of particular interest in ALS because they are thought to express amino acid transporters and secrete neurotrophic factors [79]. ALS patients are found to have increased levels of glutamate accumulated in the brain and spinal cord, which is hypothesized to be caused by a decrease in the glutamate transporter [79]. hSSCs can potentially reduce the toxicity of accumulated glutamate and be of benefit to ALS patients [79]. Optimization of an immunosuppressive protocol for spinal grafting of hSSCs to the lumbar spinal cord of a rat ALS model so that reliable assessment of potential long-term therapeutic effects associated with cell replacement is made concluded that the highest density of grafted cells was seen in animals treated with FK506 and mycophenolate [75]. This suggests that a combined, systemically delivered immunosuppression regimen including tacrolimus (FK506) and mycophenolate can significantly improve survival of hSSCs after intraspinal transplantation in G93A SOD1 rats [75].

Lumbar spinal cord injection of HSSCs has also been tested as a novel therapy for patients with ALS [76]. In a surgical procedure on 12 patients, no major surgical complications were incurred nor did the grafts exert any identifiable toxicities in the spinal cord [76]. Moreover, further quantitative clinical assessments of the participants of this study showed no evidence of disease progression, proving the safety for this phase I trial [76]. However, in order to extend the life of patients, therapeutic intervention will need to be focused at the level of the cervical spinal cord where motor neurons affecting respiratory function are located [76]. Even OECs have been engineered to produce neurotrophins to explore other potential transplantation strategies for ALS patients [26].

Most of the ongoing and completed clinical trials involve the intraspinal, intramuscular, and/or intrathecal infusion of autologous BMSCs [77,78,80, 81], umbilical cord mesenchymal stem cells (UCMSCs) [82], human spinal cord-derived neural stem cells [79, 83], autologous hematopoietic stem cells [84], autologous MSCs [85,86, 87], human neural stem cells [88], HLA-haplo matched allogenic bone marrow derived stem cells [89], iPSCs [90], and in tandem with G-CSF as adjunct therapy in the hopes of enhanced benefits [91].

Multiple Sclerosis

While there is no cure for multiple sclerosis to date, stem cell therapy has gained much attention to promote the remyelination of axons in patients [92]. Studies injecting neuronal stem cells intraventricularly have been shown to suppress MS by producing immunomodulating effects [92]. These neurospheres *in vitro* yield oligodendrocyte progenitors that mature into oligodendrocytes and migrate into the inflamed white matter where they develop glial lineages markers following transplantation, halting the progression of MS [92]. Studies injecting bone marrow mesenchymal stromal cells (BM-MSCs) intravenous or intraventricular in MS mice models showed similar results with cells homing to the inflamed lesions and differentiating into astrocytes, neuronal, and glial cells [92]. Moreover, a decrease in lymphocytic infiltrations and a significant preservation of the axons are also observed [92]. A study comparing human embryonic stem cell derived mesenchymal stromal cells (hE-MSCs) and (BM-MSCs) demonstrated hES-MSCs capable of halting the progression of MS in the MS mouse model while BM-MSCs only producing a marginal effect [93]. This superiority may be due in part to the lower expression of IL-6 and the greater ability of hES-MSCs to cross the BBB/BSCB and migrate into inflamed CNS tissue relative to BM-MSCs [93]. Moreover, histological assessment also demonstrated hES-MSCs protected against demyelination without affecting the number of surviving axons, which may be an important factor that contributes to remyelination of axons that have already lost their myelin [93].

Wharton's jelly-derived stromal cells (WJ-MSCs) display a low immunogenic phenotype, express neurotrophic factors and anti-inflammatory molecules, and inhibit the proliferation of activated T cells [94]. WJ-MSCs injected into an MS model ameliorated disease symptoms through a reduction in autoantigen-induced T cell proliferation and trophic support [94]. In 23 MS patients, human UCMSCs have been IV infused three times in a 6 week period [95]. Noteworthy, a shift from type 1 T helper (Th1) to type 2 T helper (Th2) immunity in UCMSC treated patients was observed, demonstrating a high potential for hUC-MSC treatment of MS [95].

At this time, limited clinical therapies for MS include autologous BMSCs [96] and umbilical cord tissue derived mesenchymal stem cells [97].

Multiple System Atrophy

Multiple system atrophy (MSA) is characterized by autonomic dysfunction, parkinsonism, cerebellar ataxia, and pyramidal signs in any combination, with autonomic dysfunction being an important component in the diagnosis [98]. Disease progression is rapid with a fatal prognosis, therefore, it is of dire need to develop treatment to delay disease progression of MSA. [98] In a study where autologous MSC therapy in patients with MSA demonstrated a delayed progression of neurological deficits compared with placebo treatment, intraarterial infusion was the chosen as the route of administration of MSCs to provide an increased migration and a more diffuse distribution pattern so that a larger number of engrafted cells can be homed to the brain [98]. Laboratory findings suggest the safety and feasibility of cell therapy, supporting the use of autologous MSC therapy for MSA.

Expert Commentary & Five-Year Review

While the brain is considered an immunopriviledged site, transplanting stem cells in neurological disorders need to consider host rejection as an important parameter in the successful clinical outcome of cell therapy. Allogenic transplantation does not always need immune suppression, however, as cells differentiate they may become more immunogenic [99]. One such study where immunosuppression with cyclosporine is not sufficient to prevent rejection of xenografts is evident in the transplantation of DA human embryonic stem cells (hESCs) into a Parkinsonian primate brain by the presence of large numbers of CD68 and CD45 cells in the injection sites and surrounding the grafts [100]. Because hES cell-derived neural progenitors will mature to neurons and glia in the rodent brain in about 3 months and mature neurons and glia express higher levels of major histocompatibility complex (MHC) antigens, the immune system mounts a response to the xenograft [100]. If the cells were to maintain in the progenitor stage and continue to divide, they will express a low level of MHC antigens and not elicit immune rejection in the presence of cyclosporine A [100]. Moreover, immunogenicity becomes unpredictable in cases where cells that are not intended to be used for the same essential function in the recipient as in the donor or when administered at non-physiological sites [99]. Human leukocyte antigen (HLA)-matching of donor and recipient may diminish the risk of graft-versus-host disease (GVHD), but this is often not readily achievable [99]. In a different study without systemic immunosuppression, porcine neuroblasts (pNb) cotransplanted with rat MSC intracerebrally can survive up to 120 days [101]. The loss of xenografted neurons is attributed to a host immune response at 4–6 weeks [101]. Interestingly, in the presence of MSCs, cellular and molecular events that are usually induced by intracellular transplantation in the rat brain are not observed or are strongly reduced, supporting the notion of MSC as potent immunosuppressors [101]. Human MSCs alter the maturation of dendritic cells as well as their ability to present antigens to T cells, they are also able to inhibit T-cell proliferation, and to affect the differentiation of B cells into plasmocytes [101. Thus, in optimizing cell therapy, it is important to consider the relationship between MSCs and the immune system.

The extracellular milieu of the transplanted stem cells may also influence its tumorigenic potential. Extracting stem cells from the embryo environment and enforcing it into *in vitro* culture has been thought to cause the increased tumorigenic potential of ESCs when compared to the originator cells (the inner mass of early blastocysts) [99]. The host species to which the cells are administrated is also an important factor determining the rate of teratoma formation [99]. When transplanted into a homologous species mouse ESC caused highly malignant teratocarcinomas at the site of administration, while xenotransplantation in rats resulted in migration and differentiation of the mouse ESC [99]. Additionally, stem cells can also affect the growth of existing tumour cells *in vitro* and *in vivo* [99]. The outcomes depend on the nature of the cancer cells, the characteristics of the used MSCs, the integrity of the immune system, and on the timing and site of injection [99]. MSCs either provide a supportive stroma favorable for tumor growth or may reduce immune rejection of the tumor cells which may allow continued tumor growth [99]. Potential risk of stimulation of growth of a previously undetected tumor by MSCs must be considered when administering these cells to a patient [99].

Another risk factor to consider is the homing of the administered stem cells. MSCs are known to home to specific tissues, e.g. the bone marrow, muscle, or spleen, particularly when the tissues are damaged or under pathological conditions [99]. It is unclear where the non-engrafted stem cells go to and the risks associated with distribution to undesired tissues are unknown [99]. Thus, the risk of any engraftment and its effects remains unpredictable and should be taken into account [99].

The potential number of cells needed for the beneficial effect is generally debatable, however, given the very low rate of retention and possible low cell survival, high survival rate of cells may not be required for obtaining maximal clinical benefit [99]. When MSCs are infused systemically, they are trapped into capillary beds of various tissues, especially the lungs [102]. Delivery of MSCs via the internal carotid artery significantly improved their migration and homing in the injured brain compared with injection via the femoral vein [102]. However, delivery of cells in an artery may lead to microvascular occlusions [102]. Cells may form aggregates that could cause pulmonary emboli or infarctions after infusion [99]. Injection in the portal vein or directly at the site of injury may circumvent this problem [99].

Since the inception of cell transplantation for PD in the 1980s, the last past decade has seen exponential growth of stem cell therapy as an efficacious treatment option for neurological disorders. Experimental and clinical data have given scientists knowledge on how to expand on transplant methodologies in an effort to improve outcomes in both animal models and patients with neurological disorders. Despite scientific advances that led to limited clinical applications of cell therapy, the challenges of implementing large-scale stem cell therapy in the clinic remain, requiring transplant regimen optimization and investigations into mechanisms of action underlying this treatment. Challenges that prevent the clinical entry of stem cells include but are not limited to the inadequacies in the transplantation protocol and/or study design, high tissue variability, lack of scalability, ethical concerns, inability to obtain an epidemiologically meaningful quantity of tissue, graft-induced adverse effects (such as dyskinesia in PD patients), and the lack of consensus in establishment of acceptable levels of efficacy and toxicity (e.g., graft persistence vs. acute graft survival, graft-derived growth factor secretion vs. graft-host integration and synaptic network) [20]. Finding the optimal transplant regimen is further magnified in view of recent rise in “medical tourism” of cell therapy, which is likely to result in mixed outcomes and hype due to these unregulated clinical trials or anecdotal reports. Here, we provide a basis for pursuing intracerebral route under appropriate conditions, such as when chemoattractant signaling cues wane in the chronic stage for the disease and when the blood brain barrier is prohibitive in allowing entry of stem cells from the periphery to the brain. Vis-à-vis preclinical investigations comparing the therapeutic potential between intracerebral and peripheral cell transplantation for specific neurological disorders will be needed to reveal the optimal cell delivery route that is safe and effective [47,103,104]. To this end, overexpression of migratory proteins in the donor cells may facilitate proper deposition of cells in discreet brain areas [47,103,104]. The cellular and molecular changes in the brain microenvironment that accompany the progression of the disease warrant a careful consideration of cell delivery route that will facilitate enhanced regeneration of the injured brain.

While this review focuses on the advancements of stem cell therapy from the bench to bedside it is important to highlight that stem cell therapy is still in its infancy. It is not regarded as the standard clinical treatment for neurological diseases even though extensive laboratory data may support its safety and efficacy. Thus, misconceptions regarding stem cell therapy should be deliberately contemplated. As discussed above, medical tourism offers medical treatment to patients who are willing to travel and spend their resources in areas other than their homeland, where standard treatment has not passed regulatory guidelines (such as those mandated by the US Food and Drug Administration or FDA), and where the opportunity of such cellular therapy may prove to be highly expensive [105]. However, the development of cell therapy for a disease is best done in the setting of extensive clinical trials and in a structured regulatory framework so that safety considerations, professional peer review, and the management of patient rights and obligations are considered and addressed [106]. This rigorous regulatory approach is necessary for any novel treatment, including stem cell therapy, in order to ensure safety of the patients. Moreover, it is valid to question whether practitioners are following existing regulations and whether patients are being held to false promises [106]. Unfortunately, such regulatory oversight is not a worldwide governing policy, jeopardizing patients' protection, in that patients who are blinded by the hype of stem cell therapy travel to countries with unregulated cellular therapies to avail of this treatment and likely exposing themselves to unnecessary harm. Of note, despite strict US FDA regulations, a few small companies and private clinics in the US have tried to market cell therapies to desperate patients. Many more deceitful businesses outside the US, like Mexico and China, have targeted these patients for financial gain with unproven cell therapy applications [107]. Interestingly in recent years, a stem cell registry of clinical trials (similar to the clinicaltrials.gov archival system) has been adopted by the Chinese government in an effort to regulate clinical application, but guidelines on quality control and necessary translational research needed to gain regulatory approval remains to be approved by the government [107]. With this in mind, it is important for patients to be vigilant against misconceptions and to recognize hype from hope associated with stem cell therapy. Stem cell therapy remains as an experimental treatment and is not a magic bullet for treating for neurological disorders.

In conclusion, we present that one of the key factors in translating cell therapy for CNS disorders entails carefully planned experiments in the laboratory assessing the safety and effectiveness of cell delivery routes in animal models that closely mimic the clinical scenario. To this end, the intracerebral route of cell transplantation while previously viewed as invasive may be considered as a safe and effective cell delivery approach for specific brain disorder indications after rigorous preclinical studies that equally incorporate assessments of the timing of delivery and cell dose in appropriate disease models.

Acknowledgments

The authors gratefully acknowledge the support and critical discussion offered by their colleagues at the USF Center of Aging and Brain Repair.

References

Papers of special note have been highlighted as:

*** = of interest**

1. McGovern Institute For Brain Research at MIT. Cambridge, MA: McGovern Institute For Brain Research; 2014. Brain disorders: by the numbers. Available at: http://mcgovern.mit.edu/brain-disorders/by-the-numbers
2. Harvard NeuroDiscovery Center. Boston, MA: President and Fellows of Harvard College; 2014. The challenge of neurodegenerative diseases. Available at: http://www.neurodiscovery.harvard.edu/challenge/challenge_2.html
3*. Lindvall O, Barker RA, Brustle O, et al. Clinical translation of stem cells in neurodegenerative disorders. Cell Stem Cell. 2012; 10:151–55. Overview of the challenges delaying the use of stem cells in therapy for neurodegenerative diseases. [PubMed: 22305565]
4. Lunn JS, Sakowski SA, Hur J, et al. Stem cell technology for neurodegenerative diseases. Ann Neurol. 2011; 70:353–61. [PubMed: 21905078]
5. Chaudhuri, A.; Bhattacharya, N. Human Fetal Tissue Transplantation. London: Springer London; 2013.
6. Tajiri N, Duncan K, Antoine A, et al. Stem cell-paved biobridge facilitates neural repair in traumatic brain injury. Front Syst Neurosci. 2014 published online 24 June 2014. 10.3389/fnsys.2014.00116
7. Meamar R, Dehghani L, Ghasemi M, et al. Stem cell therapy in stroke: a review literature. Int J Prev Med. 2013; 4:139–46.
8. Lim JY, Jeong CH, Jun JA, et al. Therapeutic effects of human umbilical cord blood-derived mesenchymal stem cells after intrathecal administration by lumbar puncture in a rat model of cerebral ischemia. Stem Cell Res Ther. 2011; 2:1–13. [PubMed: 21241459]
9. Schuepbach WM, Rau J, Knudsen K, et al. Neurostimulation for Parkinson's disease with early motor complications. N Engl J Med. 2013; 368:610–22. [PubMed: 23406026]
10. Ambasudhan R, Dolatabadi N, Nutter A, et al. Potential for cell therapy in Parkinson's disease using genetically programmed human embryonic stem cell-derived neural progenitor cells. J Comp Neurol. 2014; 522:2845–56. [PubMed: 24756727]
11. Glover LE, Tajiri N, Weinbren NL, et al. A step-up approach for cell therapy in stroke: translational hurdles of bone marrow-derived stem cells. Transl Stroke Res. 2011 published online 18 November 2011. 10.1007/s12975-011-0127-8
12. Lescaudron, L.; Rossignol, J.; Dunbar, GL. Stem cells and neurodegenerative diseases. London: CRC Press; 2014.
13*. Yasuhara T, Matsukawa N, Hara K, et al. Transplantation of human neural stem cells exerts neuroprotection in a rat model of Parkinson's disease. J Neurosci. 2006; 26:12497–511. Discusses the concept of multi-pronged therapeutic mechanisms underlying intracerebral transplantation of neural stem cells. [PubMed: 17135412]
14. Ghosh B, Zhang C, Smith GM. Bridging between transplantation therapy and neurotrophic factors in Parkinson's disease. Front Biosci. 2014; 6:225–35.
15*. Nikkah G, Cunningham MG, Cenci MA, et al. Dopaminergic microtransplants into the substantia nigra of neonatal rats with bilateral 6-OHDA lesions. I. Evidence for anatomical reconstruction of the nigrostriatal pathway. J Neurosci. 1995; 15:3548–61. Provides feasibility and efficacy of a high-breed microtransplantation approach for intracerebral delivery of the cells. [PubMed: 7538563]
16*. Kriks S, Shim JW, Piao J, et al. Dopamine neurons derived from human ES cells efficiently engraft in animal models of Parkinson's disease. Nature. 2011; 480:547–51. Discusses the engraftment of embryonic derived-dopaminergic cells in an animal model of Parkinson's disease. [PubMed: 22056989]
17. Kirkeby A, Grealish S, Wolf DA, et al. Generation of regionally specified neural progenitors and functional neurons from human embryonic stem cells under defined conditions. Cell Rep. 2012; 1:703–14. [PubMed: 22813745]
18. Tarazi FI, Sahli ZT, Wolny M, Mousa SA. Emerging therapies for Parkinson's disease: from bench to bedside. Pharmacol Ther. 2014; 144:123–33. [PubMed: 24854598]
19. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. Study to assess the safety and effects of autologous adipose-derived stromal in patients with parkinson's

disease. Available at: https://clinicaltrials.gov/ct2/show/NCT01453803?term=parkinson+stem+cell&rank=5

20. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. Outcomes data of adispose stem cells to treat Parkinson's disease. Available at: https://clinicaltrials.gov/ct2/show/NCT02184546?term=parkinson+stem+cell&rank=6
21. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. Safety and clinical outcomes study: SVF deployment for orthopedic, neurologic, urologic, and cardiopulmonary conditions. Available at: https://clinicaltrials.gov/ct2/show/NCT01953523?term=parkinson+stem+cell&rank=11
22. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. TRANSEURO Open Label Transplant Study in Parkinson's Disease. Available at: http://clinicaltrials.gov/ct2/show/record/NCT01898390
23. CORDIS. Cambridge, UK: CORDIS; 2014. TRANSEURO report summary. Available at: http://cordis.europa.eu/result/rcn/52712_en.html
24. Fegert, JM.; Fangerou, H.; Trapp, T. Implanted Minds: The Neuroethics of Intracerebral Stem Cell Transplantation and Deep Brain Stimulation. Bielefeld: Transcript Verlag; 2011.
25. Emerich DF, Thanos CG, Goddard M, et al. Extensive neuroprotection by choroid plexus transplants in excitotoxin lesioned monkeys. Neurobiol Disease. 2006; 23:471–80.
26. Baig AM. Designer's microglia with novel delivery system in neurodegenerative diseases. Med Hypotheses. 2014; 83:510–12. [PubMed: 25146247]
27. Mu S, Wang J, Zhou G, et al. Transplantation of induced pluripotent stem cells improves functional recovery in Huntington's disease rat model. PLoS One. 2014 published online 23 July 2014. 10.1371/journal.pone.0101185
28. Borlongan CV, Thanos CG, Skinner SJ, et al. Transplants of encapsulated rat choroid plexus cells exert neuroprotection in a rodent model of Huntington's disease. Cell Transplant. 2007; 16:987–92. [PubMed: 18351014]
29*. Bachoud-Levi AC, Remy P, Nguyen JP, et al. Motor and cognitive improvements in patients with Huntington's disease after neural transplantation. Lancet. 2000; 356:1975–79. Discusses therapeutic benefits of intracerebral grafts in Huntington's disease. [PubMed: 11130527]
30. Keene CD, Sonnen JA, Swanson PD, et al. Neural transplantation in Huntington's disease:long term grafts in two patients. Neurology. 2007; 68:2093–98. [PubMed: 17562830]
31. Cicchetti F, Saporta S, Hauser RA, et al. Neural transplants in patients with Huntington's disease undergo disease-like neuronal degeneration. Proc Natl Acad Sci U S A. 2009; 106:12483–88. [PubMed: 19620721]
32. Bachoud-Levi AC, Gaura V, Brugieres P, et al. Effect of fetal neural transplants in patients with Huntington's disease 6 years after surgery: a long-term follow-up study. Lancet Neurol. 2006; 5:303–09. [PubMed: 16545746]
33. Jiang W, Buchele F, Papazoglou A, et al. Multitract microtransplantation increases the yield of DARPP-32-positive embryonic striatal cells in a rodent model of Huntington's disease. Cell Transplant. 2011; 20:1515–27. [PubMed: 21176402]
34. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. A pre-cellular therapy observational study in early Huntingson's disease (PRE-CELL). Available at: https://clinicaltrials.gov/ct2/show/study/NCT01937923?term=huntington%27s+disease+stem+cell&rank=2
35. Kim M, Lee ST, Chu K, Kim SU. Stem cell-based therapy for Huntington's disease: a review. Neuropathology. 2008; 28:1–9. [PubMed: 18069970]
36. National Heart, Lung, and Blood Institute. Bethesda, MD: National Heart, Lung, and Blood Institute; 2014. What is stroke. Available at: https://www.nhlbi.nih.gov/health/health-topics/topics/stroke/
37. Casals JB, Pieri NC, Feitosa ML, et al. The use of animal models for stroke research: a review. Comp Med. 2011; 61:305–13. [PubMed: 22330245]
38. Rodrigues MC, Glover LE, Weinbren N, et al. Toward personalized cell therapies: autologous menstrual blood cells for stroke. J Biomed Biotechnol. 2011 published online 20 November 2011. 10.1155/2011/194720

39. Borlongan CV, Tajima Y, Trojanowski JQ, et al. Cerebral ischemia and CNS transplantation: differential effects of grafted fetal rat striatal cells and human neurons derived from a clonal cell line. Neuroreport. 1998; 9:3703–09. [PubMed: 9858383]
40. Borlongan CV, Kaneko Y, Maki M, et al. Menstrual blood cells display stem cell-like phenotypic markers and exert neuroprotection following transplantation in experimental stroke. Stem Cells Dev. 2010; 19:439–51. [PubMed: 19860544]
41. Kaneko Y, Dailey T, Weinbren NL, et al. The battle of the sexes for stroke therapy: female- versus male- derived stem cells. CNS Neurol Disord Drug Targets. 2013; 12:405–12. [PubMed: 23469849]
42. Chiang YH, Lin SZ, Borlongan CV, et al. Transplantation of fetal kidney tissue reduces cerebral infarction induced by middle cerebral artery ligation. J Cereb Blood Flow Metab. 1999; 19:1329–35. [PubMed: 10598937]
43. Borlongan CV, Lind JG, Dillon-Carter O, et al. Bone marrow grafts restore cerebral blood flow and blood brain barrier in stroke rats. Brain Res. 2004; 1010:108–16. [PubMed: 15126123]
44. Borlongan CV, Skinner SJ, Geaney M, et al. CNS grafts of rat choroid plexus protect against cerebral ischemia in adult rats. Neuroreport. 2004; 15:1543–47. [PubMed: 15232280]
45. Hara K, Yasuhara T, Maki M, et al. Neural progenitor NT2N cell lines from teratocarcinoma for transplantation therapy in stroke. Prog Neurobiol. 2008; 85:318–34. [PubMed: 18514379]
46. Hara K, Matsukawa N, Yasuhara T, et al. Transplantation of post-mitotic human neuroteratocarcinoma overexpressing Nurr1 cells provides therapeutic benefits in experimental stroke: in vitro evidence of expedited neuronal differentiation and GDNF secretion. J Neurosci Res. 2007; 85:1240–51. [PubMed: 17335085]
47. Tajiri N, Acosta S, Glover LE, et al. Intravenous grafts of amniotic fluid-derived stem cells induce endogenous cell proliferation and attenuate behavioral deficits in ischemic stroke rats. PLoS One. 2012 published online 17 Aug 2012. 10.1371/journal.pone.0043779
48. Tajiri N, Quach DM, Kaneko Y, et al. Behavioral and histopathological assessment of adult ischemic rat brains after intracerebral transplantation of NSI-566RSC cell lines. PLoS One. 2014 published online 10 March 2014. 10.1371/journal.pone.0091408
49. Ishikawa H, Tajiri N, Shinozuka K, et al. Vasculogenesis in experimental stroke after human cerebral endothelial cell transplantation. Stroke. 2013; 44:3473–81. [PubMed: 24130140]
50. Borlongan CV, Evans A, Yu G, Hess DC. Limitations of intravenous human bone marrow CD133+ cell grafts in stroke rats. Brain Res. 2005; 1048:116–22. [PubMed: 15921661]
51. Borlongan CV, Hess DC. New hope for stroke patients: mobilization of endogenous stem cells. CMAJ. 2006; 174:954–55. [PubMed: 16517763]
52. Nelson PT, Kondziolka D, Wechsler L, et al. Clonal human (hNT) neuron grafts for stroke therapy: neuropathology in a patient 27 months after implantation. Am J Pathol. 2002; 160:1201–06. [PubMed: 11943704]
53*. Kondziolka D, Wechsler L, Goldstein S, et al. Transplantation of cultured human neuronal cells for patients with stroke. Neurology. 2000; 55:565–69. Discusses safety of intracerebral grafts in stroke patients. [PubMed: 10953194]
54. Kondziolka D, Steinberg GK, Wechsler L, et al. Neurotransplantation for patients with subcortical motor stroke: a phase 2 randomized trial. J Neurosurg. 2005; 103:38–45. [PubMed: 16121971]
55. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. Autologous bone marrow mesenchymal stem cell transplantation for chronic stroke. Available at: https://clinicaltrials.gov/ct2/show/NCT01714167?term=stem+cells&recr=Open&no_unk=Y&cond=stroke&rank=1
56. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. Autologous bone marrow stromal cell and endothelial progenitor cell transplantation in ischemic stroke (AMETIS). Available at: https://clinicaltrials.gov/ct2/show/NCT01468064?term=stem+cells&recr=Open&no_unk=Y&cond=stroke&rank=2
57. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. Reparative therapy in acute ischemic stroke with allogenic mesenchymal stem cells from adipose tissue, safety assessment, a randomized, double blind placebo controlled single center pilot clinical

trial (AMASCIS-01). Available at:https://clinicaltrials.gov/ct2/show/NCT01678534?term=stem+cells&recr=Open&no_unk=Y&cond=stroke&rank=5

58. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. Study to assess the safety and effects of autologous adipose-derived stromal cells in patients after stroke. Available at: https://clinicaltrials.gov/ct2/show/NCT01453829?term=stem+cells&recr=Open&no_unk=Y&cond=stroke&rank=10
59. Zhang L, Li Y, Zhang C, et al. Delayed administration of human umbilical tissue-derived cells improved neurological functional recovery in a rodent model of focal ischemia. Stroke. 2011; 42:1437–44. [PubMed: 21493915]
60. Centers for Disease Control and Prevention. Atlanta, GA: Centers for Disease Control and Prevention; 2014. Injury prevention & control: traumatic brain injury. Available at: http://www.cdc.gov/TraumaticBrainInjury/index.html [Last accessed 20 October 2014]
61. Hayashi T, Kaneko Y, Yu S, et al. Quantitative analyses of matrix metalloproteinase activity after traumatic brain injury in adult rats. Brain Res. 2009; 1280:172–77. [PubMed: 19464272]
62. Carroll JE, Borlongan CV. Adult stem cell therapy for acute brain injury in children. CNS Neurol Disord Drug Targets. 2008; 7:361–69. [PubMed: 18991664]
63. Borlongan CV, Lind JG, Dillon-Carter O, et al. Intracerebral xenografts of mouse bone marrow cells in adult rats facilitate restoration of cerebral blood flow and blood- brain barrier. Brain Res. 2004; 1009:26–33. [PubMed: 15120580]
64. Harting MT, Sloan LE, Jimenez F, et al. Subacute neural stem cell therapy for traumatic brain injury. J Surg Res. 2009; 153:188–94. [PubMed: 18694578]
65*. Tajiri N, Acosta SA, Shahaduzzaman Md, et al. Intravenous transplants of human adipose-derived stem cell protect the brain from traumatic brain injury-induced neurodegeneration and motor and cognitive impairments: cell graft biodistribution and soluble factors in young and aged rats. J Neurosci. 2014; 34:313–26. Discusses therapeutic potential of intracerebral grafts in traumatic brain injury. [PubMed: 24381292]
66. Acosta SA, Tajiri N, Shinozuka K, et al. Combination therapy of human umbilical cord blood cells and granulocyte colony stimulating factor reduces histopathological and motor impairments in an experimental model of chronic traumatic brain injury. PLoS One. 2014 published online 12 March 2014. 10.1371/journal.pone.0090953
67. de la Pena I, Sanberg PR, Acosta S, et al. Stem cells and G-CSF for treating neuroinflammation in traumatic brain injury: aging as a comorbidity factor. J Neurosurg Sci. 2014; 58:145–49. [PubMed: 24844175]
68. Tajiri N, Kaneko Y, Shinozuka K, et al. Stem cell recruitment of newly formed host cells via a successful seduction? filling the gap between neurogenic niche and injured brain site. PLoS One. 2013 published online 4 September 2013. 10.1371/journal.pone.0074857
69. Shindo T, Matsumoto Y, Wang Q, et al. Differences in the neuronal stem cells survival, neuronal differentiation and neurological improvement after transplantation of neural stem cells between mild and severe experimental traumatic brain injury. J Med Invest. 2006; 53:42–51. [PubMed: 16537995]
70. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. Safety of autologous stem cell treatment for traumatic brain injury in children. Available at: https://clinicaltrials.gov/ct2/show/record/NCT00254722?term=stem+cells&cond=traumatic+brain+injury&rank=1
71. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. Stem cell therapy in traumatic brain injury. Available at: https://clinicaltrials.gov/ct2/show/record/NCT02028104?term=stem+cells&cond=traumatic+brain+injury&rank=3
72. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. Pediatric autologous bone marrow mononuclear cells for severe traumatic brain injury. Available at: https://clinicaltrials.gov/ct2/show/record/NCT01851083?term=stem+cells&cond=traumatic+brain+injury&rank=6
73. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. Treatment of severe adult traumatic brain injury using bone marrow mononuclear cells. Available

at: https://clinicaltrials.gov/ct2/show/record/NCT01575470?term=stem+cells&cond=traumatic+brain+injury&rank=7

74. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. Effect of recombinant erythropoietin on numbers of circulating progenitor cells in people with persistent symptoms during the subacute period after traumatic brain injury. Available at: https://clinicaltrials.gov/ct2/show/NCT02226848?term=stem+cells&cond=traumatic+brain+injury&rank=2
75. Hefferan MP, Johe K, Hazel T, et al. Optimization of immunosuppressive therapy for spinal grafting of human spinal stem cells in a rat model of ALS. Cell Transplant. 2011; 20:1153–61. [PubMed: 21669047]
76. Glass JD, Boulis NM, Johe K, et al. Lumbar intraspinal injection of neural stem cells in patients with amyotrophic lateral sclerosis: results of a phase 1 trial in 12 patients. Stem Cells. 2012; 30:1144–51. [PubMed: 22415942]
77. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. Clinical trial on the use of autologous bone marrow stem cells in Amyotrophic Lateral Sclerosis (Extension CMN/ELA). Available at: https://clinicaltrials.gov/ct2/show/study/NCT01254539?term=stem+cell&cond=amyotrophic+lateral+sclerosis&rank=1
78. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. Safety and efficacy study of autologous bone marrow derived stem cell treatment in Amyotrophic Lateral Sclerosis. Available at: https://clinicaltrials.gov/ct2/show/record/NCT01363401?term=stem+cell&cond=amyotrophic+lateral+sclerosis&rank=12
79. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. Dose escalation and safety study of human spinal cord derived neural stem cell transplantation for the treatment of amyotrophic lateral sclerosis. Available at: https://clinicaltrials.gov/ct2/show/study/NCT01730716?term=stem+cell&cond=amyotrophic+lateral+sclerosis&rank=3
80. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. Biological treatment of Amyotrophic Lateral Sclerosis (NeuStem-ALS). Available at: https://clinicaltrials.gov/ct2/show/record/NCT02193893?term=stem+cell&cond=amyotrophic+lateral+sclerosis&rank=10
81. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. Stem cell therapy for Amyotrophic Lateral Sclerosis. Available at: https://clinicaltrials.gov/ct2/show/record/NCT01984814?term=stem+cell&cond=amyotrophic+lateral+sclerosis&rank=16
82. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. The clinical trial on the use of umbilical cord mesenchymal stem cells in Amyotrophic Lateral Sclerosis. Available at: https://clinicaltrials.gov/ct2/show/study/NCT01494480?term=stem+cell&cond=amyotrophic+lateral+sclerosis&rank=2
83. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2014. Human spinal cord derived neural stem cell transplantation for the treatment of Amyotrophic Lateral Sclerosis (ALS). Available at: https://clinicaltrials.gov/ct2/show/study/NCT01348451?term=stem+cell&cond=amyotrophic+lateral+sclerosis&rank=5
84. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2014. Effect of intrathecal administration of hematopoietic stem cells in patients with Amyotrphic lateral sclerosis (ALS). Available at: https://clinicaltrials.gov/ct2/show/study/NCT01933321?term=stem+cell&cond=amyotrophic+lateral+sclerosis&rank=4
85. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2014. Mesenchymal stem cell injection in Amyotrophic Lateral Sclerosis. Available at: https://clinicaltrials.gov/ct2/show/study/NCT02116634?term=stem+cell&cond=amyotrophic+lateral+sclerosis&rank=6
86. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2014. A dose-escalation safety trial for intrathecal autologous mesenchymal stem cell therapy in Amyotrophic Lateral Sclerosis. Available at: https://clinicaltrials.gov/ct2/show/study/NCT01609283?term=stem+cell&cond=amyotrophic+lateral+sclerosis&rank=11
87. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2014. Mesenchymal stem cells for treatment of Amyotrophic Lateral Sclerosis (ALS). Available at:

https://clinicaltrials.gov/ct2/show/record/NCT01142856?term=stem+cell&cond=amyotrophic+lateral+sclerosis&rank=13

88. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2012. Human neural stem cell transplantation in Amyotrophic Lateral Sclerosis (ALS) (hNSCALS). Available at: https://clinicaltrials.gov/ct2/show/study/NCT01640067?term=stem+cell&cond=amyotrophic+lateral+sclerosis&rank=7
89. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2014. Safety study of HLA-haplo matched allogenic bone marrow derived stem cell treatment in Amyotrophic Lateral Sclerosis. Available at: https://clinicaltrials.gov/ct2/show/study/NCT01758510?term=stem+cell&cond=amyotrophic+lateral+sclerosis&rank=8
90. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2014. Derivation of induced pluripotent stem cells from an existing collection of human somatic cells. Available at: https://clinicaltrials.gov/ct2/show/NCT00801333?term=stem+cells&cond=amyotrophic+lateral+sclerosis&rank=20
91. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. The effect of GCSF in the treatment of ALS patients. Available at: https://clinicaltrials.gov/ct2/show/NCT01825551?term=stem+cells&cond=amyotrophic+lateral+sclerosis&rank=25
92. Karussis D, Grigoriadis S, Polyzoidou E, et al. Neuroprotection in multiple sclerosis. Clin Neurol Neurosurg. 2006; 108:250–4. [PubMed: 16413962]
93. Wang X, Kimbrel EA, Ijichi K, et al. Human ESC-derived MSCs outperform bone marrow MSCs in the treatment of an EAE model of multiple sclerosis. Stem Cell Reports. 2014; 3:115–30. [PubMed: 25068126]
94. Donders, R.; Vanheusden, M.; Bogie, JF., et al. Human Wharton's jelly-derived stem cells display immunomodulatory properties and transiently improve rat experimental autoimmune encephalomyelitis. Cell Transplant. 2014. published online 10 October 2014http://dx.doi.org/10.3727/096368914X685104
95. Li, JF.; Zhang, DJ.; Geng, T., et al. The potential of human umbilical cord-derived mesenchymal stem cells as novel cellular therapy for multiple sclerosis. Cell Transplant. 2014. published online 5 November 2014http://dx.doi.org/10.3727/096368914X685005
96. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2013. Safety and efficacy of autologous bone marrow mesenchymal stem cells in multiple sclerosis. Available at: http://clinicaltrials.gov/ct2/show/NCT01895439?term=stem+cells&cond=multiple+sclerosis&rank=1
97. US National Library of Medicine. Bethesda, MD: U.S. National Library of Medicine; 2014. Feasibility study of human umbilical cord tissue-derived mesenchymal stem cells in patients with multiple sclerosis. Available at: http://clinicaltrials.gov/ct2/show/NCT02034188?term=stem+cells&cond=multiple+sclerosis&rank=5
98. Lee PH, Lee JE, Kim HS, et al. A randomized trial of mesenchymal stem cells in multiple system atrophy. Ann Neurol. 2012; 72:32–40. [PubMed: 22829267]
99. Herberts CA, Kwa MS, Hermsen HP. Risk factors in the development of stem cell therapy. J Transl Med. 2011; 9:29. [PubMed: 21418664]
100. Emborg ME, Zhang Z, Joers V, et al. Intracerebral transplantation of differentiated human embryonic stem cells to hemiparkinsonian monkeys. Cell Transplant. 2013; 22:831–8. [PubMed: 23594934]
101. Leveque X, Mathieux E, Nerriere-Daguin V, et al. Local control of the host immune response performed with mesenchymal stem cells: perspectives for functional intracerebral xenotransplantation. J Cell Mol Med. 2014 published online 14 October 2014. 10.1111/jcmm.12414
102. Sohni A, Verfaillie CM. Mesenchymal stem cells migration homing and tracking. Stem Cells Int. 2013 published online 30 September 2013. 10.1155/2013/130763
103. de la Pena I, Pabon M, Acosta S, et al. Oligodendrocytes engineered with migratory proteins as effective graft source for cell transplantation in multiple sclerosis. Cell Med. 2014; 6:123–27. [PubMed: 24999443]

104. Yasuhara T, Hara K, Maki M, et al. Intravenous grafts recapitulate the neurorestoration afforded by intracerebrally delivered multipotent adult progenitor cells in neonatal hypoxic-ischemic rats. J Cereb Blood Flow Metab. 2008; 28:1804–10. [PubMed: 18594556]

105. Edelstein SA, Holmes LP. Seeking novel treatments abroad. Stem cell tourism: risks, legal issues and mitigation-proceeding of the STEMSO conference CellR^4. 2014; 2:e700.

106. California Institute for Regenerative Medicine. San Francisco, CA: California Institute for Regenerative Medicine; 2013. Patient advisory for stem cell therapy and medical tourism. Available at: https://www.cirm.ca.gov/about-cirm/newsroom/press-releases/08192013/patient-advisory-stem-cell-therapy-and-medical-tourism

107. Board on Health Sciences Policy, Board on Life Sciences, Divisions on Earth and Life Studies, et al. Stem Cell Therapies: Opportunities For Ensuring The Quality And Safety Of Clinical Offerings: Summary Of A Joint Workshop. Washington, DC: National Academies Press; 2014.

Key Issues

- Optimization of safety and efficacy remains a key gating item in translating stem cell therapy to the clinic for treatment of neurological disorders
- The route of administration of stem cells for a specific disease can vary, therefore, it is essential to determine which is most beneficial for the patient
- Intracerebral route of administration stands as an efficacious way to deliver cells directly into specific brain areas
- Understanding the disease pathology of brain disorders is critical to identifying the appropriate route of stem cell delivery
- Characterization of the donor stem cells may provide insights into their phenotypic fate (i.e., migration potential) after transplantation and can factor in deciding cell delivery route
- In addition to optimizing the transplant regimen (i.e., cell delivery route), challenges that warrant preclinical investigations include high tissue variability, lack of scalability, ethical concerns, inability to obtain an epidemiologically meaningful quantity of tissue, graft-induced adverse effects, and the establishment of acceptable levels of efficacy and toxicity