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Neurogenesis After Traumatic Brain Injury

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Despite improving rates of survival after traumatic brain injury (TBI), many head-injured patients incur permanent neurologic impairment. Each year in the United States, approximately 80,000 individuals sustain TBI that results in significant long-term disability [1]. In addition to local neuronal destruction resulting from the primary insult, mechanical brain injury secondarily induces a progressive cascade of related events that contribute to neuronal death, including ischemia, brain edema, diffuse axonal injury, excitotoxicity, radical-mediated damage, mitochondrial dysfunction, and dysregulation of calcium homeostasis [2,3]. Despite an improved understanding of the pathophysiology that occurs in TBI, clinical neuroprotection trials pharmacologically targeting these secondary mechanisms have failed to show consistent improvement in outcome for head-injured patients [4]. With the confirmation of continual neurogenesis in the adult human hippocampus [5] and subventricular zone (SVZ) [6,7], experimental paradigms have expanded to evaluate the response of endogenous neuronal progenitor cells (NPCs) to traumatic injury. Recent studies have begun to assess the potential of these cells to generate new neurons capable of functionally counterbalancing neuronal loss in TBI. Although this is a lofty goal with numerous pitfalls, accumulating data suggest that neurogenesis increases in response to mechanical brain injury in multiple areas of the adult mammalian brain. This review aims to summarize these findings and to evaluate current progress toward potential neurogenesis-targeted clinical therapy.

Neuronal loss in traumatic brain injury

Neuronal loss after TBI is focal and diffuse. Focal damage typically involves hemorrhagic lesions within the gray matter or at gray-white junctions. These contusions are typically observed at the frontal poles, orbital frontal lobes, temporal poles, and cortex above the Sylvian fissure [8]. Within contusions and adjacent neocortex of TBI patients, focal neuronal death occurs by necrotic and apoptotic mechanisms [9,10]. Among diffuse injury sites, the hippocampus is known to be damaged frequently in human beings, with neuronal loss occurring in greater than 80% of fatal TBI, even in the absence of elevated intracranial pressure (ICP) [11,12]. Additionally, apoptotic neurons have been observed in the human hippocampus up to 12 months after injury [13]. In fact, in the period after the acute phase of focal neuronal injury, hippocampal neurons may be the most vulnerable neurons in the brain because they show the earliest evidence of TBI-induced degeneration in experimental models [14]. In the two most relevant and frequently used experimental models of TBI, the lateral fluid percussion (LFP) and controlled cortical impact (CCI) injury

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models, selective neuronal death has been well described in the hippocampus (and to a lesser extent in the thalamus) [15].

More recently, these experimental TBI models have been used to investigate TBI-induced neurogenesis, with reports generally centered around two themes. The first is that although selectively vulnerable to TBI, the neurogenic hippocampus may have the unique ability to replace damaged neurons locally. Second, there is evidence that an injury stimulus may induce NPCs from the SVZ to migrate to areas of focal cortical damage. Additionally, there is some evidence for local activation of latent NPCs in the injured neocortex itself. Each of these themes is discussed separately in the following sections.

Posttraumatic hippocampal neurogenesis

In human beings, constitutive endogenous neurogenesis was demonstrated in the hippocampus after examination of postmortem tissue obtained from cancer patients who had received an intravenous infusion of bromodeoxyuridine (BrdU) for diagnostic purposes [5]. Studies from rodents demonstrate that astrocytic cells residing in the adult subgranular zone (SGZ) of the dentate gyrus continually generate neurons that migrate a short distance into the granule cell layer [16]. These new neurons demonstrate functional integration within the hippocampal circuitry by sending dendrites into the molecular layer and axons into the CA3 region [17,18] and exhibiting mature electrophysiologic properties [19].

Injury to the hippocampus has been associated with learning and memory deficits, which are the hallmarks of TBI. Neurogenesis in this region has been implicated in learning and memory functions [20], raising hope that injury-induced neurogenesis may function to replace damaged neurons, contribute to neuronal circuit repair, and restore some appreciable neurologic function in TBI patients. To explore this potential, our group and others have begun to study the effects of experimental TBI on hippocampal neurogenesis in rodents (Table 1). After LFP and CCI injury, cell proliferation increases in the dentate gyrus. We have previously reported that proliferation increases three- to fourfold beginning as early as 2 days postinjury (dpi) (Fig. 1A, B) [21,22]. Similarly, other groups have detected up to a sixfold increase in dividing cells in the ipsilateral dentate

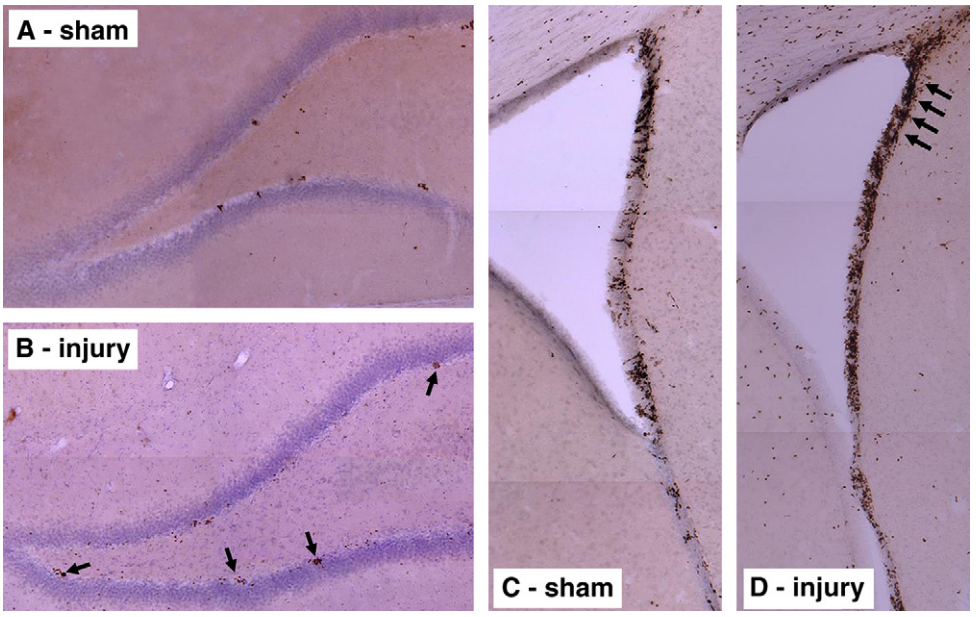

Fig. 1. Increased cell proliferation in neurogenic regions after experimental TBI. An increase in the number of bromodeoxyuridine (BrdU)-labeled cells (*brown*), observed in the ipsilateral dentate gyrus of sham animals (*A*), is apparent in LFP-injured rats at 2 days after injury (*B*). These cells are mainly clustered in the subgranular zone, as would be expected at this time point (*B, arrows*). Similarly, BrdU labeling in the ipsilateral subventricular zone of sham animals (*C*) significantly increases after injury (*D*, *arrows*).

gyrus and a significant but lesser increase in dividing cells in the contralateral dentate gyrus [21,23–28]. These data indicate the presence of a proliferative response that peaks during the first week after injury, and we have observed a return to baseline levels of proliferation in the dentate gyrus by 35 dpi [29].

Most proliferating cells in the dentate gyrus become neurons under control conditions, and increased SGZ proliferation after TBI also seems to predict an increased addition of new neurons to functional hippocampal circuitry. Several studies have reported a significant increase in the number of new cells that express neuronal markers after appropriate maturation periods, suggesting that injury-induced proliferation is primarily neurogenic in the dentate gyrus rather than a reactive gliosis. Our group has reported an approximately twofold increase in the presence of BrdU-labeled cells that express NeuN, a nuclear protein expressed by postmitotic neurons, up to 35 dpi [22,29]. Others have reported a fivefold increase in the number of injury-generated neurons, present through 60 dpi [25]. Based on studies verifying the physiologic function of morphologically mature-appearing adult-generated neurons in noninjured animals, it is reasonable to hypothesize that similar cells in injured animals achieve at least a minimum level of functional activity. In agreement with studies in uninjured rats demonstrating axonal extension into the CA3 regions of the hippocampus by newly generated granule neurons between 4 and 10 days after birth [17], retrograde labeling has been used to verify the conservation of this process in cells generated after TBI (D. Sun and colleagues, manuscript in preparation) [24]. These data confirm that experimental TBI produces a surge in the addition of stable neurons to the hippocampal circuitry after injury. Efforts to increase the magnitude of this event and to assess for subsequent neurologic improvement are discussed elsewhere in this article.

It has long been accepted that juvenile mammals recover to a greater extent than adults after TBI [30]. Because cell proliferation in the dentate gyrus decreases with age in normal rats [31], we previously hypothesized that age-related differences in hippocampal neurogenesis after TBI may be responsible, in part, for greater cognitive recovery in juveniles [22]. We found that in the first 2 days after LFP injury, juvenile rats generated nearly twice the number of new cells in the SGZ compared with adults, although this response seemed to be related to a greater level of basal progenitor cell proliferation, because the percentage increase greater than baseline was similar in young and old animals. Additionally, however, the percentage of neuronal differentiation in juvenile hippocampi was nearly double that in adult hippocampi, as measured from 7 dpi throughout the 4-week study. These findings, together with observations that new granule cell neurons functionally integrate by 14 days after birth [17] and become critically involved in the learning response within this same period [32], suggest that Morris Water Maze (MWM) data demonstrating fewer cognitive deficits and a higher index of recovery during the first 2 weeks after injury in juvenile rats compared with adults [33] may result, in part, from a greater ability of younger animals to generate new hippocampal granule cell neurons in response to injury. Similar age-related differences in hippocampal neurogenesis also were recently reported after middle cerebral artery occlusion [34].

Neither the mechanism by which injury stimulates hippocampal neurogenesis nor that by which neuronal differentiation is enhanced in younger animals is entirely clear. It may be that injury solely increases the proliferative rate of NPCs, whereas the additional increased survival of newly generated neurons occurs only in brains young enough to also generate an additional neurotrophic injury response. A related example can be found in evidence that an enriched environment and voluntary exercise increase neurogenesis by two distinctly different methods. Although exercise increases NPC proliferation, resulting in more cells that can become neurons, an enriched environment produces an increase in neurogenesis without altering cell proliferation [35]. Despite a constant decline in NPC proliferation in the dentate gyrus from adolescence through senescence [31,36,37], the adult brain upregulates proliferation to almost the same extent as the juvenile brain after TBI [22], indicating that NPCs conserve the ability to respond to proliferative signals. The greater percentage of new cells surviving as neurons in juveniles suggests a greater level of neurotrophic support that is not present or not induced in adults. Although neurotrophins are upregulated after TBI [38], the aging hippocampus has been reported to contain less brain-derived neurotrophic factor (BDNF) in comparison to the hippocampus of younger injured animals after kainic acid injury [39], suggesting that the neurotrophic response to injury in the hippocampus is age dependent. Deciphering

Table 1
Studies reporting increased neurogenesis in the hippocampus after experimental traumatic brain injury

Reference	Injury model	Region studied	Exogenous augmentation	Increase in proliferating cells (time point)		Proliferation marker	Migration	Neuronal properties (time point)	Cognitive performance
				Ipsilateral to injury	Contralateral to injury				
Chirumamilla et al, 2002	LFP	DG	None	×10 (3 dpi)	Not reported	BrdU	Not assessed	None found	Not assessed
Dash et al, 2001	CCI	DG	None	×5.9 (9 dpi); baseline (21 dpi)	×4.4 (9 dpi); baseline (21 dpi)	BrdU	Within GCL	Calbindin/BrdU (30 dpi)	Not assessed
Emery et al, 2005	LFP	DG	None	×5 (3 dpi); ×2 (14 dpi)	×5 (3 dpi); ×2 (14 dpi)	BrdU	Within GCL	Retrograde labeling from CA3 (14 dpi)	Not assessed
Kernie et al, 2001	CCI	DG	None	×5 (60 dpi)	×2 (60 dpi)	BrdU	Within GCL	Calbindin/BrdU (60 dpi)	Not assessed
Kleindienst et al, 2005	LFP	DG	Injured control	×1.9 (5 dpi); baseline (35 dpi)	Not reported	BrdU	Within GCL	NeuN/BrdU colocalization (35 dpi)	Improved water maze performance (35 dpi)
			Intraventricular S 100B	×3.4 (5 dpi); ×1.7 (35 dpi)					
Lu et al, 2003	CCI	DG	Injured control	×3 (14 dpi)	×2 (14 dpi)	BrdU	Within GCL	Hu/BrdU (42 dpi)	Reduced asymmetry deficit by mNSS (42 dpi)
			DEtA/ NONOate	×6 (14 dpi)	×4 (14 dpi)				
Lu et al, 2005	CCI	DG	Erythropoietin	×4 compared with injured control	×3 compared with injured control	BrdU	Not assessed	BrdU/NeuN (15 dpi): ×2–3 ipsilateral increase	Improved water maze performance (15 dpi)

Rice et al, 2003	LFP	SGZ	None	×3 (2 dpi) ×1.5 (8 dpi)	×2.5 (2 dpi) No change (8 dpi)	BrdU	Within GCL	β-tubulin/ BrdU (15 dpi)	Not assessed
Sun et al, 2005	LFP	SGZ, juvenile	None	> ×4 (2 dpi)	Not assessed	BrdU	Within GCL	BrdU/NeuN (28 dpi): ×1.2 sham, ×2 adult	Not assessed
		SGZ, adult	None	×3 (2 dpi)	Not assessed		Within GCL	BrdU/NeuN (28 dpi)	Not assessed
		Hilus, juvenile	None	×3 (2 dpi)	Not assessed		Within GCL		Not assessed
		Hilus, adult	None	×10 (2 dpi)	Not assessed		Within GCL		Not assessed
Yoshimura et al, 2003	CCI	DG	None	×4.3 (9 dpi)	Not assessed	BrdU	Within GCL	BrdU/NeuN (35 dpi): ×3.2 sham	Not assessed
		DG	FGF-2 overexpression	×10.3 (9 dpi)	Not assessed		Within GCL	BrdU/NeuN (35 dpi): ×6 sham, ×2.5 vector control	Not assessed

Abbreviations: BrdU, bromodeoxyuridine; CCI, controlled cortical impact; dpi, days postinjury; DG, dentate gyrus; FGF-2, fibroblast growth factor-2; GCL, granule cell layer; LFP, lateral fluid percussion; mNSS, modified Neurological Severity Score; SGZ, subgranular zone.

differences in extracellular signaling pathways and local microenvironmental cues between young and aged brains should be helpful in devising strategies to increase NPC proliferation and neuroblast survival further after TBI.

Experimental TBI induces neuronal loss and cognitive deficits despite the previously discussed evidence for increased addition of new neurons in the hippocampus, raising the question of whether neurogenesis contributes at all to functional recovery. We have found that elimination of cell proliferation in the dentate gyrus by irradiation reduces cognitive recovery, as measured by the MWM test in adult rats after TBI (D. Sun and colleagues, manuscript in preparation), suggesting that neurogenesis does contribute to functional recovery. If so, does augmentation of neurogenesis after injury result in measurable improvements in cognitive performance? This hypothesis has been tested by our group by means of intraventricular administration of S100B, a neurotrophic calcium-binding protein secreted by astrocytes [29]. After TBI and S100B treatment, the percentage of newly generated cells coexpressing NeuN in dentate gyrus was increased twofold compared with vehicle infusion at 35 dpi. These same animals underwent MWM testing on days 30 to 34 after injury. When individual average MWM performance was compared with the percentage of BrdU-NeuN–colabeled cells, improved cognitive recovery correlated strongly with the number of newly generated neurons. Note, however, that S100B has other neurophysiologic properties, such as modulation of long-term potentiation [40]. Lu and coworkers [26,27] have evaluated the cognitive recovery of animals receiving other neurotrophic factors after CCI injury. Erythropoietin administration for 14 dpi significantly increased the percentage of newly generated cells that differentiated into mature neurons in the granular cell layers of the contralateral and ipsilateral dentate gyrus [26]. Treated animals also performed significantly better than nontreated injured animals in MWM testing, although erythropoietin most likely acts as a neuroprotectant at multiple levels after injury [41]. In a separate CCI injury study, these same investigators found that treatment with intraperitoneal DEtA/NONOate, a nitric oxide donor, also increased the amount of dividing cell that survived as neurons after injury [27]. Improved behavioral outcome at 42 dpi was demonstrated by a significant reduction in asymmetry deficits compared with nontreated injured controls, as measured by the Modified Neurological Severity score. Note, again, that nitric oxide also functions in regulating cerebral blood flow and neuronal activity [42], which may affect cognition independent of any role in neurogenesis. The total number of BrdU-labeled cells in the SGZ declined to baseline levels by 42 dpi (28 days after the last BrdU injection) in animals not treated with the nitric oxide donor, whereas levels remained significantly elevated in treated animals. Cells colabeled with BrdU and Hu, an early neuronal marker, increased by twofold in the ipsilateral dentate gyrus and by a greater amount in the contralateral dentate gyrus compared with injured controls, although Hu had previously been reported as undetectable in dentate granule cells [43]. Also noted were newly born neurons in the CA regions, areas to which SGZ NPCs do not migrate, raising the possibility that some newborn neurons in the hippocampus may have been derived from the nearby SVZ. Supporting this possibility is evidence for the replacement of CA1 pyramidal neurons in response to transient forebrain ischemia by precursors migrating from the nearby posterior periventricular region [44]. Indeed, data from middle cerebral artery occlusion models indicating that SVZ neuroblasts are directed to sites of focal ischemic injury [45,46] support data indicating that similar mechanisms are initiated after TBI.

Posttraumatic subventricular zone neurogenesis

The SVZ is the largest germinal region of the adult mammalian brain, although in comparison to other species, relatively little is known regarding the contribution of human SVZ progenitor cells to adult neurogenesis. Sanai and colleagues [7] recently described a ribbon of SVZ astrocytes lining the lateral ventricles of the adult human brain that proliferate in vivo and behave as multipotent progenitor cells in vitro. These investigators subsequently undertook a more detailed analysis of the cytoarchitecture and cellular organization of this region [6]. In contrast to the SVZ of rodents and primates, which gives rise to neuroblasts that form migratory chains and travel to the olfactory bulb, where they differentiate to granule cell neurons, there was no evidence for similar neuroblast organization in the adult human brain. Nonetheless, it is reasonable to speculate that the potential for induction of migration exists, based on animal studies demonstrating that various forms of brain injury activate molecular

recruitment pathways to direct neuroblasts from the SVZ to sites of pathologic change.

Similar to models of stroke, epilepsy, and chemical demyelination or deafferentation, in which redirected migration of NPCs from the SVZ toward sites of injury has been observed in multiple studies [47], experimental TBI induces an increase in SVZ proliferation in some studies (Fig. 1C, D), followed by recruitment of NPCs to the injured cortex (Table 2). Data from our LFP injury studies demonstrate a 1.3-fold to twofold increase in ipsilateral SVZ cell proliferation in the first 5 dpi [21,29,48], whereas others have reported a twofold to fourfold increase after CCI injury, up to 14 dpi [27,49]. Contralateral SVZ proliferation also increases (but to a lesser extent) after injury in both models [27,48]. An increased rate of NPC proliferation produces a larger pool of neuroblasts available for subsequent migration, and it seems that injury-related physiologic changes recruit some of these new cells to travel outside their normal migratory pathway.

Cells labeled with BrdU and neuronal markers have been observed in the cortex, striatum, and corpus callosum after injury. Using doublecortin, a microtubule-associated protein expressed in migrating and differentiating neurons [50–52], cells colabeled with BrdU have been observed migrating into the cortex toward the site of injury as early as 3 dpi [49], whereas BrdU cells labeled with Hu have been observed in the striatum and cortex at 42 dpi [27]. In addition to increasing hippocampal neurogenesis as described previously, S100B [29] and DEtA/NONOate [27] also increase proliferation in the SVZ. DEtA/NONOate treatment resulted in a threefold increase in SVZ proliferation at 14 dpi, followed by a twofold increase in the presence of Hu/BrdU cells in the corpus callosum, striatum, and cortex at 42 dpi, suggesting that augmentation of SVZ proliferation produces a larger surge of neuroblasts that migrate to the site of injury. Migration from the SVZ after TBI has been demonstrated in other studies by labeling SVZ progenitors before injury. Intraventricular injection of fluorescent microspheres was recently used to label BrdU-positive subventricular cells, whose migration could be tracked from the SVZ throughout the corpus callosum and from the SVZ to the cortex surrounding a cortical impact contusion site [49]. In the SVZ, corpus callosum, striatum, and cortex, many of these cells were also positive for doublecortin at 3 dpi, suggesting that neuroblasts can be redirected from the SVZ by injury-specific extracellular signaling pathways. In the pericontusional cortex, expression of the mature neuronal marker NeuN by cells that presumptively migrated from the SVZ after injury has also been observed at 56 dpi after labeling NPCs by means of intraventricular injection of a lipophilic dye before injury [53].

Whether neuroblasts that migrate from the SVZ can replace cortical neurons lost in TBI remains to be determined. In the olfactory bulb of rodents, 90% of neuroblasts differentiate into GABAergic interneurons, whereas some of the remainder become dopaminergic periglomerular interneurons. Although the mechanism of this phenotypic determination is not understood, this fact does demonstrate that SVZ NPCs are not fated to a sole neuronal phenotype. We previously provided further evidence for the plasticity of SVZ NPCs by demonstrating their differentiation to calbindin-expressing neurons after transplantation to the dentate gyrus [54]. The TBI studies described here add to the greater amount of data generated from stroke models, in which the ischemic injury is related somewhat to that seen in TBI. Although NPCs do not seem to differentiate into neurons and persist in the peri-infarct neocortex after stroke, neuroblasts that migrate to the injured striatum do express markers of medium spiny neostriatal projection neurons, suggesting that they have the potential for appropriate phenotypic replacement after injury [45,46]. Taken together, these data suggest the potential for directing the phenotypic differentiation of NPCs that migrate to areas of injury once the molecular pathways governing these phenomena are better understood.

The mechanism by which recruitment of neuroblasts to injury sites occurs in the adult brain is not yet known. It has been suggested, with relation to stroke, that injury may break the physical barrier between astrocytes and migrating neuroblasts that normally prevents migration away from the SVZ [55], and this may also hold true for TBI. The various localized and overlapping pathways of growth factors, extracellular proteins, metalloproteases, neurotransmitters, hormones, and angiogenesis normally active in neurogenic niches [56] have all been hypothesized as potentially responsible for altering neurogenesis after injury [55,57]. Most recently, neuroblast migration in the rodent brain was shown to parallel cerebrospinal fluid (CSF) flow, which seems to be required for neuroblast orientation and the formation of chemorepulsive gradients in the SVZ [58]. The possible role of CSF in creating gradients in the underlying brain parenchyma in human beings is especially

Table 2
Studies reporting increased neurogenesis in the subventricular zone and cortex after experimental traumatic brain injury

Reference	Injury model	Region studied	Exogenous augmentation	Increase in proliferating cells (time point)		Proliferation marker	Migration	Neuronal properties (time point)
				Ispilateral to injury	Contralateral to injury			
Chen et al, 2003	LFP	SVZ	None	No change (14 dpi) ×3.4 (365 dpi)	No change (14 dpi) ×3.1 (365 dpi)	PCNA	Not assessed	NF-M/PCNA (365 dpi): ×2.5 ipsilateral increase
Chirumamilla et al, 2002	LFP	SVZ	None	×1.3 (3 dpi)	Not reported	BrdU	Not assessed	None found
Kernie et al, 2001	CCI	Cortex, peri, contusion	None	Qualitative increase	Not assessed	BrdU	Not assessed	Nestin/BrdU (7 dpi) GFAP/BrdU only (60 dpi)
Kleindienst et al, 2005	LFP	SVZ	Injured control Intraventricular S100B	×1.6 (5 dpi) ×1.8 (5 dpi)	Not reported	BrdU	Not assessed	Not assessed
Lu et al, 2003	CCI	SVZ	Injured control DEtA/NONOate	×3.8 (14 dpi) ×6.5 (14 dpi)	×3 (14 dpi) ×4.4 (14 dpi)	BrdU	Not assessed	HU-negative HU-negative
		Striatum	Injured control	BrdU cells identified after injury	BrdU cells identified after injury		Origin not determined	Hu/BrdU cells apparent
			DEtA/NONOate	×3 compared with injured control (14 dpi)	×3 compared with injured control (14 dpi)			Hu/BrdU (×2 control at 42 dpi)
		Cortex	Injured control	BrdU cells identified after injury	BrdU cells identified after injury		Origin not determined	Hu/BrdU cells apparent
			DEtA/NONOate	×3 compared with injured control (14 dpi)	×3 compared with injured control (14 dpi)			Hu/BrdU (×2 control at 42 dpi)

Ramaswamy et al, 2005	CCI	SVZ	None	×2 (3 dpi)	Not assessed	BrdU	cc, pericontusion, striatum, subcortical	Doublecortin/ BrdU (3 dpi)
		Striatum	None	BrdU cells identified after injury	Not assessed		From SVZ	None found (3 dpi)
		Cortex, peri, contusional	None	BrdU cells identified after injury	Not assessed		From SVZ	Doublecortin/ BrdU (3 dpi)
		Subcortical structures	None	BrdU cells identified after injury	Not assessed		From SVZ	None found (3 dpi)
Rice et al, 2003	LFP	SVZ	None	×2 (2 dpi) ×1.5 (8 dpi)	×1.3 (2 dpi) No change (8 dpi)	BrdU	Not assessed	β-tubulin/nestin (10 dpi): ×4 increase after in vitro isolation
Salman et al, 2004	CCI	SVZ	None	Not assessed	Not assessed	None	Corte, peri, contusion	Rare NeuN/DIO cells (56 dpi)

Abbreviations: BrdU, bromodeoxyuridine; cc, corpus callosum; CCI, controlled cortical impact; dpi, days postinjury; GFAP, glial fibrillary acidic protein; LFP, lateral fluid percussion; NF-M, medium chain neurofilament; PCNA, proliferating cell nuclear antigen; SVZ, subventricular zone.

intriguing, considering the extent to which this flow may be altered after TBI, secondary to increased ICP and possibly hydrocephalus, not to mention the therapeutic diversion of CSF.

Posttraumatic cortical neurogenesis

Although the hippocampus is selectively damaged in TBI, a great deal of neuronal loss obviously occurs in focal parenchymal contusions arising in locations that vary with the specific primary or secondary injury. As discussed previously, injury signals may induce migration of neuroblasts from the SVZ, but there is also evidence for the activation of latent NPCs at sites of cortical injury in the mammalian brain. The studies of Macklis and his colleagues (eg, Magavi and coworkers [59]) in adult mice have suggested the possibility that after targeted neuronal death, endogenous neural precursors from the cortex itself can differentiate into corticothalamic neurons, which survive for many months and form the appropriate long-distance corticothalamic connections. In human beings, evidence for the existence of NPCs in nongerminal cortex has thus far been restricted to the isolation of cells from surgically resected tissue, which proliferate in vitro and acquire immunochemical neuronal phenotypes in differentiating culture conditions. We have reported the isolation of putative neuroblasts from neocortical tissue surgically resected from TBI patients [60], whereas others have previously reported the isolation of neurosphere-generating cells from similar cortical tissue [61]. After CCI injury in rodents, the formation of neurospheres from neocortical cells has also been observed but only when injured tissue was isolated at 3 dpi, corresponding to a peak in nestin expression in pericontusional cortex [62]. Induction of nestin expression in the cortex adjacent to the CCI injury has also been reported at 7 dpi, supporting the possibility that resident NPCs are activated in the cortex after trauma [25]. Further studies are needed to determine whether there are cells residing in the cortex that harbor the ability to generate neuroblasts on activation by injury.

Potential for therapeutic intervention

Assuming that the adult human brain is capable of generating new neurons in response to injury, as has been observed in rodents, there are still significant blockades to actual functional neuronal regeneration, with two of the foremost being glial scar formation and inflammation. To overcome the inhibitory environment of the glial scar, combinatorial treatments to provide a growth-related pathway across lesion cavities while enhancing the ability of neurons to elongate by manipulating growth inhibitors in the glial scar environment may allow long-distance functional regeneration after TBI, as proposed in an excellent recent review [63]. Experimental neuroinflammation has been shown to inhibit hippocampal neurogenesis, an effect that can be reversed with the administration of minocycline [64], which inhibits microglial activation, or with indomethacin [65], a common nonsteroidal anti-inflammatory drug. Additionally, indomethacin seems to enhance neurogenesis after experimental stroke [66]. Indomethacin has been used clinically in the treatment of TBI to reduce ICP and increase cerebral perfusion pressure [67], and as with other neuroprotective strategies, one would like to know whether there was any effect on neurogenesis in these patients. Collection of data from patients regarding in vivo neurogenesis is, of course, quite difficult; however, postmortem analysis of proliferation in the neurogenic regions of injured brains would be especially useful in the absence of such data to date. Further into the future, it is not out of the question that efforts to development nanoimaging [68] may eventually allow for in vivo visualization of NPC proliferation and migration on the cellular level.

Of course, the introduction of new neurons to damaged circuitry might be for naught in the absence of evidence that new cells can improve patient recovery from injury. Evidence thus far supporting this idea comes predominantly from clinical cell transplantation studies demonstrating partial recovery of neurologic function in patients with basal ganglia stroke, after transplantation of human neuronal cells [69–71]. Remarkably, the autologous transplantation of putative NPCs harvested from and reintroduced into patients with open brain trauma was recently reported [72,73]. These investigators describe the harvest of NPCs from neocortical regions and subsequent improvement in neurologic function after NPC reintroduction, as measured by imaging and clinical outcome scores, although there is clearly much work to be done in verifying these results.

Summary

The relatively preliminary studies described here have begun to delineate aspects of the

neurogenic response to TBI that should determine whether this phenomenon can be manipulated for therapeutic purposes. Clarifying the time course over which NPC proliferation, migration, differentiation, and integration occur after injury is necessary for defining therapeutic windows in which to attempt to augment these processes. From studies thus far, it is known that there are an increased number of neurons added to the rodent hippocampal circuitry, which survive for at least several weeks after injury. Without longer outcome data, it is still not certain that this event is not transitory, and, of course, its verification in the human brain is still pending. Likewise, it seems that neuroblasts from the rodent SVZ migrate toward damaged cortical areas; however, the verification of this phenomenon in the human brain would be even more remarkable, because, thus far, there is no evidence for baseline long-distance SVZ NPC migration in the human brain (along the lines of the rodent rostral migratory stream). It is likely that human NPCs do proliferate in response to TBI, but measuring this occurrence must be the first step in assessing the applicability of findings in rodent studies to the potential development of neurogenic therapies to be tested in clinical trials. Additionally, it remains challenging to attribute any improvement in neurologic function to the augmentation of neurogenesis by therapeutic agents, because many neurogenic compounds have complex effects on central nervous system physiology. Nonetheless, neurogenesis remains an exciting prospect for exploration in TBI research.

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