CHAPTER 9

Progressive damage after brain and spinal cord injury: pathomechanisms and treatment strategies

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Abstract: The pathophysiology of brain and spinal cord injury (SCI) is complex and involves multiple injury mechanisms that are spatially and temporally specific. It is now appreciated that many of these injury mechanisms remain active days to weeks after a primary insult. Long-term survival studies in clinically relevant experimental studies have documented the structural changes that continue at the level of the insult as well as in remote brain structures. After traumatic brain injury (TBI), progressive atrophy of both gray and white matter structures continues up to 1 year post-trauma. Progressive changes may therefore underlie some of the long-term functional deficits observed in this patient population. After SCI, similar features of progressive injury are observed including delayed cell death of neurons and oligodendrocytes, axonal demyelination of intact fiber tracts and retrograde tract degeneration. SCI also leads to supraspinal changes in cell survival and remote brain circuitry. The progressive changes in multiple structures after brain and SCI are important because of their potential consequences on chronic or developing neurological deficits associated with these insults. In addition, the better understanding of these injury cascades may one day allow new treatments to be developed that can inhibit these responses to injury and hopefully promote recovery. This chapter summarizes some of the recent data regarding progressive damage after CNS trauma and mechanisms underlying these changes.

Keywords: traumatic brain injury; spinal cord injury; progressive damage; pathophysiology; treatment

Introduction

Recent studies from both experimental and clinical investigations have emphasized the progressive nature of central nervous system (CNS) injury. In contrast to the initial concept that the majority of damage occurs at the time of the primary ischemic or traumatic insult, new evidence emphasizes that acute injury can initiate a variety of pathophysiological cascades that lead to secondary injury mechanisms associated with subacute as well as progressive injury. These findings are important from the prospective of clarifying mechanisms underlying cell death, but more importantly provide new targets for therapeutic intervention. Indeed, the observation that processes which potentially affect long-term outcome may be active days or even months after injury provides new targets to improve outcome after CNS injury. This new way of assessing and treating acute injury is also important as we think about how acute insults such as ischemic or traumatic injuries may enhance the vulnerability of the aging brain to later occurring neurodegenerative diseases. The main objective of this chapter is to summarize recent date emphasizing the progressive nature of lesion pathology after brain and

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DOI: 10.1016/S0079-6123(06)61009-1 125
spinal cord injury (SCI) and highlight potential therapeutic strategies that may be relevant to these devastating insults.

**Traumatic brain injury**

Traumatic brain injury (TBI) is a leading cause of morbidity and mortality in the United States (Langlois et al., 2000). In 2000, half of a million new cases of moderate and severe TBI were reported. Traumatic insults commonly occur in young adults as a consequence of traffic and sporting accidents. More recently, there has been an increase in the number of traumatic brain insults in the elderly due to falls and other traumatic insults. In developing countries, a significant rise in vehicular accidents has also been observed. Thus, the magnitude of the problem internationally merits increased concern regarding the prevention as well as treatment of TBI.

The pathophysiology of TBI is complicated and involves both primary and secondary insults (Graham et al., 2000a; Bramlett and Dietrich, 2004). Primary insults due to impact injury result in the rupture of membranes that lead to acute damage of neuronal, glial and vascular components. These membrane disturbances also cause metabolic stress that initiates a cascade of cellular and molecular mechanisms that can initiate both reparative as well as destructive processes. Acute consequences of TBI include alterations in the blood-brain barrier (BBB) as well as complex changes in local cerebral blood flow (LCBF) and metabolism (Cortez et al., 1989; Hovda et al., 1991; Dietrich et al., 1994). Trauma-induced glial swelling as a consequence of both vasogenic and cytotoxic edema is also a common early consequence to TBI (Bullock et al., 1991; Kimbelberg and Nordenberg, 1994). Early metabolic and hemodynamic events on their own can lead to necrotic cell death in specific vulnerable populations of cells. Severe reductions in LCBF are observed in some TBI patients and may indicate ischemic events (Bramlett and Dietrich, 2004). In human TBI, specific gray and white matter structures including the corpus callosum and hippocampus are frequently damaged after moderate and severe TBI (Povlishock and Christman, 1995; Graham et al., 2000b; Leclereq et al., 2001). Diffuse axonal injury underlies a major mechanism for the morbidity and mortality associated with TBI (Adams et al., 1989, 1991).

Various animal models have been developed that mimic some of the structural and behavioral consequences of human TBI (Gennarelli et al., 1982; Cernak, 2005). Although no one model of TBI exactly represents the human condition of brain trauma, these models are important in the understanding of critical pathomechanisms responsible for traumatic events and the eventual testing of novel therapeutic interventions. One model, fluid-percussion (F-P) brain injury has been used commonly in these preclinical TBI studies (Dixon et al., 1987). For example, after moderate parasagital F-P brain injury, the extravasation of the protein tracer horseradish peroxidase (HRP) is observed in specific brain regions associated with acute vascular damage due to shearing forces produced by the insult (Dietrich et al., 1994). In other studies, enduring changes in axolemmal permeability has been reported in white matter tracts, not necessarily associated with overt damage (Povlishock and Pettus, 1996). Thus, abnormal permeability of a variety of cellular membranes are rendered leaky to tracers after TBI (Pettus and Povlishock, 1996).

More recently, evidence for apoptotic cell death has been demonstrated in models of TBI as well as SCI (Crowe et al., 1997; Beattie et al., 2002; Lu et al., 2003). McIntosh and colleagues (Rink et al., 1995) first provided ultrastructural evidence indicating that neurons may die by apoptotic mechanisms after F-P brain injury. In that study, neurons demonstrated the classical appearance of apoptotic bodies within the cell nucleus. More recently, molecular and biochemical approaches have been used to shed light on the various receptor and intracellular cell signaling mechanisms responsible for apoptotic cell death (Keane et al., 2001a, b). The importance of this line of research to the present discussion is that emerging evidence supports the concept that apoptotic pathways may be activated for long periods after CNS injury and that mechanisms similar to programmed cell death may be responsible for some of the
progressive changes seen in the brain late after trauma (Williams et al., 2001).

In addition to apoptosis, inflammatory and immune processes also appear to play an important role in pathophysiology of TBI (Shohami et al., 1994; Schwab et al., 2001; Morganti-Kosmann et al., 2002). Following brain injury, indicators of the activation of pro-inflammatory processes including increased expression of pro-inflammatory cytokines such as IL-1β, TNFα and IL-6 have been described (Holmin et al., 1997; Morganti-Kossman et al., 1997; Kinoshita et al., 2002). Also, specific receptors that interact with these cytokines are expressed after injury and lead to the stimulation of inflammatory signaling cascades responsible for the expression of inflammatory genes (Lotocki et al., 2004). Thus, experimental studies that have targeted inflammatory processes by applying antibodies or blockers to inflammation have in some cases led to improved outcome in the subacute post-traumatic setting. For example, in several studies, treatment with the interleukin-1 receptor antagonist has been shown to lead to improved histopathological and behavioral outcome after TBI and cerebral ischemia (Toulmond and Rothwell, 1995; Rothwell and Luheshi, 2000).

Clinical evidence for progressive injury

In patients following TBI, magnetic resonance imaging (MRI) approaches have identified evidence for progressive atrophy of specific brain regions at chronic periods after trauma (Cullum and Bigler, 1986; Anderson and Bigler, 1995; van der Naalt et al., 1999). In these clinical studies, evidence for the enlargement of ventricle structures and atrophy of specific gray and white matter structures has been demonstrated. In one study, enlargement of the lateral ventricle was identified as a relatively late occurrence after TBI. This delayed response occurred without any evidence of a systemic secondary insult and therefore had no obvious underlying mechanism.

In experimental studies of TBI, the majority of investigations have evaluated traumatic outcome in the acute or subacute post-traumatic period. Thus, until recently, little information was available to determine the more chronic consequences of experimental TBI. However, in 1997, the chronic histopathological consequences of moderate F-P brain injury were first assessed at 2 months after trauma. Bramlett et al. (1997a) reported a significant enlargement of the lateral ventricle and with associated atrophy of cerebral cortical areas within the traumatized hemisphere. This study was the first to provide histopathological evidence for structural changes continuing to occur weeks to months after TBI. In subsequent studies, other investigators using similar or different injury models have complemented and extended these initial findings by assessing even longer periods of post-traumatic injury (Smith et al., 1997; Pierce et al., 1998; Dixon et al., 1999; Bramlett and Dietrich, 2002). For example, Smith et al. (1997) reported progressive injury in various forebrain areas 1 year after moderate lateral F-P injury. In a model of controlled cortical impact (CCI) injury, Dixon et al. (1999) also reported progressive damage and long lasting behavioral deficits in that trauma model of focal damage. Thus, in various laboratories using different injury models, evidence of progressive damage has been demonstrated. Most recently, Bramlett and Dietrich (2002) assessed alterations in white matter tracts 1 year after moderate F-P brain injury. In that study, significant atrophy of various white matter structures within the traumatized hemisphere provided direct evidence of progressive white matter pathology in widespread forebrain circuits. These newer data emphasize the important fact that white matter as well as gray matter structures may be highly vulnerable to progressive damage after relatively moderate degrees of trauma. Thus, therapies may have to be developed that target both gray and white matter pathology after TBI (Medana and Esiri, 2003). The importance of white matter vulnerability will again be emphasized in the SCI discussion.
Pathomechanisms underlying progressive injury

As previously discussed, both apoptotic and inflammatory cascades are felt to underlie some of the acute and subacute pathophysiological mechanisms that are responsible for cellular dysfunction and death. Recent data also indicate that these mechanisms may be active weeks and months after trauma and also participate in the progressive nature of TBI. For example, prolonged apoptotic cell death has been demonstrated in several CNS injury models (Emery et al., 1998; Beattie et al., 2002). In these studies neuronal, microglial and oligodendrocyte apoptosis has been reported days to weeks after injury. Delayed neuronal apoptosis may lead to the continued removal of axon projections that could ultimately lead to patterns of deafferentation syndromes in brain regions remote from the primary site of damage. A good example of this type of progressive and remote damage is the delayed thalamic pathology observed after parasagittal or lateral F-P brain injury (Bramlett et al., 1997a, b). Apoptotic death of oligodendrocytes could also have devastating effects on both structure and function of an axon and/or circuit. Oligodendrocyte death would lead to demyelination of projecting axons that would be expected to result in axonal dysfunction and possible progressive damage (Waxman, 1989). Indeed, electrophysiological studies have demonstrated the adverse effects of demyelination on action potentials and axonal survival (Waxman et al., 1994). Thus, progressive apoptotic cell death can have a variety of adverse consequences on the structure and function of both gray and white matter structures.

In the area of inflammation, recent data has also emphasized the progressive nature of that response to TBI. Several studies have provided evidence for long-term inflammatory responses to TBI. In brain sections stained for macrophage activation for example, evidence for inflammatory events is sometimes also seen many weeks after a traumatic brain insult. Gentleman et al. (2004) reported microglial activation, weeks after TBI. Similarly, inflammatory cells have been observed chronically in an animal model of TBI (Rodriguez-Paez et al., 2005). In this study,
Fig. 2. Light level micrographs of toluidine blue stained thick plastic sections of control (A) and traumatized tissue, specifically the lateral posterior thalamic nuclei, (100 ×) at 3 days (B), 15 days (C), 3 months (D), 9 months (E) and 12 months (F) after TBI. (A) Control tissue shows myelinated figures oriented perpendicular and parallel to the plain of the section. Normal appearing neuronal cell bodies (N), astrocytes (black arrow) and blood vessels (V) are also apparent. (B) At 3 days, axonal abnormalities including changes in axoplasmic density, unraveling of the myelin sheath (black pointer) and irregular swollen myelinated profiles (arrowhead) were observed. There is also a proliferation of microglial cells (open arrow) associated with neuronal (N) and astrocytic (black arrow) swelling. Normal appearing oligodendrocytes (double arrows) are observed. (C) At 15 days, there is an apparent increase in overall tissue vacuolation, which appears to be associated with axonal (arrowhead) and non-axonal (black arrow) profiles. An increase in microglial cells (open arrows) as well as unraveled myelin figures (black pointer) are observed. (D) At 3 months, parenchymal vacuolation is still apparent with increased numbers of microglial cells (open arrows), swollen axonal figures (arrowhead) and irregular myelin profiles (black pointer). (E) At 9 months, a dramatic number of vacuolated profiles are observed related to inflammatory cells scattered among tissue debris. An amorphous crystallized material (black star) was observed inside the vacuolated profiles. There appears to be a progressive increase in the size of the vacuolated profiles as the post-injury time increases. (F) At 12 months, the vacuolization (large black arrows) continues to progress in association with a further decrease in numbers of myelinated axons without the presence of the amorphous material described above. Reprinted from Rodriguez-Paez et al. (2005) with kind permission of Springer Science and Business Media.
macrophage/microglia infiltration and swollen axons were observed as late as 6 months using both light and electron microscopic analysis within several vulnerable structures (Fig. 2). Additionally, a temporal decline in the number of myelinated axons within the cerebral cortex (Fig. 3) and thalamus were reported which may be due to the prolonged inflammatory response observed in this study. Nonaka et al. (1999) also provided evidence for inflammatory processes being active up to 1 year after TBI. In that study, NF-kappaB was seen in both cortical and sub-cortical brain regions undergoing progressive atrophy. Thus, the potential for macrophage/microglia released toxic substances leading to tissue damage is a real possibility even weeks after TBI. To support this assumption, recent biomarker studies that have measured levels of pro-inflammatory mediators in the cerebral spinal fluid (CSF) and plasma of TBI patients have reported elevated levels, days after injury (Rancan et al., 2004). In the future, surrogate biochemical markers of tissue damage may be developed to predict and treat progressive injury mechanisms.

Abnormal protein aggregation has been implicated in the pathogenesis of a number of neurological diseases including Alzheimer’s and Parkinson’s disease (Chaudhuri and Paul, 2006). Recent evidence indicates that abnormal protein aggregation also occurs in models of cerebral ischemia and TBI (Blumbers et al., 1994; Graham et al., 1995; Lewen et al., 1995; Bramlett et al., 1997b; Hamberger et al., 2003). In models of TBI, immunocytochemical localization of beta APP and other proteins has been identified in gray as well as white matter tracts (Sherriff et al., 1994; Bramlett et al., 1997b). Whether evidence for prolonged periods of protein aggregation can be correlated with progressive tissue damage merits continued study.

Fig. 3. Estimation of myelinated axons in the cerebral cortex of animals following TBI or Sham surgery at 3 days, 15 days, 1 month, 3 months, 6 months, 9 months and 12 months. A significant difference was found for both group (p < 0.001) and time (p < 0.001) between TBI and Sham animals. TBI results in a decrease in the number of myelinated axons compared to Sham animals at all time points analyzed. In addition, there was a temporal decrease in the number of myelinated axons in the Sham animals as well. The decrease in the axonal numbers of the Sham animals may be due to a normal aging process. Reprinted from Rodriguez-Paez et al. (2005) with kind permission of Springer Science and Business Media.
Spinal cord injury

Each year in the United States, approximately 11,000 new spinal cord injuries are recorded. Currently there are over 250,000 individuals living with chronic SCI and its devastating consequences in the United States. Similar to what has been described with TBI, evidence for lesion progression has also been demonstrated in both experimental and clinical conditions of SCI (Wallace et al., 1987; Bunge et al., 1993; Crowe et al., 1997; Bruce et al., 2000; Hill et al., 2001; Guest et al., 2005; Totoiu and Keirstead, 2005). Because SCI frequently occurs in young people, it is equally important that we understand the mechanisms underlying chronic progressive damage and develop therapeutic interventions to retard cell death, axonal degeneration and demyelination in SCI patients. The pathophysiology of acute SCI is multifactorial and like TBI, includes both primary and secondary injury mechanisms (Nashmi and Fehlings, 2001; Keane et al., 2006). Primary injury mechanisms include acute SC compression, impaction, laceration, shear damage and missile injury (Norenberg et al., 2004). These acute injury mechanisms initiate a complex cascade of secondary injury mechanisms that may remain activated for months or years after injury. These initial traumatic events lead to vascular damage and hemorrhage, alterations in spinal cord blood flow, vascular thrombosis, vasospasm and loss of autoregulation. As a consequence to cellular membrane damage, metabolic abnormalities and electrolytic shifts in ions occur (LoPachin and Lehning, 1997; Li et al., 2000). In addition, neurotransmitters are released into the extracellular space leading to the abnormal activation of various receptors and intracellular signaling processes (Keane et al., 2006). Other classical injury cascades that are activated after SCI include free radical formation, lipid peroxidation and edema formation. All of these acute processes lead to the acute destruction of gray and white matter structures (Blight, 1985; Schwab and Bartholdi, 1996; Rosenberg and Wrathall, 1997).

As in brain injury, inflammatory cascades are also activated after SCI (Blight, 1992; Crowe et al., 1997; Popovitch et al., 2002; Keane et al., 2006). In addition, calpain activation has also been emphasized as an acute injury mechanism (Ray et al., 2003). Many of these injury cascades have been targets for therapeutic interventions (Blight and Zimber, 2001).

More recently, evidence for apoptotic cell death has been reported in a number of SCI models (Springer et al., 1999; Ozawa et al., 2002; Keane et al., 2006). A complex integrated apoptotic pathway involving both extrinsic and intrinsic apoptotic cascades has been reported. Also, the study of pro- and anti-apoptotic molecules, which can control apoptotic cell death, is an exciting area of current investigation. Recent work has concentrated on mechanisms that allow for the communication between the external environment with intracellular processes involved in cell survival and death. After trauma for example, specific death receptors have been shown to accumulate within specific areas of the plasma membrane called lipid rafts that contain high concentrations of cholesterol and sphingolipid (Lotocki et al., 2004). Thus, after SCI and brain trauma, TNF1 receptors accumulate in lipid rafts and assist in the formation of cytoplasmic platforms that allow scaffolding proteins to accumulate; leading to the increased interactions of proteins that lead to the activation of intracellular cascades associated with cell survival and death (Keane et al., 2006). Thus, research is currently being undertaken to understand this relatively acute injury response to neuronal vulnerability and survival. Because a large amount of cell death occurs in the acute post-traumatic period, evidence for these pathomechanisms has been commonly reported during these periods. However, since cell death during delayed post-traumatic periods is spread out and more difficult to identify, other approaches including immunocytochemistry must be used to investigate later occurring injury mechanisms.

In addition, experimental and clinical investigations have also demonstrated that apoptotic cell death, demyelination, remyelination and axon degeneration may occur weeks to months after experimental and clinical SCI (Bunge et al., 1961; Blight, 1993; Quencer and Bunge, 1996; Schwab and Bartholdi, 1996; Crowe et al., 1997; Emery...
et al., 1998; Bruce et al., 2000; Hill et al., 2001; Guest et al., 2005; Totoiu and Keirstead, 2005). For example, in a study by Crowe et al. (1997), apoptotic cells were identified from 6 h to 3 weeks after experimental SCI. Apoptotic cell death was specifically shown to be present in the white matter tracts where apoptotic cells were shown to be positive for cellular markers of oligodendrocytes. This observation is important because it indicates oligodendrocyte cell death with resulting demyelination could be an active mechanism in the progressive injury cascades associated with human SCI. Following this particular observation, various laboratories have also reported apoptotic cell death in the spinal cord including neurons, oligodendrocytes and inflammatory cells (Emery et al., 1998; Beattie et al., 2002). In addition to clarifying mechanisms of injury, these results are also important because they provide new potential targets for therapeutic interventions directed toward the acute as well as chronic post-traumatic period (Springer et al., 1999; Ozawa et al., 2002; Demjen et al., 2004).

Evidence for apoptotic cell death and Schwannosis has also been reported in specimens obtained from patients that survived long periods after SCI. In a study by Emery et al. (1998), apoptotic cells were identified using the apoptotic marker caspase-3. In that study, oligodendrocytes stained positively for this indicator of apoptotic injury in specific white matter tracts. Again, this clinical observation is important because of the role of the oligodendrocyte in axonal myelination. We know from various electrophysiological studies that if an axon is demyelinated by a toxic or traumatic insult, that axon’s ability to propagate axon potentials is severely affected (Blight and Young, 1989; Waxman, 1989). Thus, strategies that would target degenerative mechanisms or axonal conduction blockage could potentially improve outcome in SCI subjects (Blight and Young, 1989). Hill et al. (2001) have reported on extensive dieback of the corticospinal tract chronically after SCI (Fig. 4). However, there was a regenerative response of this tract along with the reticulospinal tract evidenced by an extension of collaterals into the lesion matrix. Recently, Totoiu and Keirstead (2005) have assessed chronic progressive demyelination in an animal model of SCI. Data from that study indicated that chronic progressive demyelination after thoracic injury in rats does occur for days after injury (Fig. 5). Interesting, a process of secondary demyelination was reported around 120–450 days post injury. These studies underscore the importance of targeting demyelination in the development of therapeutic interventions and again emphasize the progressive nature of neurodegeneration after SCI.

In human tissue, evidence for chronic demyelination has also been reported (Bunge et al., 1993; Norenberg et al., 2004; Guest et al., 2005). In the study by Guest et al. (2005), evidence for axonal demyelination, even a decade after human traumatic SCI was presented. Although the response was very heterogeneous among the SCI specimens evaluated, the potential for demyelination in specific spinal cord circuits in which axon profiles remained intact was observed. In this regard, evidence for spontaneous remyelination of intact CNS axons by invading Schwann cells has also been observed (Guest et al., 2005; Totoiu and Keirstead, 2005). This observation is important because other studies have reported similar cellular responses to injury that indicate some endogenous reparative mechanisms including remyelination are activated after SCI (Hagg and Oudega, 2006).

In a clinical study by Bruce et al. (2000), evidence for Schwannosis was seen in 32 out of 65 cases that survived after 24 years. The incidence of Schwannosis rose to 82% in SCI patients who survived more than 4 months. Associated with Schwannosis was intense chondroitin sulfate proteoglycan (CSPG) staining. This observation is important because the CSPGs are thought to be inhibitory molecules that reduce axonal regeneration after CNS injury (Snow et al., 1990). Thus the continued clarification of both the positive and negative consequences of Schwann cell responses to acute and chronic SCI remains an important area of investigation.

The use of human spinal cords donated for research is also allowing other injury mechanisms to be evaluated in human tissues (Norenberg et al., 2004). At The Miami Project to Cure Paralysis, approximately 115 human spinal cords have been collected from individuals surviving from 24 h to
Fig. 4. Appearance of impact site after a 12.5 mm contusion injury 1 dpi (A–C), 3 dpi (A, D, E), 8 dpi (A, F, G), 21 dpi (A, H, I) and 14 weeks (A, J, K, L) after injury. (A) The progression of cavitation. At 1 day, the impact site is hemorrhagic but no cavitation is present, and RBCs extend up to an additional 5 mm rostral (arrow); between 3, 8 and 21 days the injury site becomes progressively more defined, and it appears as a dark region in low magnification at 8 dpi and is clearly defined at 21 days. At 21 days and 14 weeks a cellular mass is present within the cavity attached to the spared rim of white matter at several points via trabeculae (arrow). One dpi (B) RBCs and (C) damaged axons at the impact site. Three dpi (D) damaged axons (small arrow) intermixed with blood cell infiltrate (large arrow) at the impact site and (E) some macrophages (large arrowhead) have infiltrated the rim of spared white matter (small arrows). Eight dpi (F) macrophages are densely packed at the center of the cavity while (G) open spaces (small arrow) are present between groups of macrophages and other nonfluorescently labeled cells (large arrowhead) that appear in scattered bands within the cavity. Twenty-one dpi (H) trabeculae, thin tissue bridges with regions of no tissue beside them, are beginning to form (small arrow), and macrophages (large arrowhead) are still present within the cavity in association with the trabeculae or as (I) macrophage rafts. Fourteen weeks after injury (J–L) autofluorescing macrophages (large arrowhead) are present within (J) fibrous trabeculae and (K) cellular trabeculae as well as (L) the cellular infiltrate rostral to the cavity. Bar in (A), 1 mm; all other bars, 100 μm. (C, E, F), same magnification. (B, D, G–L), same magnification. dpi = days post injury. Reprinted from Hill et al. (2001) with permission from Elsevier.
24 years after injury. Corresponding MRI and immunocytochemical techniques are used to analyze the injured tissue (Bunge et al., 1993; Becerra et al., 1995; Emery et al., 1998; Guest et al., 2005). With this approach, investigators have assessed the acute as well as more chronic immunohisto-pathological consequences of traumatic human SCI. For example, in early specimens taken 3 days after injury H&E staining shows damage to gray and white matter areas with a well identified central hemorrhagic lesion (Bunge et al., 1993). If specimens are stained with various immunocytochemical approaches, evidence of gliosis in the form of increased glial fibrillary acidic protein (GFAP) staining is seen surrounding the contusive site. Also, the accumulation of invading inflammatory cells such as polymorphonuclear leukocytes (PMNL) and macrophages can be visualized (Fleming et al., 2006). These types of studies conducted on human tissues are important because they verify some of the acute and more chronic histopathological changes that are reported in preclinical animal studies. This information is currently of importance to the development of new treatments targeting inflammatory processes, which is an important secondary injury mechanism after SCI (Gris et al., 2004).

Evidence for retrograde degeneration of cortical spinal tracts (CST) has also been shown in human specimens. Following damage to thoracic cortical spinal tracts for example, evidence for retrograde degeneration can be seen in remote spinal tracts (Bunge et al., 1993; Norenberg et al., 2004). In addition to retrograde degeneration, evidence for Wallerian degeneration can also be obtained using repetitive MR imaging procedures in SCI patients (Quencer and Bunge, 1996). These studies emphasize the widespread changes that occur as a consequence to local SCI. With the development and improvement of new imaging techniques, acute damage as well as the progressive nature of human SCI will be more easily investigated. In acute injury settings, better evidence for local cord compression will provide important information to help guide surgical approaches to target ischemic events. In chronic postoperative SCI cases, both cord compression and the formation and progression of cavities can be visualized in patients experiencing neurological symptoms including chronic pain. With time, some lesions expand to form cysts.
that can have a dramatic effect on progression of neurological symptoms. These imaging techniques will continue to be improved and allow specialized treatments to be developed for an individual patient undergoing progressive injuries.

**Supraspinal alterations after SCI**

In addition to changes occurring at the level of the injured spinal cord, it is also clear that SCI leads to alterations in supraspinal areas of the neuroaxis (Jain et al., 1997; Raineteau et al., 2001; Hains et al., 2003; Hubscher and Johnson, 2006; Kim et al., 2006). In this regard, there is a rich literature on experimental SCI lesioning studies in rodents and non-human primates showing evidence for degeneration of neurons in the cerebral cortex after injury. Some data indicate that cell death occurs through apoptotic mechanisms whereas more recently, additional evidence indicates that cortical cell bodies as a consequence of SCI may only undergo severe atrophy but not actually die. In these cases, the local addition of neurotrophic factors to a particular brain region reverses some of these morphological changes.

In addition to cellular changes, evidence for circuit reorganization and plasticity resulting in deactivation and reactivation of certain brain areas has been demonstrated after clinical and experimental SCI (Jain et al., 1997; Raineteau et al., 2001). Chronic SCI has also been shown to induce changes in the response of thalamic neurons to physiological activation (Hubscher and Johnson, 2006). Thus, SCI leads to alterations in brain circuits that are responsible for assessing normal and abnormal sensation. In this regard, some researchers believe that this circuit plasticity in addition to other events including chronic inflammation may account for the late development of chronic neurogenic pain in patients with SCI. A clearer understanding of local circuit changes in response to SCI may provide important information regarding how to treat these patients with neurogenic pain.

As a consequence of chronic SCI, the cerebral cortex may also undergo plastic changes in terms of shifts in cortical map architecture (Topka et al., 1991; Jain et al., 1997; Bruehlmeier et al., 1998; McDonald et al., 2002; Sabbah et al., 2002; Lotze et al., 2006). As specific inputs to the cerebral cortex are removed after SCI, other cortical areas may take over function. Thus, electrophysiological and metabolic studies have shown evidence for cortical plasticity after SCI (Hoffman and Field-Fote, 2007; Kim et al., 2006). These observations not only emphasize the potential for plasticity occurring in remote brain regions after SCI, but also may be important as we think about repairing the nervous system in the chronically injured spinal cord. If brain circuits and/or cortical maps were significantly altered due to chronic SCI, would such a consequence affect the ability to repair the nervous system and return function? Only after successful regenerative approaches are developed in the chronically injured state can these questions be answered.

**Therapeutic interventions targeting progressive injury**

Based on the complexity of brain and SCI, it is clear that the injury mechanisms are multifactorial and may require a combinational therapeutic approach. In the area of brain and SCI, various neuroprotective agents have been evaluated (Narayan et al., 2002). Neuroprotective agents such as methylprednisolone, GM1 gangliosides, lazeroids, calcium and sodium channel blockers, growth factors as well as blockers of excitotoxic process have been reported to be effective in some animal models. More recently, the use of anti-inflammatory strategies, calpain antagonists, anti-apoptotic strategies as well as agents targeting cAMP have also been investigated with various results (Blight and Zimber, 2001).

Mild hypothermia that targets multiple injury cascades has been tested with various degrees of success after both SCI and TBI (Hayashi et al., 2004; Guest and Dietrich, 2005). Although hypothermia was used in the 60’s to target both brain and SCI, profound hypothermia (27°C) commonly produced severe effects on cardiac function and increased infection rates in patients. However, in the mid-80’s, the importance of mild to moderate hypothermic treatment was demonstrated in
ischemic and traumatic animal models. Busto et al. (1987) first reported that a reduction of just 2 or 3 degrees in the temperature of the brain provided dramatic protection of CA1 neurons within the post-ischemic hippocampus. Subsequent studies demonstrated that mild hypothermia was also protective when initiated at various times after cerebral ischemia or TBI. Recently, mild hypothermia has been shown to have a dramatic effect on SCI. In a study by Yu et al. (2000), the beneficial effects of systemic hypothermia on locomotor outcome and histopathological damage were reported after contusion SCI in rats. In that study, mild hypothermia (33°C) initiated 30 min after SCI significantly improved open locomotor function and significantly reduced contusion volume weeks after injury. Because of these exciting preclinical findings, multiple clinical trials have been initiated in patients with various neurological insults including cardiac arrest, stroke, TBI and SCI. Recent multicenter trials in cardiac arrest patients have shown a dramatic benefit with mild hypothermia in that patient population (Bernard et al., 2002; Hozler, 2002). Also, individual and multicenter trials have reported that mild hypothermia shows promise in improving outcome in severe TBI patients (Jiang et al., 2006). Clinical investigations are currently determining the effects of mild hypothermia involving SCI (Guest and Dietrich, 2005). In this regard, mild hypothermia may be used during elective surgeries or in the early post-injury periods.

The beneficial effects of mild hypothermia involve multiple injury pathomechanisms. Various studies have shown that modifying brain or SC temperatures significantly affects injury-induced excitatory neurotransmitter levels, free fatty acid formation, BBB breakdown and the formation of edema. Mild hypothermia after SCI has been shown to significantly reduce the acute inflammatory response to injury (Chatzipanteli et al., 2000). Because the early inflammatory response to injury is thought to represent an important secondary injury mechanism, the ability of mild hypothermia to limit the accumulation of inflammatory cells after SCI represents an important therapeutic target for treatment. Free radical formation can come from multiple sources including prostaglandin and nitric oxide (NO) synthesis. The inducible form of nitric oxide synthase (iNOS) produces free radicals that can be destructive to tissue survival. Post-traumatic hypothermia following TBI also reduces iNOS activity and may improve outcome by affecting free radical generation as well (Chatzipanteli et al., 1999).

Mild hypothermia has been shown to be neuroprotective and promote functional outcome after TBI (Hayashi et al., 2004). Bramlett et al. (1995) first reported that post-traumatic hypothermia improved cognitive function in rats. These studies are important because cognitive deficits are some of the most severe consequences of mild, moderate and severe TBI. In regards to potential treatments for progressive injury, post-traumatic hypothermia has also been shown to reduce the progressive nature of F-P brain injury in rats (Bramlett et al., 1997a). In that study, a period of 3 h of moderate hypothermia significantly protected against progressive damage and enlargement of the lateral ventricle at 2 months after TBI. Thus, in addition to the acute benefits of therapeutic hypothermia, this therapy may also limit the amount of progressive damage after trauma. Additional studies will be required to determine what duration of the hypothermic therapy is most beneficial in providing long-term benefits in terms of functional outcome.

Finally, recent data indicate that different types of cellular transplantation strategies can promote repair and improve recovery in animal models of brain and SCI (Schouten et al., 2004; Schwab et al., 2006; Thuret et al., 2006). In some cases, cell transplantation strategies have been initiated to replace dysfunctional or dead neurons injured by the insult. Thus, an exciting field of research is currently directed toward stem cell biology and the potential for neural stem cells to repopulate the injured nervous system and improve function. Cell survival, migration, control of cellular differentiation and the integration of new cells into existing circuits are some of the challenges currently being addressed with these approaches. Alternatively, the transplantation of specific populations of restricted progenitor cells that differentiate into myelin-forming cells is another important research direction (Cao et al., 2005; Keirstead et al., 2005;
Karimi-Abdolehraee et al., 2006). In recent studies, grafted cells have been shown to form morphologically normal-appearing myelin sheaths around damaged axons. Importantly, experimental treatments targeting neuronal or glia damage after CNS injury leads to improvements in functional recovery. Because progressive damage after brain and SCI can affect a variety of different cell types, these cellular therapies may be helpful in both neuroprotective as well as reparative strategies targeting the long-term consequences of CNS injury. Continued research into the progressive nature of brain and SCI should provide new targets for treatment and better outcome in patients that sustain these devastating injuries.

Acknowledgments

This work was supported in part by NIH grants NS30291, NS38665 and DAMD17-02-1-0190. The authors also thank the members of the Bramlett/Dietrich laboratory for their important contributions to this research field.

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