Vascular Events After Spinal Cord Injury: Contribution to Secondary Pathogenesis

Traumatic spinal cord injury results in the disruption of neural and vascular structures (primary injury) and is characterized by an evolution of secondary pathogenic events that collectively define the extent of functional recovery. This article reviews the vascular responses to spinal cord injury, focusing on both early and delayed events, including intraparenchymal hemorrhage, inflammation, disruption of the blood-spinal cord barrier, and angiogenesis. These vascular-related events not only influence the evolution of secondary tissue damage but also define an environment that fosters neural plasticity in the chronically injured spinal cord. [Mautes AEM, Weinzierl MR, Donovan F, Noble L.J. Vascular events after spinal cord injury: contribution to secondary pathogenesis. Phys Ther. 2000;80:673–687.]

Key Words: Blood-spinal cord barrier, Inflammation, Metalloproteinases, Spinal cord injury.

Angelika EM Mautes
Martin R Weinzierl
Frances Donovan
Linda J Noble
S

pinal cord injury results in the initial physical disruption of structures in the spinal cord (primary insult) and in the generation of secondary events that collectively injure intact, neighboring tissue. The concept of secondary injury was originally described in 1911 by Allen,1,2 who reported an improvement of neurological function in the injured spinal cord after myelotomy and removal of the contused tissue. These results suggested that noxious agents in the contused tissue were damaging adjacent, intact spinal cord segments and led to the development of the concept of primary and secondary injuries. Primary injury typically refers to the initial mechanical damage, whereas secondary injury is progressive cell injury that begins in the gray matter and progresses into the white matter.3

After traumatic injury, the primary mechanical injury to spinal cord blood vessels results in pronounced sequelae of secondary events, beginning with prominent hemorrhage into different compartments that correspond to the epidural, subdural, subarachnoid, and intramedullar spaces.4,5 Blood can be toxic to the central nervous system (CNS).6 In experimental models of traumatic spinal cord injury, the early intraparenchymal distribution of hemorrhage coincides at later time points with the appearance of a cavity, which is partially filled with nonneuronal structures.4,5 Together, the findings illustrate that the disruption of blood vessels, a consequence of the primary insult, is integral to subsequent pathogenesis.

Secondary pathogenesis after traumatic spinal cord injury is related not only to grossly injured vessels but also to complex responses of the intact spinal cord vasculature. These responses include the disruption of the blood-spinal cord barrier and the generation of an inflammatory response. Both barrier disruption and inflammation disturb the microenvironment and expose neurons to plasma-derived cells and molecules that can be injurious to intact, neighboring tissue.7

This review is divided into 5 sections. In the first section, we describe the blood supply to the human spinal cord. It is important to appreciate the vascularity of the spinal cord, because it is known that vulnerability within the gray and white matter is, in part, related to segmental distribution of blood vessels. In addition, the anatomy of the vascular structures provides clues regarding the development of secondary injury along the axis of the spinal cord. In section 2, we characterize the secondary tissue responses to intraparenchymal and subarachnoid hemorrhage, which are direct consequences of the physical shearing or tearing of blood vessels. In section 3, we examine inflammatory responses that are implicated in both cell injury and synaptic plasticity in the injured spinal cord. In section 4, we provide an overview of the blood-spinal cord barrier, the consequences of barrier disruption, and the factors that modulate this abnormal permeability after spinal cord injury. In the final section, we describe the metalloproteinases, which are integral to barrier disruption, inflammation, and angiogenesis in the injured spinal cord.

Blood Supply to the Spinal Cord

The blood supply to the human spinal cord is provided by longitudinally oriented vascular trunks that receive a segmental arterial supply. With few exceptions, there is remarkable overlap of vascular territories.8

Extrinsic Arteries

The mature spinal cord receives its blood supply from a variable series of arteries that arise, in most cases, from the topographically closest arteries outside the vertebral column. A segmental spinal artery enters the intervertebral foramen and divides into 3 branches outside the spinal canal at each segmental level of the spinal cord: the anterior and posterior longitudinal spinal canal arteries and the radicular artery (Fig. 1).8 The radicular artery continues along the nerve root and divides into an anterior radicular artery and a posterior radicular artery. After penetrating the dura mater, the anterior and posterior radicular arteries join 3 major arteries on the surface of the spinal cord: the anterior median longitu-
dinal spinal artery and the right and left posterolateral longitudinal spinal arteries (Fig. 1). Numerous anastomoses exist among these arteries, which collectively form an irregular net of vessels on the pia mater referred to as the “pial plexus” or “vasocorona” (Fig. 2). This anastomotic arrangement is particularly critical in the spinal cord injury, where local blood flow may be impaired by the injury.

Branching of each radicular artery into anterior and posterior radicular arteries to the spinal cord is the exception in humans. On average, there are 7 or 8 ventral vessels and approximately 15 dorsal vessels. The most substantial vessels in the cervical region are radicular vessels that typically arise from the deep cervical artery and enter the spinal cord between C5 and C7. There is one large anterior radicular artery that serves the thoracolumbar region. This artery, initially described in 1882 by Adamkiewicz, is integral to the blood supply to this region. Thoracolumbar ischemia has been demonstrated in animal models when the anterior radicular artery is occluded.

**Intrinsic Arteries**

The intrinsic arteries of the spinal cord consist of central arteries, which originate from the anterior median longitudinal spinal artery and traverse the ventral median fissure sulcus, and arterioles, issuing from the pial plexus and the posterolateral longitudinal spinal artery (Fig. 2). The distribution of these central arteries varies along the axis of the cord, with the densest distribution in the cervical region (8–13 arteries per centimeter) and in the thoracolumbar region (2–3 arteries per centimeter).

The intramedullary territories of arterial supply can be roughly divided into 2 zones (Fig. 2). The first zone encompasses approximately two thirds of the cross section of the spinal cord and is supplied by the anterior medial longitudinal spinal artery (including the central artery and the vasocorona). The second zone serves the dorsal white matter and the dorsal horn and is supplied by arteries issuing from the posterolateral longitudinal spinal artery and the posterior part of the vasocorona. An intermediate zone is variably supplied by one system or the other. These intramedullary territories of arterial supply are characterized by different flow directions, involving both centrifugal (central artery) and centripetal (vasocorona) systems. Together, these systems may define a watershed zone.

**Concept of Watershed Zones**

A *watershed zone* is a region that does not receive a direct blood supply but rather is dependent on overlapping terminal vascular fields. Regions within a watershed zone, therefore, may be particularly vulnerable if any of the contributing vascular fields are interrupted. Adamkiewicz was the first clinician to differentiate territories of blood supply in the spinal cord. The early studies emphasized the variability in the number of vessels in certain areas of the spinal cord as well as the presence of overlapping vascular territories. The medullary segments with the highest incidence of arterial tributaries correspond to the regions with the highest density of neurons (ie, the cervical and thoracolumbar enlargements). In these areas, the anterior medial longitudinal spinal artery is also of large caliber. In the thoracic region, the vessels are fewer and smaller, and they anastomose with a more diminutive anterior longitudinal artery. These distinct differences in the vascularity in the thoracic cord are thought to contribute to the watershed zone.

Although watershed zones have been implicated in secondary pathogenesis after spinal cord injury, there is reason to challenge this concept. The segmental variability in blood supply may not be predictive of vulnerability but may reflect local metabolic requirements. Neuronal activity depends on an adequate blood supply. In the spinal cord, there is a close correlation between segmental supply and the volume of gray matter. Thus, the segmental, vascular arrangements may be indicative of local metabolic needs that are dictated by neuronal activity.

**Venous Drainage of the Spinal Cord**

Large anterior and posterior central veins collect blood from both sides of the spinal cord accompanying the entering arteries but do not necessarily drain blood from the same areas as supplied by the nearest artery. The posterior central vein has no arterial counterpart and drains into a posterior median longitudinal vein. Between the anterior and posterior central veins there is a central anastomosis that encircles the central canal. Radial veins join the venous pial plexus around the circumference of the spinal cord. From the anterior and posterior longitudinal veins, the blood drains into anterior and posterior radicular veins that lie parallel and adjacent to the nerve root.

**Partial Flow Theory**

The blood flow to the spinal cord is not unidirectional, and blood flow can reverse within some vessels. There are ascending and descending blood flow currents in the ascending and descending branches of the radicular arteries forming the anterior median and posterolateral longitudinal spinal arteries. This partial flow theory, originally described by Adamkiewicz and verified more recently with spinal angiography, acknowledges that blood flow into each spinal cord segment reflects the partial contribution of both ascending and descending flow currents. In spinal cord injuries, this multidirectional flow may serve to protect the spinal cord when
local blood vessels are injured and incapable of supporting surrounding tissue.

Reversal of blood flow has been observed in response to normal physiological stimuli (changes in the posture of the spinal cord) and pathological conditions. This reversing capability may likewise confer added protection in spinal cord injuries by diverting flow to those areas of greatest need.

**Experimental Models of Spinal Cord Injury**

Experimental models of spinal cord injury include contusions, compression, ischemia, and crush injuries. Each model generates injuries that mimic certain clinical aspects of the mechanical damage and the ensuing posttraumatic ischemia. Researchers have also used either transection or hemisection to study injury. Transection and hemisection are less clinically relevant but offer the distinct advantage of consistent reproducibility. The hemisection model offers the ability to compare cellular responses in the ipsilateral and contralateral spinal cord. The common denominator among the experimental models is destruction of both gray and white matter. The lesioned site is characterized by early hemorrhage, breakdown of the blood-spinal cord barrier, neuronal cell loss, and infiltration of monocytes and macrophages and of neutrophils. Progressive necrosis and active phagocytosis are key processes that act in concert to remodel the cytoarchitecture of the spinal cord. Several months postinjury, the injured spinal cord typically exhibits marked axonal degeneration and a fluid-filled cavity that occupies at least part of the gray matter and adjacent white matter.
Perhaps one of the most commonly used experimental models for studying spinal cord injury is the weight-drop contusion model. This model, originally described in 1911 by Allen, involves dropping a calibrated weight a defined distance onto the exposed dura of the spinal cord. Since its original description, the procedure has been modified in order to better produce a graded, reproducible injury. The model has been instrumental in the development of optimal therapies for treating spinal cord injury, including the use of methylprednisolone (MP), which was shown to be neuroprotective after experimental contusion injuries. These studies provided an impetus for subsequent evaluation of MP in human spinal cord injury, where it was found to improve neurologic function when given within 8 hours after injury.

The contusion model has also offered important insights into the pathogenesis of secondary spinal cord injury. A component of early cell death after spinal cord contusion injury is related to vascular events. The disruption of vessels leads to intraparenchymal hemorrhage; the disruption of the blood-spinal cord barrier, which is closely associated with edema formation; the release of vasoactive molecules that influence the extent of spinal cord perfusion; and the loss of autoregulation. Together, these vascular events result in varying degrees of spinal cord ischemia. Ischemia not only results in the death of cells but also triggers a cascade of secondary events, including excitotoxicity, that lead to further damage to the spinal cord. Given this background, it is not surprising that there has been a considerable research effort directed toward the develop-
Development of strategies for limiting the extent of ischemic injury. Particular attention has been directed at the penumbral zone surrounding the ischemic, necrotic core. This region exhibits progressive cell death over time and thus is a target for therapeutic intervention. The clinical relevance of the penumbral zone is based, in part, on an important observation that was first identified in an experimental model of spinal cord contusion injury, namely that effective locomotion can occur with only 5% to 10% of the original axonal population. Thus, protecting the penumbral zone could have an important impact on the extent of functional recovery.

**Spinal Cord Injury and Hemorrhage**

**Relationship to Secondary Pathogenesis**

Mechanical trauma to the spinal cord causes immediate vasospasm of the superficial vessels and intraparenchymal hemorrhage, which is initially localized in the highly vascularized and most vulnerable central gray matter. The generation of central gray matter injury may be best understood by a consideration of the distribution of the mechanical forces in the traumatized spinal cord. For example, an anteroposterior compressive force on a flexible tube filled with gelatin, a physical model of the contused spinal cord, produces longitudinal stress that is most intense in the center of the tube. Thus, the vessels on the surface of the spinal cord are relatively spared from initial insult, whereas the microvasculature in the gray matter is subjected to stretching and shear stress, a consequence of the difference in compliance between the gray and white matter. The immediate mechanical damage to gray matter microvasculature impairs the microcirculation and impedes perfusion, leading to a profound reduction of spinal cord blood flow accompanied by impairment of autoregulation. The impaired blood flow in the traumatized spinal cord may be further compromised by systemic responses, including posttraumatic hypotension, bradycardia, and decreased cardiac output, that exacerbate the ischemic damage.

**Intraparenchymal Hemorrhage**

Noble and Wrathall studied the anatomical characteristics of central hemorrhage after graded contusive injury in the rat. The size of the hemorrhage, which is directly proportional to the severity of the initial impact, is maximal at the injury site and extends into the dorsal columns in the rostral and caudal segments. The central hemorrhage occupies the gray matter and a variable proportion of adjacent white matter at the epicenter of the injury. At distant sites, the hemorrhage is typically found in the most central part of the dorsal column (Fig. 3).

There is both indirect and direct evidence that hemorrhages may be damaging to the CNS. Perhaps the most compelling indirect evidence relates to the evolution of a fusiform-shaped cavity that closely approximates the distribution of early intraparenchymal hemorrhage. Evidence that tissue damage may be a consequence of this intraparenchymal hemorrhage comes from a study by Bullock and Fujisawa, who demonstrated that injection of blood into the extra-axial space resulted in cell damage.

Several factors may contribute to the tissue necrosis following central hemorrhage. Blood flow is reduced in tissue around hemorrhage, resulting in different degrees of ischemia. Ischemic damage may arise from one or all of the following events: (1) vasogenic edema as a consequence of blood-spinal cord barrier breakdown, (2) direct compression by adjacent tissue, and (3) vasospasm, which occurs as a result of mechanical trauma or due to exposure to red blood cell components, including oxyhemoglobin and endothelin. Free radicals are rapidly generated upon reperfusion of ischemic tissue or hypoperfused tissue. Activation of phagocytic cells and liberation of catalytic metal ions, which are present in high concentrations during the degradation of hemoglobin, is one process that results in the production of free radicals. The CNS is particularly prone to free radical damage. The cell membranes are rich in polysaturated fatty acid chains, which are sensitive to free radical attack. In addition, the CNS has
limited antioxidant defense mechanisms. The brain and spinal cord exhibit low levels of catalase activity and only moderate levels of superoxide dismutase and glutathione peroxidase.  

Heme Oxygenase-1

The experimentally contused spinal cord generates substantial intraparenchymal hemorrhage. A focus of our laboratory has been to define how neural and glial cells respond to hemorrhage. This research interest has led us to investigate the role of heme oxygenase-1 (HO-1) in the traumatized spinal cord. Heme oxygenase (HO) is the rate-limiting enzyme that is involved in the degradation of heme to biliverdin, carbon monoxide, and iron. This pathway is of particular importance because bile pigments are potent antioxidants and may therefore serve an important function in cellular defense against free radical-mediated cell damage.

There are at least 3 HO enzymes: (1) HO-1, the inducible form found mainly in microglia, (2) HO-2, the constitutive form, and (3) HO-3, whose transcripts have been recently reported in the CNS. Heme oxygenase-1 is induced by a variety of stimuli, including heme. In the intact spinal cord, basal expression of HO-1 is restricted to neurons. However, we have demonstrated marked induction of HO-1 in astrocytes and in microglia and macrophages after contusion injury. The pattern of induction corresponds to regions exhibiting posttraumatic hemorrhage. Hemorrhage, therefore, may be a potent inducer of HO-1 after spinal cord injury. To begin to test this hypothesis, we have exposed the subarachnoid space, overlying the intact spinal cord, to lysed blood. Preliminary data demonstrate focal induction of HO-1 in glia, a finding that suggests these cells may play an important role in sequestering and metabolizing heme (Fig. 4).

The central question, however, is whether glial induction of HO-1 is protective or detrimental to the traumatized spinal cord. Although somewhat controversial, there is growing evidence that HO-1 is protective in CNS injury. Panahian et al. in a recent study using transgenic mice that overexpress HO-1, found an attenuation of cell injury after cerebral ischemia. Whether similar protection occurs in the hemorrhagic traumatized spinal cord remains to be determined.

Inflammation

Inflammation: The Patient With Spinal Cord Injury

Inflammation is central to secondary pathogenesis after spinal cord injury. In the clinical setting, there is compelling evidence to establish early and prolonged inflammation in the traumatized spinal cord. This inflammation is exemplified in a clinical retrospective analysis involving 1,917 patients over a period of 10 years. The number of white blood cells was found to be elevated in the cerebrospinal fluid within the first 7 days after spinal cord injury. It is likely that the increased number of leukocytes is indicative of an early immune response. The inflammation is not limited to the acutely traumatized spinal cord, but rather occurs over a period of weeks in regions of white matter undergoing wallerian degeneration. Recent research has shown elevated plasma levels of inflammatory mediators, including cytokines (ie, interleukin-2, interleukin-6), the soluble interleukin-2 receptor, and intercellular adhesion molecule-1 (ICAM-1), in patients with long-standing spinal cord injury. Such findings emphasize prolonged inflammatory responses in human spinal cord injury.

Inflammatory Cells

Microglia, leukocytes (lymphocytes, neutrophils, and monocytes), and astrocytes contribute to the cellular inflammatory response after experimental spinal cord injury. Our review focuses on those cells that are most closely associated with the vasculature, namely leukocytes and macrophages.

Time Course of the Inflammatory Response

Traumatic spinal cord injury results in both a primary injury and a cascade of secondary processes that collectively lead to additional loss of tissue. Post-traumatic inflammation, characterized, in part, by the accumulation of activated microglia and macrophages, is thought to contribute to secondary pathogenesis.
Infiltration of neutrophils signals an early inflammatory response after spinal cord injury. Neutrophils infiltrate the traumatized cord within the first hour post-injury, peak at 24 hours post-injury, and begin to diminish by 48 hours post-injury, and are negligible by 7 days post-injury. The early appearance of neutrophils most likely reflects the early hemorrhage. Activated macrophages are apparent early after injury, but they plateau between 2 and 4 weeks post-injury in the contused spinal cord. There is a rapid transformation of resident microglia into macrophages, which precedes infiltration of macrophages derived from infiltrating monocyte precursors. B and T lymphocytes appear in the injured spinal cord within hours of the injury and persist up to 7 days post-injury. There is marked infiltration of B lymphocytes between 3 and 6 hours post-injury, and both B and T lymphocytes are identified in the spinal cord by 4 days post-injury. There is a marked decline in these cells by 7 days post-injury.

The role of inflammation in spinal cord injury is controversial. Inflammatory cells are associated with delayed neuronal death and demyelination, and they also may be integral to neural regeneration. Infiltrated leukocytes and endogenous glia, acting as macrophages, mediate tissue damage, including myelin vesiculation and lipid peroxidation, through the generation of a variety of toxic molecules (eg, reactive oxygen and nitrocellular radicals, cytokines and chemokines). These molecules are thought to damage surrounding healthy tissue. This hypothesis is supported by a recent study by Carlson et al that demonstrated a correlation between the axial extent of tissue damage and the numbers of macrophages and microglia. Strategies aimed at blocking neutrophil or macrophage influx and at inhibition of phagocytic and secretory activity of macrophages in the injured spinal cord have resulted in neuroprotection and improved locomotor function.

There is also evidence indicating that inflammation may play a beneficial role in the traumatized spinal cord. Macrophages and microglia promote regeneration of axons by scavenging myelin and neuronal debris, by producing the proregenerative cytokine, transforming growth factor-beta (TGF-β), and by enhancing neurite outgrowth. The enhancement of neurite outgrowth is exemplified by a recent study that demonstrated that the extent of axonal regeneration in the injured rat spinal cord was correlated with the presence of phagocytic cells. In that study, nitrocellulose membranes, which were treated with TGF-β or coated with microglia, were co-transplanted with fetal spinal cord tissue into the injured spinal cord of an adult rat. Cut dorsal roots were apposed to both sides of the nitrocellulose. Four weeks later, regenerated sensory axons were found to be associated with macrophages.

Conversely, this axonal regeneration could be experimentally inhibited by implanting a nitrocellulose strip containing a macrophage inhibitory factor, suggesting a prominent role for phagocytic cells in the regenerative response.

The Blood-Spinal Cord Barrier

Arteries in the subarachnoid space become arterioles as they penetrate into the substance of the spinal cord. The subarachnoid space surrounds the penetrating arteriole and is referred to as the “Virchow-Robin space” (Fig. 5). This space contains not only cerebrospinal fluid but also pia-arachnoid cells, which have the capacity to behave as phagocytes. The arteriole wall is composed of endothelial cells, which are surrounded by a smooth muscle coat. The arteriole terminates at the level of the capillary. The capillary has several distinguishing features; it is smaller in diameter than that of the arteriole, lacks a smooth muscle coat, and is marginally contacted by pericytes. A basal lamina surrounds the endothelial cell and splits to enclose each pericyte. Astrocytic foot processes abut the parenchymal side of the basal lamina (Figs. 6 and 7).

The blood-brain/spinal cord barrier is located at the level of the capillary and is composed of specialized endothelial cells that regulate and restrict transport of molecules into the CNS. This specialized interface provides a stable microenvironment, which is necessary for normal neuronal function. The morphologic basis of the blood-brain/spinal cord barrier is attributed, in part, to the presence of tight junctions between adjacent endothelial cells that effectively block the intercellular movement of large molecules, including plasma proteins. The restrictive nature of the barrier is also attributed to a complex glycoprotein-rich glycocalyx, which is located on the luminal (humeral) front of the endothelial cell. This glycocalyx is anionic (negative) in charge (Fig. 8) and thus serves as a repulsive interface to circulating plasma proteins that bear a similar charge. The basement membrane maintains a close relationship with the abluminal (parenchymal) side of the endothelial cell. This structure is thought to play a critical role in maintaining the integrity of the barrier, in part, by providing structural support to the endothelial cell wall.

Time Course of Barrier Disruption

Disruption of the blood-spinal cord barrier after spinal cord injury is characterized by a transient loss of anionic charged sites along the endothelial glycocalyx and indiscriminate extravasation of plasma proteins (Figs. 8 and 9). The time course for barrier disruption to circulating molecules varies from 4 to 28 days postinjury and is not restricted to the injured site but extends along the axis of the spinal cord. A recent study by Popovich et al demonstrated that the blood-spinal cord barrier...
remains permeable to small molecules such as \([14C]-\) aminoisobutyric acid up to at least 28 days postinjury. Such an extended time course for barrier breakdown has been confirmed in a recent magnetic resonance imaging study after a spinal cord contusion.97

**Importance of Barrier Disruption**

Altered barrier permeability exposes the spinal cord to the toxic effects of inflammatory cells.7 Amino acid neurotransmitters such as glutamate and glycine, when present at high concentrations, also can be toxic to cells.7 As noted earlier, there is substantial evidence that infiltrating leukocytes can be injurious to neural structures. There is also evidence that barrier disruption, in the absence of an inflammatory response, can be detrimental. For example, barrier breakdown resulting from a transient rise in blood pressure has been shown to produce a pathology consistent with irreversible tissue damage.98 Some researchers99,100 have demonstrated that transient opening of the blood-brain barrier consistently induces the stress proteins HSP32 and HSP70. The stress protein HSP70 is an established marker of cell injury,101 whereas HSP32 has been reported to be an indicator of oxidative stress.102,103 Taken together, these findings emphasize the importance of barrier breakdown in secondary pathogenesis after spinal cord injury.

**Factors That Contribute to Abnormal Barrier Permeability**

The traumatized spinal cord is exposed to a wide spectrum of substances, including cytokines32 and vasoactive peptides that have been implicated in barrier disruption and edema formation.32,104 In our laboratory, we have been particularly interested in the contribution of the vasoactive peptide endothelin-1 (ET-1) in modulation of the blood-spinal cord barrier after spinal cord injury.

Endothelin-1 is a 21-amino acid peptide that was originally identified by Yanagisawa et al105 in the supernatant of aortic endothelial cells. Since the characterization of ET-1, 3 additional isoforms have been identified: endothelin-2 (ET-2), endothelin-3 (ET-3), and vasoactive intestinal constrictor polypeptide (VIC). These peptides, encoded by separate genes, consist of 21 amino acid residues joined by 2 disulfide bridges with 6 conserved amino acid residues at the carboxy terminus.106

There is evidence that implicates ET-1 in barrier disruption after spinal cord injury. There is an increased expression of this peptide in the injured spinal cord107 which correlates with the pattern of blood-spinal cord barrier breakdown.108 Intrathecal administration of ET-1 in the intact spinal cord results in disruption of the blood-spinal cord barrier.108 Finally, pharmacologic

---

**Figure 5.**
Schematic illustration of the Virchow-Robin space, which begins at the surface of the spinal cord and terminates at the level of the capillary.
blockade of ET-1–mediated vasoconstriction attenuates breakdown of the blood-spinal cord barrier after traumatic spinal cord injury.\textsuperscript{108}

The biology of ET-1–mediated pathology relates, in part, to the endothelin receptor subtypes. There are at least 3 endothelin receptor subtypes, designated ETA, ETB\textsubscript{1}, and ETB\textsubscript{2}, that influence vascular reactivity in the CNS.\textsuperscript{106,109} The ET\textsubscript{A} receptor is localized in vascular smooth muscle and mediates prominent, sustained vasoconstriction. The ET\textsubscript{B} receptor is subdivided into the ET\textsubscript{B\textsubscript{1}} and ET\textsubscript{B\textsubscript{2}} types. The ET\textsubscript{B\textsubscript{1}} subtype is localized in vascular endothelial cells and generates vasodilation, whereas the ET\textsubscript{B\textsubscript{2}} receptor is present in vascular smooth muscle and produces vasoconstriction. Thus, ET-1–mediated vascular changes (ie, barrier disruption, vasoconstriction) in the traumatized spinal cord could be executed through either the ET\textsubscript{A} or ET\textsubscript{B\textsubscript{2}} receptor subtype. The ET\textsubscript{A} receptor has a particularly close link to vasospasm, secondary to subarachnoid hemorrhage,\textsuperscript{46} and prolonged vasoconstriction,\textsuperscript{6} concomitant with ischemic damage.\textsuperscript{110}

There are 3 probable sources of ET-1 after hemorrhage. The first source may be derived from the humoral compartment. Increased ET-1 levels, which are observed after subarachnoid hemorrhage,\textsuperscript{111} could enter the CNS through a disrupted blood-CNS barrier. The second source may be the injured tissue itself, and the third source may be red blood cells. An in vitro study by Tippler et al\textsuperscript{112} demonstrated a 3-fold increase in ET-1 immunoreactivity in the lysate of disrupted red blood cells.

The mechanism whereby ET-1 disrupts the blood-spinal cord barrier may be related to its role as a prominent vasoconstrictor. Endothelin-1 has the ability to produce prolonged periods of vasospasm,\textsuperscript{6} resulting in ischemic damage to neurons\textsuperscript{110} and disruption of the barrier.\textsuperscript{104} Most recently it has been reported that intrathecal administration of endothelin generates free radicals.\textsuperscript{113} Such a finding suggests that endothelin-mediated barrier disruption may reflect an oxidative insult.

**Metalloproteinases and Spinal Cord Injury**

Matrix metalloproteinases (MMPs) are a large family of enzymes that mediate extracellular matrix degradation and release of growth factors and cytokines from the matrix. This important family of inflammatory enzymes has become a new area of interest in the study of CNS injury. Some researchers\textsuperscript{114,115} have demonstrated the presence of MMPs in disease and injury in the brain and spinal cord. Experimental models suggest that MMP activity is required for inflammatory cell infiltration and may contribute to both abnormal blood-brain barrier permeability and ischemia-induced angiogenesis following injury.
MMP Family
The MMPs are zinc- and calcium-dependent endopeptidases that together can hydrolyze essentially all of the components of the extracellular matrix. These endopeptidases are roughly classified by substrate classes and therefore fall into the categories of collagenases, gelatinases, stromelysins, membrane-type MMPs, and “other MMPs.”

MMPs and Inflammation
Matrix metalloproteinase activity is required for the inflammatory cell infiltration that occurs following spinal cord injury and most likely contributes to early barrier disruption. The early inflammatory response involves an initial wave of infiltrating neutrophils, followed by migration of monocytes and macrophages into injured segment. Each of these inflammatory cells expresses MMPs, including MMP-2 (gelatinase A), MMP-8 (neutrophil collagenase), MMP-9 (gelatinase B), MMP-11 (stromelysin-3), and MMP-12 (metalloelastase). Together, these MMPs are thought to participate in infiltration and migration, tissue destruction, extracellular matrix degradation, blood-spinal cord barrier disruption, and edema.116–122

Matrix metalloproteinase activity is thought to be required for the infiltration of inflammatory cells because they must degrade the basal lamina that surrounds blood vessels, a structure that is normally a barrier to cell migration. The degradation of this basal lamina leads to the next consequence of MMP activity: increased permeability of the blood-spinal cord barrier. In the CNS, vascular endothelial cells form a tight junction, and, together with astrocyte processes, form a specialized basal lamina. During the infiltration of inflammatory cells and during angiogenesis, this basal lamina becomes degraded and the barrier becomes permeable.123 Several experimental models of brain injury demonstrate the relationship between MMP activity and blood-brain barrier permeability. For example, injection of proinflammatory cytokines or tumor necrosis factor-α induces MMP-9 expression in the brain.124 The subsequent breakdown of the blood-brain barrier
can be directly attributed to MMP activity in that treatment with MMP inhibitors blocks this abnormal barrier permeability. A similar relationship has been established in ischemic brain injury. Romanic et al demonstrated that an MMP-9 neutralizing antibody reduced infarct size, thus underscoring the contribution of MMP activity in the ischemic brain.

**MMPs and Angiogenesis**

Angiogenesis occurs following spinal cord injury in response to localized tissue hypoxia. Membrane type 1 MMP (MMP-1) is found on endothelial cells and is required for endothelial cell migration in angiogenesis. Hypoxic and ischemic conditions occur as a result of the physical damage to blood vessels as well as the hypoperfusion and vasodilation that accompany spinal cord injury. Proangiogenic factors released from the injured site cause rapid increases in vascular permeability. Fibrinogen leaking from the microvascular bed is polymerized into a fibrin matrix. Endothelial cells require MMP-1 activity to degrade this matrix and invade the surrounding tissue. Many researchers have demonstrated increased MMP-1, -2, and -9 activity at sites of angiogenesis. Vascular endothelial growth factor expression, known for its proangiogenic as well as endothelial cell permeability activities, also strongly correlates with MMP-2 and -9 expression and is thought to up-regulate MMP-9 expression. Matrix metallocollagenase proteolysis has 4 consequences: (1) it permits invasion of endothelial cells into the surrounding matrix, (2) it generates extracellular matrix degradation products that are chemotactic for endothelial cells, (3) it activates and releases growth factors localized in the extracellular matrix, and (4) it results in increased permeability of the blood-spinal cord barrier, as discussed earlier. Whether angiogenesis is beneficial or detrimental to the traumatized spinal cord is not clear. Recent data support a role for angiogenesis in promoting neural regeneration, but other data suggest that inhibition of angiogenesis is neuroprotective.

**Summary**

Spinal cord injury results in injury to both neural and vascular elements. The extent to which the vascular injury contributes to secondary pathogenesis is dependent on not only the initial disruption of blood vessels, leading to prominent intraparenchymal hemorrhage, but also on progressive disruption of the blood-spinal cord barrier coincident with the infiltration of inflammatory cells. Collectively, these events influence both acutely and chronically injured spinal cords and define, in part, the extent of functional recovery.

**References**


74 Hirschberg DL, Yoles E, Belkin M, Schwartz M. Inflammation after axonal injury has conflicting consequences for recovery of function: rescue of spared axons is impaired but regeneration is supported. J Neuroimmunol. 1994;50:9–16.


84 Colton CA, Gilbert DL. Production of superoxide anions by a CNS macrophage, the microglia. FEBS Lett. 1987;223:284–288.


