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# Drug development in spinal cord injury: What is the FDA looking for?

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**Abstract—**It has long been recognized that much of the post-traumatic degeneration of the spinal cord following injury is caused by a secondary injury process that occurs during the first minutes, hours**,** and days after spinal cord injury (SCI). A key biochemical event in that process is reactive oxygen-induced lipid peroxidation (LP). Indeed, the administration of a high-dose regimen of the glucocorticoid steroid methylprednisolone (MP) has been shown to inhibit post-traumatic LP in animal models of SCI, and to improve neurological recovery in spinal cord-injured humans. This resulted in the registration of high-dose MP for acute SCI in several countries, although not in the U.S. Nevertheless, this treatment quickly became the standard of care for acute SCI, since it was already on the U.S. market for many other indications. Subsequently, it was demonstrated that the nonglucocorticoid 21-aminosteroid tirilazad could duplicate the antioxidant neuroprotective efficacy of MP in SCI models, and evidence of human efficacy has been obtained. This article explains the process of the discovery, development, and Food and Drug Administration regulation of new drugs for SCI; reviews the past development of MP and tirilazad for acute SCI; identifies the regulatory complications involved in future SCI drug development; and suggests some promising therapeutic approaches that could either replace or be added to high-dose MP.

**Key words:** lipid peroxidation, methylprednisolone, secondary injury, spinal cord injury, tirilazad.

## INTRODUCTION

Although spinal cord injury (SCI) can victimize active individuals at any age, most injuries occur in young adults between the ages of 16 and 34. Those who survive their initial injuries can now expect to live long lives because of improvements in medical and surgical care, although intensive rehabilitation and prolonged disability exacts a significant toll on the individual, the family, and society. Effective ways of restoring and maintaining function could markedly improve the outlook for those with traumatic SCI by enabling higher levels of independence and productivity.

**Abbreviations:** ATP = adenosine triphosphate, CDER = Center for Drug Evaluation and Research, CNS = central nervous system, FDA = Food and Drug Administration, GM1 = monosialoganglioside, IND = Investigational New Drug Application, LP = lipid peroxidation, MP = methylprednisolone, NASCIS = National Acute Spinal Cord Injury Study, NDA = New Drug Application, NIH = National Institutes of Health, NINDS = National Institute of Neurological Disorders and Stroke, OP-1 = osteogenic protein 1, ROS = reactive oxygen species, SCI = spinal cord injury.

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The potential for pharmacological intervention to either preserve or restore neurological function after SCI exists, because most traumatic injuries to the spinal cord do not involve actual physical transection of the cord, but rather damage to the spinal cord as a result of a contusive, compressive, or stretch injury. Typically, residual white matter containing portions of the ascending sensory and descending motor tracts remains intact, allowing for the possibility of neurological recovery. However, during the first minutes and hours following injury, a secondary degenerative process is initiated by the primary mechanical injury that is proportional to the magnitude of the initial insult. Nevertheless, the initial anatomical continuity of the injured spinal cord in the majority of cases, together with our present knowledge of many of the factors involved in the secondary injury process, has led to the notion that pharmacological treatments that interrupt the secondary cascade, if applied early, could improve spinal cord tissue survival, and thus preserve the necessary anatomic substrate for functional recovery to take place.

## PROCESS OF DRUG DEVELOPMENT AND FDA REGULATION

The Federal Food, Drug, and Cosmetic Act, passed by Congress in 1963, provides the current legal basis for federal regulation of the development and marketing of new compounds for use in treating human disease. That act set forth standards for demonstrating that new chemical entities intended for use in humans are not only reasonably safe, but that they also possess therapeutic efficacy in the setting of their intended indication. In order to meet those regulations, a Food and Drug Administration (FDA)-mandated process of new drug testing, first in animals and then in humans, has been developed. The branch of FDA that is responsible for overseeing the testing and approval process for new drugs is the Center for Drug Evaluation and Research (CDER).

The preclinical testing phase, carried out by the sponsor (usually a pharmaceutical company), involves a demonstration that the compound exhibits pharmacological activity in animal models consistent with its intended use in humans, and that toxicological studies carried out in at least two species (e.g., rat and dog, rat and monkey) do not show any acute toxicities that suggest unreasonable risks are likely to be encountered in early-stage clinical studies. At this point in the discovery/development process, an Investigational New Drug Application (IND) is filed with the FDA that documents the preclinical pharmacological and toxicological testing, as well as the chemical purity, stability and manufacturability, and planned early clinical protocols. The CDER has 30 days in which to respond to the IND by issuing either a clinical hold or approval of the early clinical testing plan put forth by the sponsor. In the case of compounds intended for clinical use for unmet medical needs that are associated with high morbidity (e.g., SCI) or mortality, the FDA has created an expedited drug development and review process ("fast track" under Subpart E of the FDA Commissioner's Initiative). This is aimed at speeding up the testing and approval process. For compounds that justifiably fall into this category, FDA-CDER personnel, of necessity, become involved in the pre-IND planning of clinical testing via face-to-face meetings between the sponsor and the FDA.

Phase I clinical testing involves the demonstration of the safety of single and then multidose (if relevant) administration of the compound in healthy volunteers by the intended route of administration. Allowable clinical doses are initially held to one tenth the dose level that has been previously shown to be maximally tolerable in animal toxicology studies. If Phase I testing fails to turn up any toxicities that would preclude subsequent testing in patients, the compound then moves into Phase II, in which safety and efficacy is tested within the intended patient population. Commonly, Phase II trials are subdivided into Phase IIa and IIb. Phase IIa studies are initially carried out to establish safety in a small number of patients, and typically involve a carefully monitored open label, dose-escalation approach. These are followed by Phase IIb studies, in which further safety, clinical pharmacology, and initial therapeutic efficacy determinations are made in a larger number of patients. Phase IIb studies may be conducted in an open label or a blinded fashion. If at the end of Phase II it is determined that the compound is safe for wider patient testing and appears to show the predicted efficacy, the compound can then graduate into Phase III clinical trials that are randomized, controlled with either a placebo or an active, already approved, control drug, and double blind. The ultimate purpose is to determine whether the compound produces a statistically significant therapeutic effect without any toxicities that severely limit its use in the effective dose range. In the case of compounds designated as fast-track, it is expected that the sponsor and the FDA will keep in close contact, and in essence, collaborate in the design of

the development process and the interpretation of results. To ensure this, face-to-face meetings take place before and after each phase.

Subpart E also allows for the clinical development phases to be partially overlapped (i.e., Phase IIa studies can begin before the completion of the later, multi-dose Phase I trials), in order to further expedite the development of promising agents. Furthermore, the FDA regulations allow for the filing of an Emergency Use IND, which allows the use of the experimental drug in an emergency situation involving patients who do not meet the criteria of existing study protocols. Alternatively, a Treatment IND can be submitted to request permission for limited usage of experimental drugs that show promise in Phase II clinical testing for serious or immediately life-threatening conditions while controlled clinical testing is conducted and the FDA review takes place.

The approval of a drug for full marketing is based on the demonstration in Phase III clinical trials of a statistically significant therapeutic effect in the absence of unacceptable toxicities. During these trials, safety and efficacy are monitored by an independent safety monitoring committee, which can vote to stop the trial before completion if one of the groups is showing either unpredicted morbidity or mortality or a remarkable therapeutic response. Moreover, the FDA typically requires the successful completion of at least two similar and well monitored Phase III trials that both demonstrate "substantial evidence" of efficacy (i.e., $p < 0.05$, comparing untreated and drug-treated outcomes). This requirement, however, has been slightly liberalized by the Food and Drug Administration Modernization Act of 1997, which states that "if the secretary (Health and Human Resources) determines, based upon relevant science, that data from one adequate and well-controlled clinical investigation and confirmatory evidence (obtained prior to or after such investigation) are sufficient to establish effectiveness, the Secretary may consider such data and evidence to constitute substantial evidence" required for FDA approval. An example of how this might work is the coupling of a single successful Phase III trial with an earlier Phase IIb trial that tended strongly to show therapeutic efficacy. In order for the Phase III trial to be usable, it would almost certainly have to have been conducted in a randomized, controlled, and double blinded fashion.

At the end of Phase III, the sponsor files a New Drug Application (NDA) with the FDA that encompasses, in excruciating detail, the experimental drug's life story from initial chemical synthesis through completion of Phase III trials. After a laborious review, the FDA will approve the compound if they reach the determination that (1) the drug is safe and effective in its proposed use(s), and the benefits outweigh the risks; (2) the drug's proposed labeling (i.e., package insert) is appropriate; and (3) that the methods used in manufacturing the drug are adequate to preserve the drug's identity, strength, quality, and purity. Although the legal approval of the drug is strictly within the FDA's authority, the FDA typically employs an Advisory Committee, consisting of independent experts knowledgeable in the field of the drug's intended use, to make a recommendation concerning the approvability of the drug for marketing. Approval of a drug without a positive (majority) recommendation of the Advisory Committee, although possible, is highly unlikely.

## DEVELOPMENT OF HIGH-DOSE METHYLPREDNISOLONE AND TIRILAZAD FOR NEUROPROTECTION IN ACUTE SCI

Although the secondary injury process in acute SCI is biochemically and physiologically complex, as shown in **Figure 1**, cell membrane (plasma and organellar) lipid peroxidation (LP) has been conclusively demonstrated to be a key mechanism. LP, which is post-traumatically initiated by highly reactive oxygen species (ROS), damages both microvascular and parenchymal cell membranes. Within the context of the overall secondary injury cascade, the process of LP is a consequence of post-traumatic

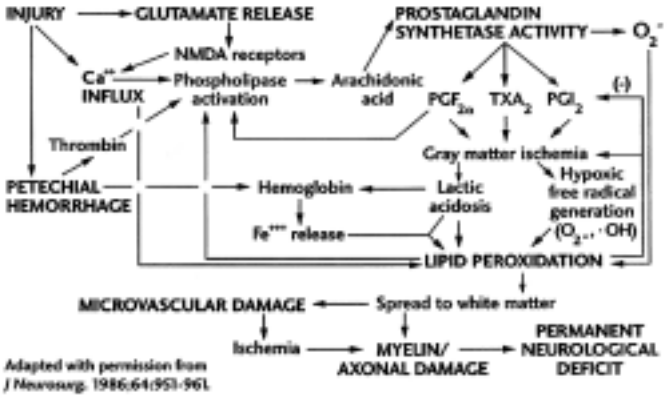


**Figure 1.**
Secondary injury cascade after acute spinal cord injury.

glutamate release, activation of the arachidonic acid cascade, and the production of prostaglandins, resulting in vasoconstriction and microembolism and the formation of oxygen radicals that initiate LP [1–4]. Iron is a powerful catalyst that accelerates the propagation of LP reactions. Anaerobically derived lactate promotes LP by stimulating the release of iron from storage sites; e.g., ferritin. In addition, primary and secondary petechial hemorrhages supply hemoglobin-bound iron. In addition, dysfunctional mitochondria appear to be an important source of ROS within the injured cord. LP occurs in neurons and blood vessels, directly impairing neuronal and axonal membrane function, and causing microvascular damage and secondary ischemia that indirectly contributes to the secondary injury to neurons and axons.

## High-Dose Methylprednisolone Inhibition of Lipid Peroxidation

Knowledge of this mechanism prompted the development of neuroprotective pharmacologic strategies aimed at antagonizing oxygen radical-induced LP in a safe and effective manner. Beginning in the early 1970s, attention was focused on the hypothetical possibility that glucocorticoid steroids might be effective inhibitors of post-traumatic LP, based on their typically high lipid solubility and known ability to intercalate into artificial membranes between the hydrophobic polyunsaturated fatty acids of the membrane phospholipids and to thereby limit the propagation of LP chain reactions throughout the phospholipids bilayer [5,6]. Interest in the glucocorticoid steroids per se was also enhanced by their already widespread empirical use to treat SCI and traumatic brain injury, based on the notion that they would attenuate post-traumatic central nervous system (CNS) edema. This notion was born out of the rather profound effects that glucocorticoid steroids had displayed in suppressing peritumoral edema.

My colleagues and I became interested in the LP hypothesis of secondary SCI during our parallel investigations of the effects of high-dose methylprednisolone (MP) (15–90 mg/kg, i.v.) on spinal cord electrophysiology, as those might serve to improve impulse conduction and recovery of function in the injured spinal cord [7]. Consequently, we decided to test the possibility that a similar high dose of MP that enhanced spinal neuronal excitability and impulse transmission might also be required to inhibit LP in vivo. In an initial set of experiments in cats, we demonstrated that the administration of an i.v. bolus of MP could indeed inhibit post-traumatic LP in spinal cord tissue [8], but that the doses required for this effect were much higher (30 mg/kg) than previously hypothesized or than those empirically employed in the acute treatment of clinical CNS injury. Further experimental studies also conducted in cat SCI models showed that at the 30 mg/kg dose of MP not only prevented LP, but in parallel inhibited post-traumatic spinal cord ischemia [9,10], supported aerobic energy metabolism (i.e., reduced lactate and improved adenosine triphosphate (ATP) and energy charge) [11–13], improved recovery of extracellular calcium (i.e., reduced intracellular overload) [9], and attenuated calpain-mediated neurofilament loss [13]. However, the central effect in this protective scenario is the inhibition of post-traumatic LP (**Figure 2**). With many of these therapeutic parameters (LP, secondary ischemia, aerobic energy metabolism), the dose-response for the MP follows a sharp U-shaped pattern. The neuro- and vaso-protective effect is partial with a dose of 15 mg/kg, is optimal at 30 mg/kg, and diminishes at higher doses (60 mg/kg) [6].

The neuroprotective action of MP is closely linked to the drug's tissue pharmacokinetics [6,12,14,15]. For instance, when MP tissue levels are at their peak following administration of a 30 mg/kg i.v. dose, lactate levels in the injured cord are suppressed. When tissue MP levels decline, spinal tissue lactate rises. However, the administration of a second dose (15 mg/kg, i.v.) at the point at which the levels after the first dose have declined by 50 percent, acts to maintain the suppression of lactate seen at the peak of the first dose [12]. This prompted the hypothesis that prolonged MP therapy might better

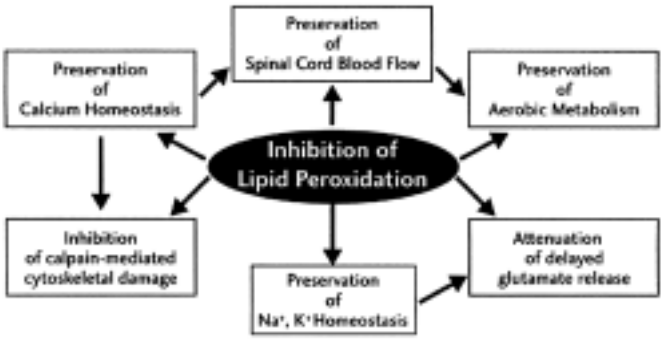


**Figure 2.**
Neuroprotective actions of high-dose methylprednisolone therapy in injured spinal cord showing that central mechanism is inhibition of reactive oxygen-induced lipid peroxidation.

suppress the secondary injury process and lead to better outcomes compared to the effects of a single large initial intraveneous dose. Indeed, subsequent experiments in a cat spinal injury model demonstrated that animals treated with MP using a 48-hour antioxidant dosing regimen had improved recovery of motor function over a 4-week period [16,17].

## NASCIS II

These studies successfully inspired the Second National Acute Spinal Cord Injury Study (NASCIS II) [18], even though an earlier NASCIS trial, which came to be known as NASCIS I, failed to show any efficacy of lower MP doses, even when administered over a 10-day period [19,20]. The NASCIS II trial compared MP, naloxone, and placebo for the treatment of acute SCI. A priori-trial hypotheses included the prediction that SCI patients treated within the first 8 postinjury hours would respond better to pharmacotherapy than patients treated after 8 hours. Indeed, the results demonstrated the effectiveness of 24 hours of intensive MP dosing (30 mg/kg, i.v. bolus, plus a 23-hour infusion at 5 mg/kg per hour) when treatment was initiated within 8 hours. Significant benefit was observed in individuals with both neurologically complete and incomplete injuries. Moreover, the functional benefits were sustained at 6-week, 6-month, and 1-year follow-up [18,21–23]. Although predictable side effects of steroid therapy were noted, including gastrointestinal bleeding, wound infections, and delayed healing, these were not statistically significant with regard to frequency compared to placebo-treated patients [18].

## Discovery of Tirilazad

MP is a potent glucocorticoid that possesses a number of glucocorticoid receptor-mediated anti-inflammatory actions. The principal mechanism of the neuroprotective action for MP in the injured spinal cord was determined to be inhibiting post-traumatic LP, rather than it being mediated via glucocorticoid receptor-mediated activity [24–26]. This prompted our speculation that modifying the steroid molecule to enhance the anti-LP effect, while eliminating the steroid's glucocorticoid-related effects, would result in more targeted therapy devoid of the typical side effects of steroid therapy. This avenue led to the development of more potent inhibitors of LP, i.e., the 21-aminosteroids or “lazaroids,” which lack the glucocorticoid side effects that limit the clinical utility of high-dose MP. One of these, tirilazad, was selected for development.

**Figure 3** compares the structures of the glucocorticoid, MP, and the nonglucocorticoid 21-aminosteroid, tirilazad.

## NASCIS III

The demonstrated efficacy of a 24-hour dosing regimen of MP in human SCI in NASCIS II [18], and the discovery of tirilazad [24–26], led to another trial of these agents in human SCI, NASCIS III [27,28]. In the NASCIS III trial, 3 groups of patients were evaluated. The first (active control) group was treated with the 24-hour MP dosing regimen that previously had been shown to be effective in NASCIS II. The second group was also treated with MP, except that the duration of MP infusion was extended to 48 hours. The rationale for this regimen was to determine whether extension of the MP infusion from 24 to 48 hours resulted in greater improvement in neurological recovery in acute SCI patients. The third group of patients was treated with a single 30 mg/kg i.v. bolus of MP, followed by the 48-hour administration of tirilazad. No placebo group was included because it was deemed ethically inappropriate to withhold at least the initial large bolus of MP. Another objective of the study

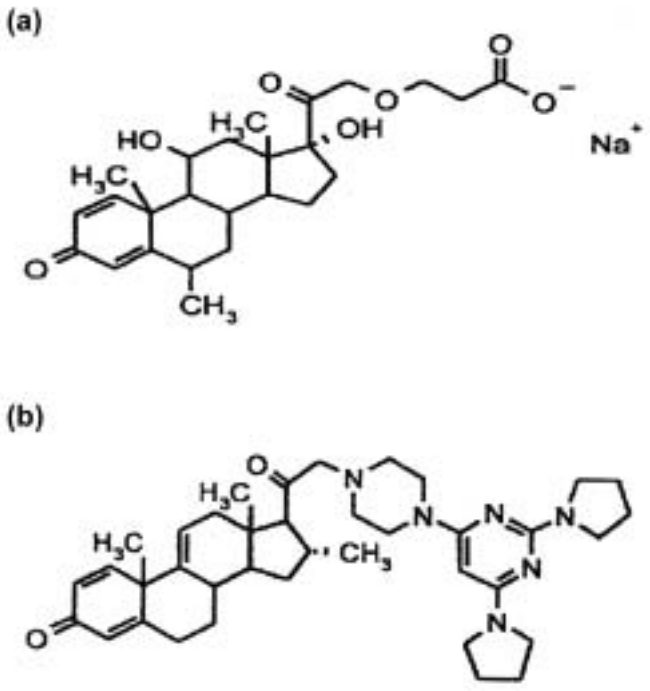


**Figure 3.**
Chemical structures of **(a)** glucocorticoid steroid methylprednisolone (shown as sodium salt of 21-hemisuccinate ester) and **(b)** nonglucocorticoid 21-aminosteroid (lazaroid) tirilazad mesylate.

was to ascertain whether treatment initiation within 3 hours following injury was more effective than when therapy was delayed until 3 to 8 hours post-SCI.

Upon completion of the NASCIS III trial, it was found that all 3 treatment arms produced comparable degrees of recovery when treatment with each was begun within the shorter, 3-hour window. When the 24-hour dosing of MP was begun more than 3 hours post-SCI, recovery was poorer in comparison to the cohort treated within 3 hours following SCI. However, in the 3- to 8-hour post-SCI cohort, when MP dosing was extended to 48 hours, significantly better recovery was observed. In the comparable tirilazad cohort (3–8 hours post-SCI), recovery was slightly, but not significantly, better than in the 24-hour MP group, and poorer than in the 48-hour MP group. These results showed that (1) initiation of treatment within the first 3 hours is optimal, (2) the nonglucocorticoid tirilazad is as effective as 24-hour MP therapy, and (3) if treatment is initiated more than 3 hours post-SCI, extension of the MP dosing regimen is indicated, from 24 to 48 hours. However, in comparison with the 24-hour dosing regimen, significantly more glucocorticoid-related immunosuppression-based side effects were seen with more prolonged dosing; i.e., the incidence of severe sepsis and pneumonia significantly increased. In contrast, tirilazad showed no evidence of steroid-related side effects, suggesting that this nonglucocorticoid 21-aminosteroid would be safer for extension of dosing beyond the 48-hour limit used in NASCIS III [27,28].

## REGULATORY STATUS OF MP AND TIRILAZAD FOR SCI

Because MP had already been successfully developed, approved by the FDA, and marketed over several years for a wide variety of anti-inflammatory conditions, the clinical testing of MP in both NASCIS I and II was much easier than the typical scenario of new drug development. MP in different oral, intramuscular, and intravenous formulations had been approved in the early 1960s and was actually off patent in the U.S. by the time of the NASCIS trials. Furthermore, with regard to the testing of high or mega-doses of the steroid in SCI patients, there was already considerable clinical experience with the intraveneous administration of doses as high, or higher, than 30 mg/kg in several clinical studies concerned with the potential use of MP in various critical-care indications. Thus, the safety of this high-dose treatment for a short period had already been established, even in severely compromised patients. Therefore, the approval of the IND for testing it in human SCI did not pose a significant hurdle. Moreover, the trials were not initiated or controlled by the drug's original sponsor (The Upjohn Company), but rather by the NASCIS group headed by Dr. Michael Bracken (Professor of Epidemiology and Public Health at Yale University). Although Upjohn provided the supplies of their already marketed MP formulation (Solu Medrol) and the aqueous vehicle (placebo) in support of NASCIS I and II, the trials were funded solely with grant support from the National Institutes of Health (NIH), National Institute of Neurological Disorders and Stroke (NINDS). NASCIS III was also NIH-supported, although Upjohn, in addition to providing MP at no cost, also shouldered some of the monitoring costs relevant to tirilazad, which was still under premarketing clinical development. However, the clinical data analysis was carried out at Yale University, completely independent of The Upjohn Company.

Following the demonstration of the efficacy of the 24-hour MP dosing regimen in NASCIS II, Upjohn successfully achieved registration of the drug in the early 1990s for use in acute SCI in Canada, several western European countries, and most Far Eastern countries, where the drug was already marketed for anti-inflammatory uses. However, because of the U.S. FDA requirement for two well controlled clinical trials that both demonstrate substantial evidence of efficacy, the submission of an NDA for the use of MP in SCI was not possible on the strength of NASCIS II alone. Nevertheless, because MP was already marketed in the U.S. for several therapeutic indications, its extensive use in human SCI (albeit unapproved) was possible, even though The Upjohn Company could not promote it for the SCI indication. Furthermore, because SCI represented an unmet medical need, the 24-hour NASCIS II MP dosing protocol quickly became the standard of care for human SCI in the U.S. as well as in other countries in which it was registered for SCI.

Subsequent to NASCIS II, two other groups of investigators in Japan [29] and France [30] reported replications of the therapeutic efficacy of the NASCIS II MP protocol in SCI patients. However, Upjohn, after becoming Pharmacia & Upjohn in 1995, still elected not to file an NDA, even though the requirement for two successful trials had presumably been met. This decision not to seek U.S. registration for acute SCI was based, first of all, on

the fact that the drug was already widely used in SCI in the U.S. in the absence of registration for that particular indication. Secondly, the difficulties in successfully gathering the necessary clinical data from non-company, offshore investigators made the pulling together of required NDA documents exceedingly difficult. Thirdly, as recently reviewed by Bracken [31], there was reason to believe that these non-U.S. replications may not have been conducted in a rigorous enough manner to support FDA approval. For instance, in the Japanese trial [29], there was a differential loss to follow up between the untreated and MP-treated patients; and, it was not clear that the French MP SCI trial was carried out in a fully blinded manner [30].

From a regulatory standpoint, the NASCIS III trial did little to facilitate the potential filing of an MP for SCI NDA in the U.S., since it was not a placebo-controlled trial. In designing NASCIS III, the NASCIS clinical investigators concluded, on the basis of NASCIS II results showing significant efficacy of 24-hour, high-dose MP in comparison to placebo-treated patients, that it was no longer ethical to withhold high-dose MP from SCI victims. Consequently, NASCIS III [27] became not only a comparison of 24-hour versus 48-hour MP dosing, but also a simultaneous comparison of the neurological outcome of patients treated with a single i.v. bolus of MP followed by 48 hours of tirilazad dosing. Although the results showed that patients treated after 3 hours postinjury did significantly better when dosed with MP for 48 hours in comparison to only 24 hours, the absence of a placebo left the placebo-controlled results of NASCIS II unconfirmed, except by the Japanese and French groups, whose trials may not have been as rigorously controlled as the NASCIS trials [31].

In the case of the MP bolus plus tirilazad group, those patients recovered as well as the 24- and 48-hour MP-treated patients when treatment was initiated within the first 3 hours, and in between the 24- and 48-hour MP groups in the 3- to 8-hour treatment cohort. Nevertheless, even though these NASCIS III results suggest that the nonglucocorticoid steroid tirilazad may duplicate MP's neuroprotective efficacy without the same side effects, the ultimate approval of this compound for SCI in humans would require at least another trial comparing it against placebo in order to have any hope of becoming registered by the FDA. Furthermore, a registration-worthy Phase III trial of MP versus tirilazad is precluded by the fact that the comparator drug MP is not registered for SCI in the U.S; Phase III clinical trials destined for inclusion in NDAs cannot be conducted with one unapproved drug being compared to a second unapproved drug. Consequently, a scenario in which tirilazad could be successfully approved for use in SCI in the U.S. is not apparent, unless it were deemed to be ethically appropriate to test it against placebo (i.e., without MP).

## FUTURE SCI DRUG DEVELOPMENT: PROBABLE NEED TO BUILD ON HIGH-DOSE MP THERAPY

At present, high-dose MP therapy, although not officially approved in the U.S. for acute SCI treatment, continues to be the standard of care in the U.S. for many neurosurgeons. This is also the case in most countries where MP is marketed. However, the use of high-dose MP in acute SCI is controversial. Some neurosurgical clinicians and researchers believe that the risks outweigh what they feel are, on average, modest neurological benefits [32–34]. Although many appear to believe in the neuroprotective efficacy of MP, there is little doubt that many use it in their patients on a defensive basis to prevent possible malpractice litigation were they not to use the treatment. In any event, it is likely that future trials of new drug treatments will have to be evaluated on top of high-dose MP because, apparently, most clinicians who are faced with treating acute SCI victims are not prepared to withhold MP from their patients. The only situation in which this may be avoidable will be when preclinical studies have shown that the neuroprotective effects of MP, and/or the second drug, are offset when the two are used simultaneously or when there is a documented dangerous interaction of the two drugs. For instance, it has been reported that the coincident use of MP and the monosialo-ganglioside (GM1) in a preclinical SCI model resulted in an attenuation of MP's neuroprotective efficacy and an increase in post-SCI mortality [35]. Thus, a recently completed Phase III trial of GM1 in SCI patients was successfully carried out that involved administration of GM1 beginning after the completion of the NASCIS II 24-hour MP dosing protocol [36–38]. The results of that trial indicated that use of GM1 after MP therapy resulted in a faster achievement of peak neurological recovery, although the extent of neurological recovery was not greater than that in patients who only received MP.

This future development scenario involving the necessary design of clinical trials in which a second neuroprotective (or neurorestorative) drug is administered at the same time as, or in series with, high-dose MP, raises a significant dilemma for both the sponsor of the new agent and the FDA. From the perspective of the sponsor, there is the need not only to define the neurological effects in both animals and humans of the combination of the new compound with and without MP therapy, but also to carefully study toxicological and pharmacokinetic interactions of the two agents. From the vantage point of the FDA, the problem will be evaluating the effects of the combination of MP plus a second drug against MP, without ever having made the determination that MP, by itself, is better than placebo treatment. In other words, they will be evaluating one experimental drug against a combination of two experimental drugs. If there is any precedent for this regulatory situation, it will probably have to come from cancer chemotherapy drug review, where combination treatments are typically the norm.

## SOME NEW NEUROPROTECTIVE OR NEURORESTORATIVE APPROACHES

### Novel Scavengers of ROS

In view of the clear role of reactive oxygen or oxygen radical-induced LP in the pathophysiology of post-traumatic spinal cord degeneration, and the demonstrated benefits of antioxidant compounds with neuroprotective activity such as MP and tirilazad, it is logical to pursue the development of novel antioxidant compounds. Recent work suggests that the most critical ROS in acute SCI may be peroxynitrite, which is formed from the combination of superoxide and nitric oxide radicals [39,40]. Peroxynitrite is capable of causing widespread damage to lipids, proteins, and nucleic acids. Prototypical scavengers of peroxynitrite include penicillamine and Tempol, both of which are neuroprotective in cell culture and in vivo models of acute CNS injury [41,42].

### Dual Inhibition of LP and Peroxynitrite

A third antioxidant-based approach that shows promise concerns dual inhibition of LP and neuronal nitric oxide synthase (an enzyme that contributes to the production of peroxynitrite). Such a dual-inhibitor compound, BN-80933, has been reported to attenuate post-traumatic and postischemic degeneration in in vivo models [43]. In comparison with a neuronal nitric oxide synthase inhibitor alone, BN-80933 has been shown to have superior neuroprotective efficacy.

### Combination Therapy

A fourth approach would be to try combination therapy of antioxidant therapy and agents with complementary mechanisms of actions. Some of the logical candidates for combination with MP, tirilazad, or other antioxidants include calpain inhibitors [44,45], antiapoptotic compounds [46,47], anti-inflammatory agents [48], and the beta-2 agonist, clenbuterol [49]. Another possibility would be to combine a neuroprotective agent with a neurorestorative agent. Neurorestorative agents enhance the inherent plasticity of surviving neurons. A number of agents have been identified—e.g., OP-1 (osteogenic protein 1), which stimulates dendritic branching and growth [50]; neuroimmunophilins, such as FK506 [51,52] and V-10,367 [53,54], which stimulate axonal sprouting and inhibitors of the myelin-derived Nogo protein, which acts to inhibit axonal growth [55]. Neuroprotective and neurorestorative agents also have potential applications in cell transplantation, where they could serve to protect transplanted cells and promote their differentiation and growth.

## CONCLUSIONS

To move a new pharmacological neuroprotective and/or neurorestorative treatment for SCI from animals to humans (i.e., from preclinical to Phase I), the FDA will fundamentally require (1) evidence of neurological efficacy in acute SCI animal models with and without concomitant MP treatment (although data from a nonrodent SCI model would be good, it is unlikely that it will be required to move to humans); (2) preclinical pharmacokinetics of the new compound by itself and in combination with MP and correlation of therapeutic blood or spinal cord levels with neuroprotective/neurorestorative efficacy; and (3) preclinical pathology/toxicology and toxicokinetics in two species (e.g., rat and dog, rat and monkey), looking at the new compound alone and in combination with MP.

In order to move from Phase I to Phase II, and from Phase II to Phase III, the FDA will expect (1) involvement of FDA personnel in trial design for each phase (assuming accelerated drug development has been agreed

upon); and (2) careful pharmacokinetic and safety assessment in each phase. However, apart from safety concerns, the decision to go to large, and expensive, Phase III trials in the face of equivocal evidence of efficacy in Phase IIb will depend on the sponsor's willingness to take the risk. The FDA will not stop the sponsor unless there are toxicology considerations that preclude further development.

Ultimately, to gain registration of a new compound for marketing approval in humans, the sponsor will have to provide unequivocal evidence of neurological benefit in double-blinded Phase III clinical trials, which will mean that a *p* value of 0.05 (double-tailed) has been achieved on top of Bonferroni or other corrections for multiple comparisons (i.e., placebo or MP vs. different doses of the compound under development). As noted above, the FDA may or may not require the completion of two Phase III trials. In any event, the primary efficacy endpoint will have to be an index of functional recovery improvement. Although surrogate endpoints such as MRI imaging showing sparing of spinal cord tissue or other biochemical markers of neuroprotective or neurorestorative effects will be helpful, they are currently not viewed as sufficient by themselves to drive FDA marketing approval. Furthermore, the neurological benefits will have to outweigh the risks (i.e., side-effect potential) of the therapy, whether it is administered alone or in combination with MP.

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