Neurological outcome correlated with spinal evoked potentials in a spinal cord ischemia model

MAN-KAI CHENG, M.D., CLAUDIA ROBERTSON, M.D., ROBERT G. GROSSMAN, M.D., RICHARD FOLTZ, B.A., AND VICK WILLIAMS, M.D., PH.D.

Department of Neurosurgery, Baylor College of Medicine, Houston, and Department of Anatomy, University of Texas Health Science Center at San Antonio, San Antonio, Texas

Occlusion of the abdominal aorta of the rabbit by inflating the balloon of a Swan-Ganz catheter positioned in the aorta is a simple and reliable method of producing spinal cord ischemia. The electrophysiological, neurological, and neuropathological correlates of ischemia with progressively longer durations and of ischemia after drug interventions were studied with the goal of developing an easily monitored, reproducible model for central nervous system ischemia. The percentage of animals developing paraplegia after varying periods of ischemia was zero after 15 minutes, 30% after 17 minutes, 33% after 20 minutes, 38% after 25 minutes, and 100% after 60 minutes of ischemia. After 25 minutes of ischemia the percentage of animals developing paraplegia was 87% when they were awake and not ventilated during ischemia and reperfusion, but dropped to 38% in animals that were paralyzed, sedated with ketamine, and ventilated, and when the metabolic acidosis that follows aortic occlusion was corrected during reperfusion. Pretreatment with thiopental, hypothermia, naloxone, methylprednisolone, and verapamil changed the percentage of animals developing paraplegia after 25 minutes of ischemia to 0%, 0%, 25%, 40%, and 100%, respectively. The component waves of the spinal somatosensory evoked potential (SSEP) disappeared sequentially during ischemia in the following order: P2, N4, N3, N2, and N1. After reperfusion, the SSEP components returned in reverse order of their disappearance. In the untreated animals, absence of the N3 wave for more than 10 minutes during ischemia was always followed by a neurological deficit. Pretreatment with thiopental or hypothermia permitted longer periods of electrophysiological silence without permanent neurological deficit. The ratio of the amplitude of N3 to N1 (N3/N1) was at least 70% of the control level, and N4 and P2 amplitudes were at least 30% of the control level at 120 minutes after reperfusion in all animals that had a normal outcome. Return of the N3/N1 amplitude to at least 90% of the control level or return of N3/N1 to 70% to 89% of control and P2 to at least 60% of control at 120 minutes after reperfusion reliably correlated with a normal 48-hour motor examination in animals with and without drug interventions.

KEY WORDS spinal cord □ spinal somatosensory evoked potentials □ ischemia □ infarction □ paraplegia

Occlusion of the abdominal aorta has been used for many years as a method of studying ischemia of the spinal cord. Over 200 years ago, Stenonis described hindlimb paralysis in a dog after ligation of the descending aorta which resolved when the ligature was loosened. Methods of producing occlusion of the aorta have included external mechanical clamps that compress the aorta against the vertebral column, clamping or clipping the aorta through an abdominal incision, and inflating a balloon catheter positioned in the aorta. Because the blood supply to the spinal cord of the rabbit originates from segmental arterial branches of the aorta, reproducible lesions can be caused by obstructing blood flow at a given level in the aorta. Easily recognizable neurological deficits result, and the systemic effects of producing the lesion are not so severe as to prevent survival of most animals. However, there are systemic changes with this method which might affect the neurological outcome. The lower extremities and the large bowel are also supplied by the distal aorta and become ischemic with the spinal cord. When the balloon is deflated, the lactate produced by the ischemic tissues is released into the systemic circulation. The hypotension and the hypocarbia that result from the metabolic acidosis may exacerbate the injury during the reperfusion phase.

Since the neurological deficits evolve with time, the
Outcome and evoked potentials in spinal ischemia

animals must be observed for several days to determine neurological outcome. Animals that become paraplegic, however, require special care, and are more likely to die postoperatively than are normal animals. This skews outcome results if only surviving animals are counted, and requires that more animals be studied. A method of reliably predicting neurological outcome at the time the ischemia is produced would reduce the time and cost of this method of studying the pathophysiology of ischemia of the central nervous system.

Evoked potentials of the spinal cord may give information as to whether or not a population of sensory neurons, motor neurons, or interneurons can survive a certain period of ischemia by demonstrating how long during ischemia they remain functional. Although spinal somatosensory evoked potentials (SSEP's) do not give direct information about the function of motor neurons, during the global ischemia produced by complete aortic occlusion the survival of motor neurons might correlate with survival of the dorsal interneurons.

The purpose of our present study was to refine a classical model of spinal cord ischemia to include the following features: 1) rapid, simple preparation using a relatively noninvasive balloon catheter occlusion; 2) the ability to control or at least measure systemic factors that might contribute to ischemic injury; and 3) the capability of predicting neurological outcome at the time of the study by monitoring SSEP recovery.

Materials and Methods

Occlusion Protocol

Sixty-seven male New Zealand albino rabbits, each weighing 3.5 to 4.5 kg (mean 4.2 kg), were anesthetized with methohexital sodium (Brevital), up to 10 mg/kg total dose, during the surgical preparation. A No. 5 French pediatric Swan-Ganz catheter* was modified so that pressure-monitoring ports lay proximal and distal to the balloon, and was inserted through the right femoral artery into the abdominal aorta. Under fluoroscopic arteriography, the catheter was positioned so that the tip was just distal to the origin of the left renal artery. Figure 1 shows the usual catheter position in the abdominal aorta and demonstrates the segmental arterial branches supplying the spinal cord; in the rabbit, there is no major artery supplying the lumbosacral cord more proximal than the renal artery.

After preparation as described below, ischemia of the spinal cord was produced by inflating the balloon with 0.4 ml of air. The port distal to the balloon was used for drawing arterial blood gases and monitoring systemic blood pressure, and the port proximal to the balloon was used to monitor pressure in the caudal abdominal aorta to demonstrate that the occlusion remained complete. After the desired period of ischemia, the balloon was deflated and the catheter withdrawn so that only 2 to 3 cm of the catheter remained in the femoral artery to minimize the risk of thrombosing the abdominal aorta. Heparin, 500 units, was given intravenously prior to femoral catheterization, and 100 to 150 units were given intravenously each hour as long as the catheter remained in place.

After awakening from anesthesia, the animals were returned to their cages. They were given fluids by intraperitoneal injection until they were able to drink adequately, and cephalothin (Keflin) was administered intramuscularly in a dose of 25 mg/kg every 12 hours. The Credé maneuver was used to empty the bladders of the paraplegic animals at least twice daily.

After the final neurological examination and/or recording of SSEP's, the rabbits were sacrificed by left cardiac perfusion with a buffered solution of 4% paraformaldehyde and 0.5% glutaraldehyde. The spinal cord from T-5 to S-5 was removed en bloc and placed in a phosphate-buffered solution for neuropathological studies. At necropsy, the abdominal aorta was checked for patency.

Experimental Groups

Three experimental designs were used to study the factors responsible for producing paraplegia and to correlate electrophysiological, neurological, and neuropathological findings.

---

* Swan-Ganz catheter, No. 702215, manufactured by Edwards Laboratories, 17221 Red Hill Avenue, Santa Ana, California.
**Group 1.** In Group 1 animals, the SSEP's were compared to neurological outcome following spinal cord ischemia of varying time intervals. In this group, 31 of the rabbits were intubated and ventilated with a Harvard rodent respirator† with supplemental oxygen, and with the tidal volume and rate adjusted to maintain normal arterial blood gases. The mean blood gas values were: pO₂ 119 ± 43 torr, pCO₂ 32 ± 5 torr, and pH 7.39 ± 0.04. Anesthesia was maintained with intramuscular ketamine, 44 mg/kg, followed by 20 mg/kg/hr intramuscularly. Rectal temperature was kept at normal levels (38.6° ± 0.5°C) using a heating lamp.

Both sciatic nerves were exposed just proximal to their bifurcations, and cuff bipolar silver stimulating electrodes were placed around each nerve. The muscle-twitch threshold was in all cases less than 0.05 mA at a pulse duration of 0.1 msec. Three needle electrodes‡ were inserted into the midline interspinous ligament so that they were in contact with the lamina at the L3-4, L4-5, and L5-6 levels.

A 1-mg dose of pancuronium bromide (Pavulon), administered intravenously, followed by supplemental doses as needed, was given to maintain muscle paralysis during recording of SSEP's to prevent recording of the muscle-twitch artifact evoked by sciatic nerve stimulation. The stimuli used were square-wave pulses of 0.1-msec duration and 0.5-mA intensity delivered at 3.1 Hz.

The SSEP's were recorded in a bipolar fashion from the L5-6 to the L4-5 interspaces, and from the L4-5 to the L3-4 interspaces. The potentials were displayed so that a negative potential (N) at L5-6 with respect to L4-5, or at L4-5 with respect to L3-4, was an upgoing deflection (see Fig. 2). Recordings were made with a Nicolet signal averager.§ The signal was amplified 30,000 times by amplifiers with a bandwidth of 5 Hz to 3 kHz (low and high bandpass, respectively), and 50 repetitions were averaged.

Ischemia of the spinal cord was produced by inflation of the balloon of the Swan-Ganz catheter for 15, 17, 20, 25, or 60 minutes. A mean dose of 3 ± 2 mEq sodium bicarbonate was given intravenously to correct the metabolic acidosis that occurred during reperfusion. The SSEP's were recorded prior to spinal cord ischemia, at intervals during ischemia, and for 120 minutes after reperfusion. Neurological examinations were performed daily, and the SSEP recordings were repeated at 48 hours and at 7 days.

To compare the SSEP's recorded during ischemia and reperfusion to the control SSEP's, the amplitude and latency of each component wave was measured. In addition, to compensate for small differences in the position of the recording electrodes in different animals, and between the first and subsequent recordings in the same animal, the ratio of the amplitude of N₃ to N₁ (N₃/N₁) was calculated. The information obtained from these Group 1 rabbits was used to establish the normal amplitudes and latencies of the SSEP components, the normal response of the SSEP's to ischemia and reperfusion, and the criteria for prediction of neurological outcome from SSEP recordings.

**Group 2.** In Group 2, neurological outcome after 25 minutes of spinal cord ischemia in awake rabbits was determined. The 15 rabbits in this group were allowed to awaken from anesthesia after placement of the Swan-Ganz catheter, and then underwent ischemia of the spinal cord by inflation of the balloon of the catheter for 25 minutes. Neurological examinations were performed daily for 7 days.

**Group 3.** In the Group 3 rabbits, the SSEP's were correlated with the neurological outcome following 25 minutes of spinal cord ischemia and drug administration. The 21 rabbits in this group were intubated and ventilated with a Harvard rodent respirator with supplemental oxygen, and with the tidal volume and rate adjusted to maintain a normal pO₂, pCO₂, and pH. Anesthesia was maintained with intramuscular ketamine, 44 mg/kg, followed by 20 mg/kg/hr intramuscularly.

The left sciatic nerve was exposed just proximal to its bifurcation, and a cuff bipolar silver stimulating electrode was placed around the nerve. Two Teflon-coated silver wire electrodes were implanted at L5-6 and L4-5 and sutured in place so that, for recordings

---

† Harvard rodent respirator manufactured by Harvard Apparatus, Inc., 150 Dover Road, Millis, Massachusetts.
§ Nicolet signal averager manufactured by Nicolet Instrument Corp., 5225 Verona Road, Madison, Wisconsin.

**Fig. 2.** Characteristic rabbit spinal somatosensory evoked potentials elicited by sciatic nerve stimulation and recorded at L5-6, with reference to L4-5. Negativity (N to N₁) is indicated by upward deflection. The inflection points used for measuring the amplitudes and latencies of the component waves are illustrated by arrows.
on future days, the electrode position would remain constant. Pancuronium bromide was given to maintain muscle paralysis during SSEP recordings. The stimuli and recording parameters were otherwise as described in Group I.

The rabbits received one of five treatments: intravenous administration of either thiopental, 30 mg/kg; methylprednisolone, 30 mg/kg; verapamil, 0.3 mg/kg, each for 30 minutes before ischemia; naloxone, with a 10-mg dose followed by 1 mg/min during ischemia and for 90 minutes after reperfusion; or surface cooling to a rectal temperature of 30°C. Ischemia was then produced by inflation of the balloon of the Swan-Ganz catheter for 25 minutes. Sodium bicarbonate was given intravenously to correct the metabolic acidosis that occurred during reperfusion. The SSEP’s were recorded prior to spinal cord ischemia, at intervals during ischemia, and for 120 minutes after reperfusion. Neurological examinations were performed daily, and the SSEP recordings were repeated at 48 hours.

The results in this group of animals were used to confirm that the SSEP criteria for predicting neurological outcome which was developed with the Group 1 rabbits, remained reliable when drugs were administered.

Results

The Normal SSEP Pattern

The morphology of the SSEP’s recorded from L5–6 and L4–5 was virtually identical in all animals. A typical recording at L5–6 is shown in Fig. 2, which demonstrates the component waves P1, N1 to N4, and P2. Measurement of the wave amplitudes from the pre-stimulus baseline value gave the clearest description of the behavior of the waves during ischemia and reperfusion. The means and standard deviations of these baseline amplitudes and the latencies of N1 to N4 and P2 in the 31 Group 1 rabbits are also shown in Fig. 2. The mean value of the N3/N1 amplitude was 0.49 ± 0.16.

Obtaining this characteristic pattern was dependent upon the stimulus parameters used to record the SSEP’s and the position of the recording electrodes. As shown in Fig. 3 left, with a stimulus current of 0.1 mA, the N1 and N2 components were evoked, but the N3, N4, and P2 waves were absent. With a stimulus current of 0.2 mA, all components were evoked, but maximum amplitudes of N3 and N4 were obtained only with a stimulus current of 0.5 mA or above. Stimulus frequencies from 0.5 to 3.1 Hz produced identical SSEP’s; however,
at 24 Hz and above, the amplitudes of $P_2$ and $N_4$ decreased, and the latency of $N_4$ increased slightly (Fig. 3 right). Recordings from spinal segmental levels varying from L2-3 to L5-6 are shown in Fig. 4. Two levels, L5-6 (referred to L4-5) and L4-5 (referred to L3-4), had the highest amplitude and clearest separation of the components, and were used in this study.

The latencies of $N_1$ to $N_4$ were not significantly different when the SSEP's were evoked by stimulation of the right or left sciatic nerve. The SSEP's were unchanged by insertion of the Swan-Ganz catheter into the right femoral artery. The drugs given in the Group 3 rabbits did not significantly change either the amplitudes or the latencies of the component waves; however, hypothermia both increased the latencies and decreased the amplitudes of all the waves. The return of the SSEP's during reperfusion in the animals with hypothermia was compared to a control SSEP taken at the same temperature during the process of cooling. Although methohexital can obliterate the late components of the cortical evoked potentials, the SSEP's remained unchanged by doses up to 8 mg/kg.

**Physiological Events During Ischemia and Reperfusion**

When the balloon of the Swan-Ganz catheter was inflated in the abdominal aorta, systemic blood pressure abruptly increased and heart rate decreased. The arterial pressure distal to the inflated balloon fell to near zero and no pulsations were recorded. While the balloon remained inflated, systemic blood pressure and heart rate slowly decreased. Upon deflation of the balloon, systemic blood pressure abruptly decreased and heart rate increased and then slowly returned to normal. A fall in arterial $pH$ was usually measured due to release into the systemic circulation of the lactate produced by ischemia of the tissues supplied by the distal abdominal aorta (Fig. 5).

The treatments given prior to aortic occlusion had varying effects on cardiovascular parameters. Thiopental caused a transient drop in blood pressure which returned to normal levels prior to the onset of ischemia; blood pressure was well maintained during reperfusion. Verapamil caused a decrease in blood pressure and heart rate which persisted until the onset of ischemia. There was also prolonged hypotension during early reperfusion in the animals pretreated with verapamil. Hypothermia, naloxone, and methylprednisolone did not consistently change pre-ischemia blood pressure or the blood pressure response to ischemia and reperfusion.

There was no difference in the pre-ischemia blood pressure, $pO_2$, $pCO_2$, $pH$, or temperature between the animals that had a normal neurological outcome and those that became paretic or paraplegic. However, the animals that were paretic or paralyzed had significantly lower blood pressures during reperfusion and required more sodium bicarbonate to correct their acidosis than those that were normal.
Outcome and evoked potentials in spinal ischemia

**SSEP Changes During Ischemia and Reperfusion**

At the onset of ischemia, the component waves of the SSEP's became smaller and their latency increased, then disappeared in the following sequence: P2 followed by N4, N3, N2, and N1 (Fig. 6). The wave amplitudes were more sensitive to ischemia than were their latencies; that is, the change in amplitude occurred earlier than the change in latency. As shown in Fig. 7, the later waves (N3 and N4) were more sensitive to ischemia than were waves N1 and N2.

Pretreatment with drugs in Group 3 rabbits did not change the sequence of SSEP changes, but did affect the time course. The average time required for the amplitude of the waves to decrease by 50% during ischemia (T89) in the Group 1 rabbits was 13.5, 5.8, 6.0, 3.8, and 2.3 minutes for waves N1 to N4 and P2, respectively. In Group 3 animals, agents that improved neurological outcome during 25 minutes of ischemia, as described below, increased the T89 of the waves, while verapamil, which worsened neurological outcome, shortened the T89 of the waves. Thus, the T89 of the waves, especially the late waves which are more sensitive to ischemia, provided an estimate of the relative protection of a drug. The average T89 of N3 was 15.0, 14.2, 12.4, 8.9, and 2.8 minutes with hypothermia, thiopental, naloxone, methylprednisolone, and verapamil, respectively.

The length of time that the late waves were absent during ischemia appeared to have some relationship to neurological outcome in Group 1 rabbits. Animals in which the N3 wave was absent for less than 10 minutes were always normal, regardless of the total ischemia time. Animals in which N3 was absent for more than 10 minutes were always paretic or paralyzed. Animals in which the N3 wave was absent for exactly 10 minutes had a variable outcome: three were normal, two were paretic, and three were paraplegic. When drug interventions were added, however, this relationship was not consistent. One rabbit that was hypothermic had a normal outcome even though N3 was absent for 18 minutes during ischemia.

The SSEP component waves returned after deflation of the balloon in reverse order of their disappearance (Fig. 6), and were identifiable in almost all animals after reperfusion, regardless of their neurological outcome. The latency of the individual waves normalized faster than the amplitude.

The amplitude of the SSEP component waves in most of the paretic and paralyzed animals decreased over time, being greater at 120 minutes than at 48 hours. In some animals this decline in amplitude was observed to begin in the first 120 minutes after reper-
Neurological outcome of 31 rabbits in Group I*

<table>
<thead>
<tr>
<th>Ischemia Time (mins)</th>
<th>No. of Animals</th>
<th>Outcome</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Normal</td>
</tr>
<tr>
<td>15</td>
<td>5</td>
<td>5 (100)</td>
</tr>
<tr>
<td>17</td>
<td>10</td>
<td>5 (50)</td>
</tr>
<tr>
<td>20</td>
<td>6</td>
<td>2 (33)</td>
</tr>
<tr>
<td>25</td>
<td>8</td>
<td>4 (50)</td>
</tr>
<tr>
<td>60</td>
<td>2</td>
<td>0</td>
</tr>
</tbody>
</table>

* Numbers in parentheses are percentages of the total number of rabbits studied at each time. For a description of Group 1 see text.

Neurological Outcome

Motor function was used for classifying the animals as normal, paretic, or paralyzed. Rabbits were considered to have normal motor function if they were able to hop with no muscle spasticity, and if their muscle strength was 4/5 or 5/5. Sensory abnormalities were noted, however, in almost all of the animals. They were usually hyperalgesic between L-1 and L-6 and hypalgesic below L-6. A few of the animals that had normal motor function at 48 hours exhibited spasticity for a few hours after awakening from anesthesia, but then their neurological function improved. Paretic animals could move about with spastic hindlimbs, but were unable to hop. Hypalgesia below L-6 was present. Paralysis was defined as the absence of muscle tone or contraction. Anesthesia was present below the L-6 level. Manual evacuation of the bladder and bowel was necessary in these animals. The flaccid muscles atrophied quickly, and by 7 days the body weight had decreased by as much as 500 gm.

Table 1 shows that the neurological outcome of the 31 Group 1 animals was dependent on the duration of spinal cord ischemia. After occlusion of the abdominal aorta in five animals for 15 minutes, all animals retained normal motor function. After ischemia in two animals for 60 minutes, both were paraplegic. Intermediate ischemia times resulted in paraplegia in 30% to 38% of the rabbits, and 12% to 33% were paretic.

Neurological outcome of the 36 rabbits in Groups 2 and 3*

<table>
<thead>
<tr>
<th>Anesthesia</th>
<th>Treatment</th>
<th>No. of Animals</th>
<th>Outcome</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Normal</td>
</tr>
<tr>
<td>none</td>
<td>none</td>
<td>15</td>
<td>0</td>
</tr>
<tr>
<td>ketamine</td>
<td>none</td>
<td>8†</td>
<td>4</td>
</tr>
<tr>
<td>ketamine</td>
<td>hypothermia</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>ketamine</td>
<td>thiopental</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>ketamine</td>
<td>naloxone</td>
<td>4</td>
<td>3</td>
</tr>
<tr>
<td>ketamine</td>
<td>methylprednisolone</td>
<td>5</td>
<td>3</td>
</tr>
<tr>
<td>ketamine</td>
<td>verapamil</td>
<td>4</td>
<td>0</td>
</tr>
</tbody>
</table>

* All animals underwent 25 minutes of ischemia. For a description of groups see text.

† These eight rabbits are the Group 1 animals that underwent 25 minutes of ischemia: they are shown for comparison but are not included in the total for Groups 2 and 3.

FIG. 8. Early recovery and late failure of the spinal somatosensory evoked potential's (SSEP's) after 25 minutes of ischemia in a rabbit undergoing stimulation of the left sciatic nerve (A) and the right sciatic nerve (B). In this animal, N3 and N4 were abolished by 20 minutes of ischemia. Reperfusion was started at 24 minutes of ischemia. After 120 minutes, the SSEP's had partially recovered. The animal was paretic but able to walk at 24 hours after reperfusion. At 48 hours, the animal was paraplegic and the N3 and N4 waves were absent.
Outcome and evoked potentials in spinal ischemia

**TABLE 3**

*Predictions based on SSEP recordings correlated with 48-hour neurological outcome*

<table>
<thead>
<tr>
<th>Features Compared</th>
<th>Group 1</th>
<th>Group 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>no. of 48-hr survivors</td>
<td>16</td>
<td>17</td>
</tr>
<tr>
<td>predicted normal</td>
<td>9</td>
<td>11</td>
</tr>
<tr>
<td>outcome normal</td>
<td>10</td>
<td>11</td>
</tr>
<tr>
<td>predicted abnormal</td>
<td>7</td>
<td>6</td>
</tr>
<tr>
<td>outcome paretic/paralyzed</td>
<td>6</td>
<td>6</td>
</tr>
</tbody>
</table>

*SSEP = spinal somatosensory evoked potential. For a description of groups see text.*

Paretic or paraplegic. These animals had no ventilatory or blood pressure support or correction of acidosis during ischemia and reperfusion. Fifty percent of animals that were ventilated, sedated with ketamine and paralyzed during ischemia, and had the metabolic acidosis corrected during reperfusion had a normal motor outcome. Pretreatment with hypothermia, thiopental, or naloxone, in addition to supportive care, increased the proportion of animals with normal motor outcome to 100%, 100%, and 75%, respectively. Pretreatment with methylprednisolone did not significantly change neurological outcome; in contrast, pretreatment with verapamil resulted in all rabbits becoming paraplegic.

**Neurological Outcome Correlated with Recovery of SSEP's**

In 16 of the Group 1 rabbits, SSEP's were monitored for 120 minutes after reperfusion and were compared to the neurological outcome at 48 hours. In these rabbits, the behavior of the SSEP's during ischemia and reperfusion was studied for predictors of long-term neurological outcome. The N1 and N2 waves returned during reperfusion in almost all animals regardless of the neurological outcome. The degree to which the late waves returned, however, correlated with the final neurological outcome.

As shown in Fig. 9, the N3/N1 amplitude returned to at least 70% of control levels, the N4 amplitude to at least 40% of control, and the P2 amplitude to at least 30% of control by 120 minutes after reperfusion in all animals that were normal at 48 hours. No animal that was paretic or paraplegic at 48 hours had the N3/N1 or N4 amplitude return to greater than 89% of the control level, or the P2 amplitude return to greater than 59% of control at 120 minutes of reperfusion. Thus, an animal that had a 90% or greater return of the N3/N1 amplitude by 120 minutes after reperfusion could be predicted to have a normal outcome, and an animal in which the N3/N1 amplitude returned to less than 70% of control at 120 minutes after reperfusion could be predicted to be paretic or paralyzed by 48 hours. Seven rabbits, however, had the N3/N1 amplitude return to 70% to 89% of the control level. Their neurological outcome at 48 hours was variable; five were normal and two were paraplegic. In these animals with intermediate return of the late negative waves, however, neurological outcome could still be predicted by measuring P2. The P2 amplitude returned to greater than 60% of the control level in all but one that had a normal outcome, and to less than 60% of the control in all that were paralyzed.

The SSEP criteria that predicted a normal neurological outcome in the Group 1 animals, an N3/N1 amplitude equal to or greater than 90% of control, or an N3/N1 amplitude of 70% to 89% of control and a P2 amplitude greater than 60% of control at 120 minutes after reperfusion, were used to predict the outcome of the 17 Group 3 rabbits that survived 48 hours. Eleven were predicted to be normal and all were normal at 48 hours; six were predicted to be abnormal and all were paretic or paralyzed at 48 hours (Table 3).

**Pathological Examination**

Histopathological examination of the spinal cords from 16 representative animals revealed lesions in 10 of the rabbits. The lesions, almost entirely confined to the gray matter, were typical infarctions, characterized mainly by areas of neuronal destruction and replacement by mononuclear phagocytic cells. Damaged areas of the spinal cords usually extended from the midlumbar to the upper sacral levels.
The histology of these spinal cords showed that longer periods of ischemia are more likely to produce lesions, and the histological lesions correlated with the neurological deficits produced by the ischemia. Only one of the four animals subjected to 15 minutes of ischemia developed a small lesion; all had a normal motor examination. Seventeen minutes of ischemia resulted in lesions in three of six animals; the three animals with lesions were either paretic or paralyzed. The one rabbit that was ischemic for 20 minutes was parietic and exhibited a lesion, and both animals that were paralyzed after 60 minutes of ischemia developed severe lesions (Fig. 10). A more detailed description of the neuropathology of these lesions will be reported in a forthcoming study.

Discussion

Balloon occlusion of the abdominal aorta has a number of advantages as a method of producing spinal ischemia. The surgical procedure is simple, quickly accomplished, and less invasive than ligating the aorta through a laparotomy. The adequacy of aortic occlusion by the balloon can be monitored throughout the time of ischemia by recording the pressure distal to the balloon, although collateral circulation may still supply some blood flow to the cord despite complete occlusion of the abdominal aorta. Normal systemic arterial blood gases and pH can be maintained by ventilating the animal and by correcting post-ischemia acidosis with sodium bicarbonate to assure that the injury is not exacerbated by systemic hypoxia or acidosis.

As in previous studies of spinal cord ischemia, the neurological outcome in this present investigation was found to be dependent on the duration of ischemia: ischemia of 15 minutes' duration is well tolerated by the rabbit spinal cord; ischemia of 60 minutes' duration always results in paraplegia; and ischemia of 17 to 25 minutes' duration results in paresis or paraplegia in about 50% of the animals. Most other studies have found 25 minutes of ischemia to cause paraplegia in all rabbits.\(^6\) The difference from the present study probably results from variations in supportive care and anesthesia, and possibly from the pancuronium given to animals in this study, since our animals that were metabolically unsupported during 25 minutes of ischemia were all parietic or paraplegic.\(^1\) The heparin that these animals received to prevent thrombosis around the catheter may also have contributed to the improved outcome.

For screening of drugs that might provide protection against ischemia, 25 minutes of ischemia may be a useful time, allowing detection of both improvement and impairment of outcome. Because the purpose of the investigation in the Group 3 animals was primarily to confirm that the SSEP response to ischemia was unchanged by pretreatment with a variety of drugs, the number of animals in each group is too small to permit us to draw definite conclusions about change in neurological outcome. However, use of hypothermia and administration of thiopental and naloxone (drugs that have been shown to improve neurological outcome in other ischemia models) allowed almost all animals to tolerate 25 minutes of ischemia without a deficit.\(^5\) Methylprednisolone, which appears to have little effect on outcome from ischemia, resulted in a distribution of deficits similar to that noted without drug administration. Verapamil, which has not improved outcome in other models of central nervous system ischemia, actually worsened outcome in this model; all rabbits became paraplegic.\(^\text{8}\)

Monitoring of SSEP's evoked by stimulation of the sciatic nerve appears to be a promising method of predicting neurological outcome after cord ischemia. The SSEP is highly characteristic, consisting of an initial positive component (P1), four negative components (N1 to N4), and a longer, smaller late positive wave (P2). If the generators of these waves were known, their differential sensitivity to ischemia would throw light on the sites of damage produced by cord ischemia. The generators of these waves are not known with certainty, but some reasonable hypothesis can be advanced based upon the physiological behavior of the potentials. It appears that N1 and N2 are presynaptic components on the basis of their early latency, large amplitude, and their presence even at high stimulating frequencies, and the fact that they are strongly resistant to ischemia. Since N1 was not recorded rostral to L-2, this wave may represent dorsal root and dorsal column fibers that do not ascend higher in the cord. The contrasting behavior of N3 and N4 to that of the first two negative waves suggests a postsynaptic origin.\(^3,11\) The N3 and N4 waves are not the same component, since N4 is more sensitive to N3 to ischemia, and N4 disappears at lower stimulation rates than N3. The P2 component is the most sensitive of all of the waves to ischemia, and to increasing rates of stimulation. Those characteristics as well as its time course and polarity

---

Fig. 10. Photomicrograph of a cross section of spinal cord at L-6 from a paraplegic rabbit sacrificed 4 days after aortic occlusion for 25 minutes. Bilateral infarction of the anterior gray columns can be seen. H & E, x 13.
Outcome and evoked potentials in spinal ischemia

suggest that P2 is the extracellularly recorded field potential generated by hyperpolarizing inhibitory postsynaptic potentials. Gelfan and Tarlov\(^3\) have demonstrated that the inhibitory interneurons in the dog’s spinal cord are particularly vulnerable to ischemia.

The degree to which the N3 and P2 components of the SSEP had recovered at 120 minutes after reperfusion could be correlated with the final neurological outcome. Animals in which the N3/N1 amplitude returned to 90% or more of the control level or in which the N3/N1 amplitude returned to 70% to 89% of control and the P2 amplitude was greater than 60% of control at 120 minutes were found to be normal at 48 hours. Other animals were either parietic or paraplegic at 48 hours. These criteria were valid even when drugs were given. Future studies using this model to investigate protection of the spinal cord from ischemia might use SSEP’s to predict neurological outcome rather than relying on long-term neurological observation.

Monitoring of SSEP’s permitted identification of animals in which there was an initial recovery of the N3 and N4 waves (presumably postsynaptic potentials) after periods of ischemia, but in which the eventual result was paralysis. This observation appears to be of some significance because it demonstrates that rather prolonged periods of ischemia do not destroy the cellular processes necessary for synaptic transmission and generation of postsynaptic potentials at the time of ischemia. Rather, it appears that these processes can recover from the ischemia, only to deteriorate later as a result of secondary processes which are poorly understood. This observation suggests the possibility of therapeutic modification of these secondary processes within the first 24 to 48 hours after spinal ischemia.

Acknowledgments

The technical assistance of Susan Irish, the editorial assistance of Sharon Kahler, and manuscript preparation by Bonnie Savage are gratefully acknowledged.

References


Manuscript received August 3, 1983.
This research was supported by The William H. Lane Fund for Neurological Research.
Joseph E. Levine, M.D., Ph.D., participated in preliminary studies of recording and balloon catheterization.
Address reprint requests to: Robert G. Grossman, M.D., Department of Neurosurgery, Baylor College of Medicine, 1200 Moursund Avenue, Houston, Texas 77030.