An Intraaortic Solution Trial to Prevent Spinal Cord Injury in a Rabbit Model†

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Objectives: to evaluate the effectiveness of an intraaortic delivered solution on preventing spinal cord injury.

Design: forty rabbits were allocated into five equal groups.

Materials and Methods: one clamp was placed just distal to the left renal artery, and another was placed just above the iliac bifurcation for 40 min. Group 1 was not infused (control group). Through a 24G vascular catheter inserted into the isolated aortic segment, 20 ml of LR solution at room temperature (Group 2) 20 ml of LR solution at 3 °C (Group 3), and 20 ml of LR solution at 3°C containing 30 mg/kg of methylprednisolone (Group 4) were infused over 3 min. In Group 5, 10 mg/kg of vitamins E and C were delivered two days before the experiment, and 20 ml of LR solution at 3°C containing 30 mg/kg of methylprednisolone, and 10 mg/kg of vitamins E and C was infused at the operation. Postoperative spinal cord function was assessed using Tarlov’s criteria.

Results: the neurologic status of Groups 3, 4, and 5 was significantly superior to that of Groups 1 and 2. No paraplegia was observed in Groups 4 and 5. Spastic paraplegia occurred in all rabbits of Groups 1 and 2, and in 20% of Group 3. In the electron microscopic evaluation of spinal cord specimens, normal histologic structure was observed in Groups 4 and 5, whereas, some derangements were observed in all others.

Conclusions: intraaortic infusion of a hypothermic blended solution containing methylprednisolone, vitamins C and E provided best protection against postischaemic spinal cord dysfunction.

Key Words: Spinal cord protection; Paraplegia; Ischaemic injury.

Introduction

Ischaemia of spinal cord is a major complication (6–40%) following operations on the thoracoabdominal aorta.1–5 However, with the introduction of adjunctive methods, the neurologic risk associated with type I thoracoabdominal or total descending thoracic aortic repair has dramatically reduced to less than 5%.6–8

Numerous methods have been developed to protect the spinal cord against ischaemic injury. These range from purely mechanical, such as preservation and reimplantation of important intercostal and lumbar arteries, cerebrospinal fluid drainage or perfusion of the aorta beyond the cross-clamp, to purely pharmacologic, such as the use of vasodilators, neutrophil-blocking antibodies, and free radical scavengers.9–11

Other, more experimental models of spinal cord protection, such as intraaortic delivery of various pharmacologic agents including vitamins, retrograde venous perfusion, perfusion of the subarachnoid space with cold solutions or oxygenated perfluorocarbons, have also been investigated.11–13

Veno and coworkers found that intraaortic delivery of a hypothermic lactated Ringer’s (LR) solution containing methylprednisolone, and vitamins C and E had a neutroprotective effect on postischaemic-reperfusion injury of spinal cord in rabbits.12 In the present study we additionally wanted to assess the effect of an extrasupplementation of vitamins C and E given two days before the experiment.

Materials and Methods

Forty New Zealand white rabbits, weighing 2.5 to 4 kg, were used in compliance with the “Guide for the Care...
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Fig. 1. Experimental model and delivery of infusions.

Inferior mesenteric artery

Heat exchanger

Infusion pump

and Use of Laboratory Animals” published by the “National Institutes of Health (NIH publications, No. 85-23, revised 1985)”. Five groups of 8 animals each underwent preparation, anaesthetic management and operation.

A midline laparotomy incision was used to expose the aorta between the left renal artery and iliac bifurcation. Heparin 150 IU/kg was given intravenously before the cross-clamping and was not revised. One clamp was positioned just below the left renal artery, and another was placed at the bifurcation. This produces spinal cord ischaemia in the rabbit and after 40 min all control animals are rendered paraplegic.11,14

In Group 1, no further procedure was performed. In all other groups, a 24G vascular catheter was inserted into the isolated aortic segment (Fig. 1). In Group 2, 20 ml of LR solution at room temperature, in Group 3, 20 ml of LR solution at 3 °C, and in Group 4, 20 ml of LR solution plus 30 mg/kg methylprednisolone (MP) were delivered. In Group 5, 10 mg/kg of vitamins C and E were delivered intravenously two days before the operation, and 18.7 ml of LR solution plus 30 mg/kg methylprednisolone plus 10 mg/kg each of vitamins C (0.1 ml) and E (0.2 ml) were infused at the experiment. The total volume in each infused group was 20 ml. The puncture site was sutured with 7.0 polypropylene.

Statistical analyses were performed by SPSS/PC+ (version 8.0) computer program. The probability (p) less than 0.05 was considered significant. The mean and standard deviation values of all data were calculated and indicated. For the changes in neurologic status, Mann–Whitney U-test and Kruskal–Wallis analysis of variance were used. Paired t-test was used in the comparison of arterial pressures.

Clinical evaluation

Spinal end function was assessed at 24 and 48 h by Tarlov’s criteria: grade 0, spastic paraplegia with no movement of the hind limbs; grade I, spastic paraplegia with slight movement of the hind limbs; grade II, good movement of the hind limbs, but unable to stand; grade III, able to stand, but unable to walk normally; grade IV, normal walking and complete recovery.

Histopathology evaluation

At 48 h animals were sacrificed and their spinal cords removed. Specimens of lumbosacral spinal cords were prepared and fixed by a solution containing 5% glutaraldehyde phosphate. Thereafter, electron microscopic evaluation of the specimens were performed.

Results

At 48 h, the neurological status of Groups 3, 4, and 5 was significantly superior to that of Groups 1 and 2
Table 1. Neurologic evaluations according to Tarlov’s criteria and arterial blood pressures (mean ± standard deviation) of all groups on postoperative day 2.

<table>
<thead>
<tr>
<th>Blood pressure</th>
<th>Group 1</th>
<th>Group 2</th>
<th>Group 3</th>
<th>Group 4</th>
<th>Group 5</th>
</tr>
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<tbody>
<tr>
<td></td>
<td>76.25 ± 2.92</td>
<td>76.75 ± 2.25</td>
<td>77 ± 2.39</td>
<td>77 ± 2.39</td>
<td>77.38 ± 2.81</td>
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<tr>
<td>Neurologic status</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Grade 0</td>
<td>6</td>
<td>5</td>
<td>2</td>
<td>0</td>
<td>0</td>
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<tr>
<td>Grade 1</td>
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<td>3</td>
<td>0</td>
<td>0</td>
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<tr>
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</tr>
<tr>
<td>Grade 4</td>
<td>0</td>
<td>0</td>
<td>3</td>
<td>5</td>
<td>8</td>
</tr>
</tbody>
</table>

Fig. 2. Foldings on nuclear membrane, derangement and swelling of mitochondria forming vacuoles, and decreased ribosomes (lead cytrate–uranyle acetate×7000, n: nucleus, m: mitochondria, and arrows indicate foldings on nuclear membrane).

(Table 1). None of the rabbits in both group 4 and group 5 showed paraplegia (grade 0 or 1).

At electron microscopy there was nuclear membrane following vaculation of mitochondria and a reduction in decreased ribosomes in Group 1 (Fig. 2). Group 2, nuclear condensation, disintegration of the nuclear membrane and vacuolisation of cytoplasm to structures was observed (Fig. 3). In Group 3, derangements on membranes of nucleus and mitochondria, and disintegration of nuclear membrane were detected (Fig. 4). The completely normal, well preserved histologic structure of spinal cord was observed in healthy animals of Groups 3, 4, and 5 (Fig. 5).
Deep hypothermia with total cardiopulmonary bypass and circulatory arrest has been proposed to protect the spinal cord in thoracic and thoracoabdominal aortic aneurysm repairs. However, haemorrhagic, pulmonary, and neural complications of this method limit its usefulness.  
In the present study local hypothermia had a protective effect on spinal cord injury. Numerous extensive studies have been performed to clarify anatomic, physiologic, and biochemical aspects of traumatic spinal cord injury. Oxygen radical-mediated lipid peroxidation has been suggested as an important factor in posttraumatic neuronal degeneration. Lipid peroxidation can involve circumferentially the undamaged neuronal tissue, leading to collapse of microcirculation and to irreversible damage to myelin and axons. Similar pathophysiologic changes also occur in ischaemic spinal cord injury, such as calcium influx, eicosanoid production, hypoxic free-radical generation and subsequently developed lipid peroxidation. Therefore, a number of pharmacologic methods applied to treat traumatic spinal cord injury have been used to reduce ischaemic or reperfusion injury of the spinal cord and to prevent
paraplegia. Methyl prednisolone inhibits lipid peroxidation. Moreover methylprednisolone causes local vasodilation, reverses intracellular calcium influx, inhibits lipid hydrolysis, maintains the aerobic energy metabolism, and enhances neuronal excitability and synaptic transmission. In the present study, methyl prednisolone provided a statistically superior neurologic outcome.

Vitamin E can function as an antioxidant and membrane stabilizer in spinal cord injury. Vitamin C also reduces free radical induced lipid peroxidation. However, both intravenous must be given preoperatively to provide maximal beneficial effect against spinal cord injury.

In conclusion, the combined delivery of hypothermic (3 °C) LR solution, 30 mg/kg methyl prednisolone, 10 mg/kg each of vitamins C and E added to pretreatment of 10 mg/kg each of vitamins C and E provided the best neurologic outcome and recovery.

References


