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Introduction  Treatment using mild hypothermia, temperatures from 33˚C to 35˚C, has been reported to be a very effective therapeutic method on central nervous system damage such as brain trauma and ischemia. Until now, there have been reported the effectiveness of hypothermic treatment for the treatment of brain (1) and spinal cord (2) injuries. Systemic hypothermic treatment under anesthesia has been introduced for heart and aortic surgery (3) and recently, also introduced into the emergency clinical field (4). The efficacy of this treatment has been established, but several serious side effects have been reported. Among them, sepsis induced by the suppression of the immune system is one of the most serious and fatal side effects (5). To avoid these serious systemic side effects, local cooling should be considered. The ideal conditions of hypothermic treatment for spinal cord were 1) under awake, and 2) easy and adequate control of local temperature. To achieve the conditions mentioned above, we used the Peltier modules for a core part of our thermoelectric cooling device. Peltier modules use the Peltier effect to create a heat flux between the junction of two different types of materials. Peltier modules transfer heat from one side to the other side against the temperature gradient, with consumption of electrical energy. In the present study, we developed a new device for spinal cord local hypothermia and tested the device for the treatment of experimental thoracic spinal cord injury in rats. We evaluated the efficacy of the device both in rat hind-limb function and histological change after the traumatic spinal cord injury.

Materials and methods  Rat spinal cord contusion injury was performed at the 11th thoracic vertebral level using a MASCIS impactor. Our new device for local hypothermia consist of the extracorporeal electrically cooling component using Peltier modules and intracorporeal aluminum arched plate which was placed on the lamina. Since the lamina was removed for the contusion injury, the dura mater of the 11th vertebra level was closed to the aluminum plate (about 1.5mm). The plate ending (about 3 cm) was projected from the surface of the skin for connection to extracorporeal component. After the operation including spinal cord contusion injury and insertion of the intracorporal aluminum plate, the extracorporeal cooling component was connected to the aluminum plate (Fig. 1). The temperature controlling was performed using this cooling device and animal body temperature controller by infrared rays irradiation (IFR-100, Unique Medical Co. Tokyo, Japan). The spinal cord cooling (33˚C; hypothermic animal) was performed for 48 hour after the contusion injury. During the hypothermic treatment, rats were awake and could move in the cage. The normothermic animal received the same procedure and the temperature of spinal cord maintained to 37˚C. The devise was removed after the hypothermic or normothermic treatment. Hind-limb motor function was assessed with the Basso, Beattie and Bresnahan (BBB) scoring scale (6). The BBB scale is a 21-point scale that ranks no locomotion as 0 points and a normal gait as 21 points. For the histological examination, spinal cord axial sections (10 µm thickness) were stained by anti-OX-42 antibody and anti-TNF-α antibody. The number of OX-42-positive cells and TNF-α-positive cells in 220µm x 170µm area were counted in the slice 1cm rostral to injured site.

Results  The hind-limb function, evaluated by BBB scale, in the hypothermia-treated animals was significantly better than that in the normothermia-animals from two weeks to six weeks after the SCI (Fig. 2). In the lateral funiculi, the number of OX-42-positive macrophage in hypothermia-treated animals (3.7±1.15; n = 6) was significantly (p < 0.05) less than that in the normothermia animals (24.1±5.04; n = 6) three days after the injury. In the dorsal horn, the number of TNF-α-positive cells in hypothermia-treated animals was 1.95 ± 0.6 (n = 6) and 1.73 ± 0.09 (n = 6) at three days and seven days after the contusion injury. In the normothermia animals, the numbers of TNF-α-positive cells in the dorsal horn are 6.2 ± 0.73 (n = 6) and 6.23 ± 1.32 (n = 6), at three days and seven days after contusion injury, respectively. Significant difference between the hypothermic and normothermic animals was observed (p<0.05, P<0.01 in three and seven days after SCI).

Discussion  The efficacy of hypothermia on the recovery of motor function after spinal cord injury has been established (7). Ha et al.(8) reported the effect of moderate hypothermia by epidural infusion of cool liquid after spinal cord injury in rats. Until now, there is no report of electrically controlling local hypothermic system for the treatment of spinal cord injury. Stable temperature controlling system may achieve local cooling for a long duration without general anesthesia. Microglia, an immune cell in the central nervous system (CNS), mainly takes charge of the inflammatory response against infections. However, not only against infections, but also in several CNS pathological conditions such as trauma (9) and ischemia (10), microglia has been reported to proliferate and be activated. Activated microglia seems to accelerate neuronal damage by release of nitric oxide, superoxide, and cytokines such as tumor necrosis factor-α (TNF-α). The result in this study suggest that local hypothermia inhibit the inflammatory response which accelerate secondary neuronal damage.