Studies on therapeutic mechanisms of bone marrow-derived mononuclear cell and involvement of hepatocyte growth factor in acute spinal cord injury

Abstract of Doctoral Thesis

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Bone marrow-derived mononuclear cell (BM-MNC) transplantation therapy is the only cell source that can be used during the peracute phase of spinal cord injury. The reported therapeutic effect of BM-MNC including cell protective effect and angiogenesis is speculated to be due to the paracrine of growth factors. However, the detailed mechanism remains elusively unclear. The elucidation of its therapeutic mechanisms will bring clarifications on the treatment targets and the suitable timing for treatment. In addition, a clarification of the main therapeutic mechanism will lead to the development of effective therapeutic methods. The purpose of this study was to clarify the therapeutic mechanism of BM-MNC in the treatment of the acute spinal cord injury. At first, green fluorescent protein (GFP)-traced BM-MNC was transplanted into a rat model of spinal cord injury, and findings that were considered to be associated with therapeutic mechanisms were analyzed with immunohistochemically. As a result, part of transplanted BM-MNC showed characteristics of perivascular-localized macrophage at the site of injury, although this occurs only transiently. BM-MNC survived in the injured spinal cord at 7 days after transplantation and produced various growth factors at the site of injury. The expression rate of hepatocyte growth factor (HGF) was found to be the highest. Besides, the mechanism of the cytoprotective effect of BM-MNC was analyzed, and the findings showed that BM-MNC caused a phosphorylation of c-Met, HGF receptor which was expressed by rat adrenal pheochromocytoma cell line, a neuronal-model, significantly reduced the intracellular production of reactive oxygen species (ROS) and cell death. Suppression of ROS production and cell death were significantly decreased in the presence of c-Met inhibitor. To explore new therapy of acute spinal cord injury, single-dose intraspinal administration of HGF was trialed and compared its therapeutic effect with that of BM-MNC transplantation therapy. As a result, fractional anisotropy value of diffusion tensor imaging in HGF group showed significantly higher than that of control group at 14 and 28 days after administration. Besides, positive area of neuron, axon, and astrocyte markers in HGF group were significantly preserved compared with

control at 28 days after administration, but did not have enough effects compared with BM-MNC transplantation. The present study suggested that BM-MNC suppresses ROS-induced cell death, at least partly, by the paracrine of HGF. Besides, single-dose administration of HGF showed efficacy of a new therapy, although it is not enough effective compared with BM-MNC transplantation in vivo. In addition, our study revealed that BM-MNC exhibited a characteristic behavior that they adhered to blood vessels in an injured spinal cord, suggesting that they were associated with an angiogenesis promoting effect. In the future, more detailed analyses may potentially lead to the finding of new healing mechanisms and to the development of more effective therapeutic methods.