Neuroprotection and neurosupplementation in ischaemic brain

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Abstract
Possible strategies for treating ischaemic stroke include: (i) neuroprotection (preventing damaged neurons from undergoing apoptosis in the acute phase of cerebral ischaemia), and (ii) neurosupplementation (the repair of broken neuronal networks with newly born neurons in the chronic phase of cerebral ischaemia). In this paper, we review our recent progress in development of these distinct new strategies for treatment of damaged brain following a stroke. Firstly, we investigated the role of endogenous IL-6 (interleukin-6), which is one of the cytokines drastically induced by ischaemic stimuli, by administering IL-6RA (anti-IL-6 receptor monoclonal antibody) to mice. We found that endogenous IL-6 plays a critical role in neuroprotection and that its role may be mediated by STAT3 (signal transducer and activator of transcription-3) activation. Secondly, we studied the endogenous sources of the newly born neurons in the ischaemic striatum by region- and cell-type-specific cell labelling techniques. The results revealed that the SVZ (subventricular zone) is the principal source of the neuronal progenitors that migrate laterally towards the infarcted regions, and differentiate into newly born neurons. Finally, we developed a restorative stroke therapy with a bio-affinitive scaffold, which is an appropriate poly-porous structure releasing bioactive substances such as neurotrophic factor. This bio-affinitive scaffold is able to give an appropriate environment for newly born neurons. In future, we will combine these strategies to develop more effective therapies for treatment of strokes.

Key words: cerebral ischaemia, interleukin-6, neuroprotection, neurotrophic factor, scaffold, subventricular zone.

Abbreviations used: bFGF, basic fibroblast growth factor; EGF, epidermal growth factor; GFAP, glial fibrillary acidic protein; IL-6, interleukin-6; IL-6RA, anti-IL-6 receptor monoclonal antibody; MCAO, middle cerebral artery occlusion; STAT3, signal transducer and activator of transcription-3; SVZ, subventricular zone.

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Strokes have traditionally been associated with a high mortality and morbidity; however, effective therapy is not yet available. A new strategy for patients who have suffered a stroke is urgently required. Possible strategies for treating ischaemic strokes are broadly categorized into two groups: (i) neuroprotection, which is preventing damaged neurons from undergoing apoptosis in the acute phase of cerebral ischaemia, and (ii) neurosupplementation, which is the repair of broken neuronal networks with newly born neurons in the chronic phase of cerebral ischaemia [1] (Figure 1).

Neuroprotection
As effective candidates for neuroprotection, we have been interested in inflammatory cytokines, because the results of previous studies have suggested that the inflammatory reaction plays an important role in contributing to the pathophysiology of the ischaemia [2–4]. To clarify the role of endogenous IL-6 (interleukin-6), one of the cytokines induced by ischaemic stimuli [5,6], we administered IL-6RA (anti-IL-6 receptor monoclonal antibody) [7] to mice after MCAO (middle cerebral artery occlusion) and investigated its effects on downstream signal pathways, neuronal survival, infarct volume and neurological function. At 6 h after MCAO, IL-6RA administration had resulted in a significant reduction in the amount of STAT3 (signal transducer and activator of transcription-3) protein, which is one of the key effectors downstream of IL-6 [8]. At 24 h after MCAO, blockade of IL-6 signalling had led to an increase in number of apoptotic cells and enlargement of the infarct, with an adverse effect on neurological function. These results suggest that endogenous IL-6 plays an important role in neuroprotection in the acute phase of cerebral ischaemia and that its role may be mediated by STAT3 activation [9]. IL-6 and its downstream pathway can be used as a new target for stroke therapy.

Neurosupplementation
To supply new neurons into damaged brain after a stroke, two tactics are proposed. One tactic is the activation of intrinsic neural stem cells [10,11]. The other tactic is the transplantation of extrinsic neural stem cells [12] (Figure 1). In our present research, we are focusing on intrinsic neural stem cells. In a normal brain, GFAP (glial fibrillary acidic protein)-expressing cells in the SVZ (subventricular zone) of the lateral ventricles include neural stem cells that give rise to olfactory bulb neurons only [13–16]. To clarify whether neural stem cells supply new neurons to areas injured by cerebral ischaemia, we performed region- and cell-type-specific cell labelling and long-term tracing experiments. GFAP-expressing cells in the SVZ were found to generate neuroblasts that
Figure 1 | Therapeutic strategy against ischaemic stroke
Increase in cerebral blood flow (CBF) and primary neuroprotection, in which endogenous IL-6 plays an important role, is principally an important partner for essential neuroprotection. On the other hand, stem cell strategy is essentially a neurosupplement therapy. This strategy is composed of two tactics: (i) activation of intrinsic neural stem cells, whose principal origin is SVZ, as we have shown, and (ii) transplantation of extrinsic neural stem cells. Gene therapy is able to support both tactics. NTFs, neurotrophic factors.

migrated towards the injured striatum after MCAO. Long-term (90 days) tracing of the green fluorescent-labelled cells with a Cre–loxP system [17] revealed that the SVZ-derived neuroblasts differentiated into mature neurons in the striatum, in which they expressed neuronal-specific nuclear protein and formed synapses with neighbouring striatal cells [18]. These results demonstrate the role of the SVZ in neuronal regeneration after a stroke and its potential as an important therapeutic target.

For prolonged survival of newly born neurons, an appropriate environment should be prepared in ischaemic brain. Therefore we developed new methods using a new porous gelatin–siloxane hybrid derived from the integration of gelatin and 3-(glycidoxypropyl) trimethoxysilane [19]. The porous hybrid implanted into a defective part of the cerebral cortex remained at the same site for 60 days, kept integrity of the brain shape and attached well to the surrounding brain tissues. Marginal cavities of the maintained scaffolds were occupied by newly formed tissue in the brain, where newly produced vascular endothelial, astroglial and microglial cells were found with bromodeoxyuridine double positivity. In addition, those cells increased in a dose-dependent manner following the addition of bFGF (basic fibroblast growth factor) and EGF (epidermal growth factor) [1,20] (Figure 2). These results suggest that this new porous gelatin–siloxane hybrid has biocompatibility after implantation into a lesion of the central nervous system, and thus provided a potential scaffold for cell migration and angiogenesis with dose-dependent effects following the addition of bFGF and EGF.

This paper briefly highlights our recent progress in the development of these distinct new strategies for the treatment of damaged brains following a stroke. To achieve more effective therapies for patients who have suffered a stroke, it is important to combine these strategies in acute or chronic phase following the stroke.

References

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