CCL2 Is Necessary but not Sufficient to Promote Transplantation of Intra-arterially Delivered Neural Progenitor Cells to the Mouse Brain

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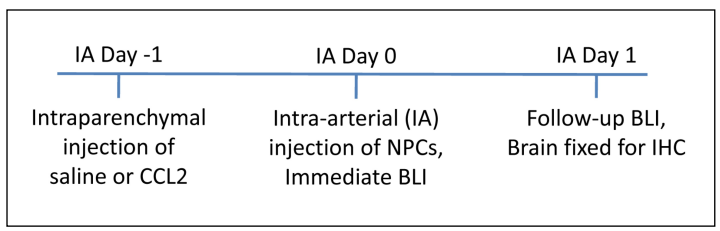
Introduction

Intra-arterial transplantation of neural progenitor cells (NPCs) to treat ischemic stroke requires efficient mechanisms for cellular migration. NPCs possess receptors to detect molecular signals released by ischemic tissue and can diapedese through blood vessel walls, potentially obviating the need for more invasive cell transplantation directly into brain parenchyma. Our group has previously demonstrated that C-C chemokine ligand 2 (CCL2) and its receptor, CCR2, play a pivotal role in directing the migration and therapeutic ability of transplanted NPCs. However, it remains unclear whether this signaling is sufficient to allow for NPC diapedesis, or if the blood-brain barrier compromise which accompanies stroke is also required. guided by CCL2 and other chemokines. Here we examine the sufficiency of CCL2 to enhance such homing following intracarotid NPC delivery in non-stroked mice.

Methods

Non-stroked FVB mice received stereotactic intraparenchymal (IP) injections of CCL2 to the striatum. 24 hours later, 500,000 NPCs expressing green fluorescent protein (GFP) and luciferase were injected into the ipsilateral common carotid artery with the external carotid artery ligated. Bioluminescence imaging (BLI) was performed immediately and 24 hours post-transplant to characterize in vivo cell distribution. Brains were then sectioned for immunohistochemical detection of GFP, ionized calcium binding adaptor molecule-1 (Iba-1), and glial fibrillary acidic protein (GFAP), the latter two to examine inflammatory effects of the injection process with or without CCL2.



Results

Post-transplant BLI showed no significant difference in signal intensity or retention of NPCs transplanted after intraparenchymal CCL2 versus saline ($p = 0.87$). IHC showed increased Iba-1 and GFAP staining specifically along the needle track.

Figure 1

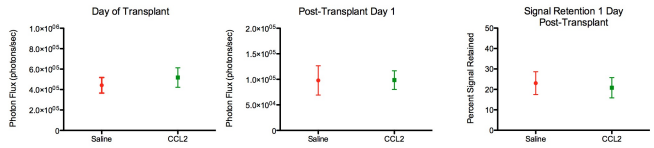


Figure 1: BLI photon flux over injection target immediately and one day after IA NPC transplantation to mice that received IP injections of either saline or CCL2. There is no signfiicant difference between groups at either time point or in the percent of retained signal.

Figure 2

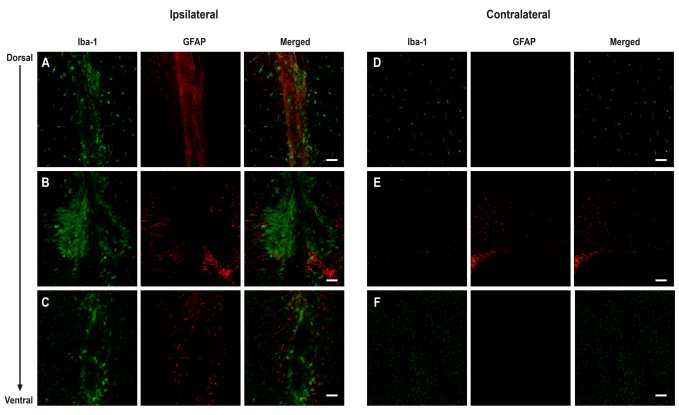


Figure 2: Iba-1 and GFAP immunoreactivity is increased along the IP injection route compared to the nontreated hemisphere. A coronal brain section from a recipient of an IP saline injection and IA NPC transplant was imaged in 20x ROIs at A. the cortical needle insertion site, B. the needle track passing through the corpus callosum, and C. the injection site in the striatum. D-F show matched ROIs in the contralateral hemisphere.

Conclusions

Although necessary, CCL2 is likely by itself insufficient to increase NPC recruitment following intra-arterial delivery in non-stroked mice. While in vitro migration data suggest some role from direct signaling, contributions from inflammation and blood-brain barrier compromise that accompany intraparenchymal injection and, to a greater degree, stroke appear to be important in allowing intravascularly-delivered cells access to the brain compartment. They may then migrate along chemokine gradients to reach sites of therapeutic potential.

Learning Objectives

After viewing this poster, participants should be able to:

1) Describe the importance of CCL2 and the post-stroke/post-injection inflammatory response in directing the transendothelial migration of intravascularly delivered neural progenitor cells.
2) Discuss, in small groups, how to use these and other methods to identify additional key mediators of effective neural progenitor cell delivery to ischemic brain regions and select for or upregulate these factors when evaluating NPC-based therapies.
3) Identify an effective treatment for ischemic brain injury using catheter-based delivery of a CCR2-enriched neural progenitor cell population, likely after thrombolysis with intra-arterial tPA.

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