Effect of Mesenchymal Stromal Cell Delivery Through Cardiopulmonary Bypass in a Piglet Model

Alice Chen[1], Kei Kobayashi[2,3], Nobuyuki Ishibashi[1,2,3]
[1]The George Washington University, [2]Department of Cardiac Surgery and [3]Center for Neuroscience Research, Children's National Hospital

THE GEORGE WASHINGTON UNIVERSITY
WASHINGTON, DC

Children's National

Introduction

Background

- Congenital heart disease (CHD) is the most common congenital disorder in the U.S.
- Increased severity of CHD has been associated with increased frequency of neurological deficits.[1]
- Young children with complex CHDs who require cardiopulmonary bypass (CPB) surgery have a significantly higher incidence of developmental abnormalities.[1]
- Microglia are resident immune cells in the central nervous system and routinely migrate toward and cluster around sites of neuronal degeneration induced by excitotoxic injury.[2]
- Mesenchymal stromal cells (MSCs) are multipotent, non-hematopoietic cells that possess immunomodulatory and regenerative properties and regulate microglia activation.

Objective

- To analyze the neuroprotective effects of intra-arterial MSC delivery through CPB in a survival porcine model

Methods

Study Design

- Two-week old female piglets were randomly assigned to one of three groups:
  (1) Control; no surgery
  (2) CPB; 34°C full flow for 150 min.
  (3) CPB+MSC; CPB followed by bone marrow-derived MSC (BM-MSC) administration before weaning, 120 min. into procedure

Data Analysis

- Morphological changes of microglia among all three treatment groups in three different cortical regions were assessed using Imaris software.
- Variables tested: 1) number of branch points from nuclei, 2) lengths of microglial processes
- P-values were determined by two-way ANOVA with Tukey comparisons using Prism to identify effect of MSC delivery on microglial structure and shape.

Results

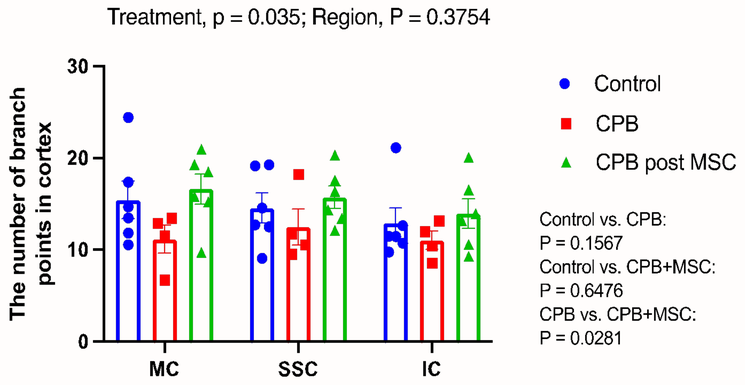

Figure 1. The number of branch points in microglia in three regions of the cortex. Data values are expressed as mean SEM. P-values were evaluated by two-way ANOVA with Tukey comparison (n=6, 4, 4).

- Four weeks post-surgery, CPB reduced the number of branch points compared to control in motor cortex (MC), somatosensory cortex (SSC) and insular cortex (IC)

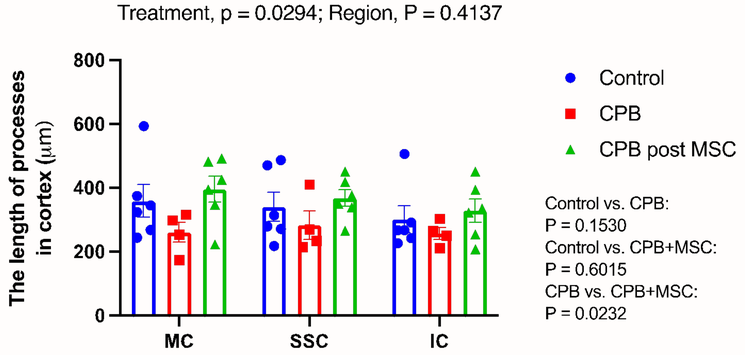

Figure 2. The lengths of microglial processes in three regions of the cortex. Data values are expressed as mean SEM. P-values were evaluated by two-way ANOVA with Tukey comparison (n=6, 4, 4).

- CPB also reduced the lengths of microglial processes in all three cortical regions
- CPB+MSC revealed significantly longer microglial processes (p=0.0281) and more branch points (p=0.0232) compared to CPB alone, indicating that morphological changes were normalized post-MSC delivery

Discussion

- In contrast to their ramified morphology in a naïve state under normal conditions, activated microglia undergo structural remodeling and adopt an amoeboid morphology with highly retracted processes, a hallmark of brain inflammation, as shown in the CPB group.[2]
- In a previous study, CPB and circulatory arrest increased microglia number three days after surgery.
  - Microglia expansion was prolonged for up to four weeks.[3]
- In MSC-treated animals, there are significantly fewer amoeboid microglia with an associated increase in the length of processes and number of branch points to mitigate the effects of CPB, indicating attenuated activation of the microglia.
- MSC delivery during CPB is highly effective and shows translational potential to minimize CPB-induced systemic inflammation and microglial activation in children with CHD.[4]
- In future studies, repeated cell administration and different dosages of MSCs should be tested to further optimize MSC treatment for CHD patients

References

1. Wernovsky G. (2006). Current insights regarding neurological and developmental abnormalities in children and young adults with complex congenital cardiac disease. *Cardiol Young*, 16(1): 92-104.
2. Nayak D, Roth TL, & McGavern DB. (2014). Microglia development and function. *Annu Rev Immunol*, 32: 367-402.
3. Korotcova L, Kumar S, & Ishibashi N. (2015). Prolonged White Matter Inflammation after Cardiopulmonary Bypass and Circulatory Arrest in a Juvenile Porcine Model. *Ann Thorac Surg*, 100(3): 1030-37.
4. Sarkislali K, Somaa F, Ishibashi N, et al. (2017). Mesenchymal Stem/stromal Cell Delivery Through Cardiopulmonary Bypass for Reduction In Systemic- and Neuro-inflammation in a Juvenile Porcine Model. *Circulation*, 136(1).