Characterization of blood Microparticles in an EAE model

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Introduction

Microparticles (MP) are membrane fragments shed by activated cells after a variety of stimuli such as stress or inflammatory processes. They play an apparently role in extracellular communication with no direct contact. They also have been shown to contain genetic material, mainly RNA and miRNA, which produce genotypic modifications in the target cell (1). The MP cellular origin can be determinate due to the presence in their membrane of characteristic marker of the cell they come from. It has been proposed that the concentration of some MP can be use as inflammatory state marker (2).

MP are related to immune processes that occurs in EAE. The characterization of this relationship can gives us information about the role that plays MP in the inflammatory and autoimmune process in MS and open new horizons for research.

In the EAE group it is expected to found a higher MP count number due to the specific leukocyte activation. In the other hand in the immunized group a lower increased in MP count is expected according to an inespecific immune activation.

The normal clinical onset in EAE model is on the 7th dpi. However, in our model it was during the 23th dpi. This may be due to the rigorous asepsis conditions because of variables that are outside the scope of this work.

Objective

Analysis of concentration and origin on blood MP in the EAE murine model.

Methods

<table>
<thead>
<tr>
<th>Time: days post immunization (dpi)</th>
<th>Monitoring: 0-30 dpi (weight and score)</th>
<th>sacrifice and necropsy: 30 dpi</th>
</tr>
</thead>
<tbody>
<tr>
<td>Immunization (0dpi)</td>
<td>EAE group: FAC, MOG, Mycobacterium</td>
<td></td>
</tr>
<tr>
<td></td>
<td>tuberculin, pertussis toxin</td>
<td></td>
</tr>
<tr>
<td>Immunized group: FAC, M.66, Mycobacterium</td>
<td>tuberculin pertussis toxin</td>
<td></td>
</tr>
<tr>
<td>Control group: no intervention</td>
<td>Flow cytometry: Gating strategy</td>
<td></td>
</tr>
</tbody>
</table>

Results

- The total MP results do not exhibit the same correlation as the CD45+ MP shown. This could be due to changes in MP from other cellular origins (platelet, endothelial).

Discussion and conclusions

- The methodology used for obtaining MP is appropriated in this animal model. However the results indicate that one mouse hearth exhaustion is not enough to achieve the necessary blood volume to perform a MP analysis. This required us to carry out a blood pool for each group.
- The normal clinical onset in EAE model is on the 7th dpi. However, in our model it was during the 23 th dpi. This may be due to the rigorous aspecific conditions in which the animals are handle and stabilize leading to a delayed immune response in a naive animals immune system (5).
- A higher number of CD45+MP was found in the EAE and immunized animals than controls. This demonstrates an immune response in the first two groups. This data corroborate partially the initial hypothesis but is not capable to discern specific from unspecific immune response. We speculate that the similar MP levels in EAE and immunized groups may be due to migration of leucocytes through the blood brain barrier (BBB) into the CNS in EAE group (5).

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REFERENCES


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