Exploring the influence of Modic Changes and spinal level on macrophage differentiation in disc herniations

Co-authors: Niek Djuric, Geraldine Lafeber, Sjoerd G. van Duinen, Carmen L.A. Vleggeert-Lankamp
Disclosures

Nothing to declare
Introduction

1. Discs can herniate in multiple ways

2. Extrusions have a higher exposure to systemic circulation after herniation, which stimulates neovascularization and may allow macrophages to enter.

3. Macrophages are believed to play a role in disc resorption

4. Modic Changes (MC) could interfere with resorption through altering the inflammatory environment, which could affect macrophages

Hypothesis

Macrophage differentiation may vary depending on the characteristics of the disc lesion and condition of the vertebral end plates

Macrophage differentiation profiles can be pro-inflammatory (M1) or anti-inflammatory (M2) macrophages

We expect higher percentages of M2 macrophages in patients without MC and higher percentages of M1 macrophages in patients with MC.
To assess the efficacy of immune-histological methods to optimally visualize pro-inflammatory M1- and anti-inflammatory M2 macrophage differentiation patterns in herniated intervertebral disc tissue
38 patients - with lumbar or cervical HNP

MRI baseline - presence of MODIC

Discectomy

Immuno-histochemistry - T-cells (CD3)
- Macrophages: CD68 (macrophage marker), CD163 (M2), CD40 (M1), Arg1 (M2) and iNos (M1).
Results: Examples of antibody distribution

**Fig. 1** Examples of antibody distribution
Fig 2. Distribution of macrophage markers between cervical and lumbar samples

- Lumbar samples showed significantly more macrophage infiltration as assessed by CD68 (p=0.045)
- Lumbar samples showed higher number of M1 and M2 markers (iNos p=0.002)
- Lumbar samples showed higher number of T-cells (p=0.026)
### Results

**Table 1** overview of inflammatory marker expression in subgroups for location of herniation and MC status

<table>
<thead>
<tr>
<th>Subgroups</th>
<th>CD68/cm²</th>
<th>CD3/cm²</th>
<th>CD15/cm²</th>
<th>CD163 %</th>
<th>Arg1 %</th>
<th>CD40 %</th>
<th>iNOS %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cerv_MC+</td>
<td>40</td>
<td>0</td>
<td>2</td>
<td>21%</td>
<td>0%</td>
<td>0%</td>
<td>0%</td>
</tr>
<tr>
<td>Cerv_MC-</td>
<td>16</td>
<td>0</td>
<td>0</td>
<td>59%</td>
<td>0%</td>
<td>2%</td>
<td>0%</td>
</tr>
<tr>
<td>Lumb_MC+</td>
<td>231</td>
<td>0</td>
<td>3</td>
<td>8%</td>
<td>2%</td>
<td>0%</td>
<td>4%</td>
</tr>
<tr>
<td>Lumb_MC-</td>
<td>223</td>
<td>0</td>
<td>3</td>
<td>26%</td>
<td>0%</td>
<td>0%</td>
<td>6%</td>
</tr>
</tbody>
</table>

The values are medians for CD68, CD3 and CD15, and percentages of CD163, Arg1, CD40 and iNOS are relative expressions compared to CD68+ cells.
Limitations

- Small sample size
- Possible overestimation of the number of inflammatory cells in patients with large amounts of tissue
- Absence of correction by multiple testing
- Lumbar herniated discs have more infiltrated macrophages compared to cervical discs

- M2 macrophages (CD163+) are the dominant type of inflammatory cells in disc herniations

- Inflamed cervical herniated discs contain a higher % of M2 (CD163+) and lower % of M1 (iNOS+) macrophages compared to lumbar ones

- M2 macrophages and are more dominant in MC- as compared to MC+ patients, suggesting quicker recovery rate after surgery in patients without MC with macrophage infiltration