Superiority of Bone Marrow derived-Osteoblastic Cells (ALLOB®) over Bone Marrow derived-Mesenchymal Stromal Cells

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SUMMARY

Bone Therapeutics is a bone cell therapy company addressing high unmet medical needs in the field of bone fracture repair, more specifically in non-union and delayed-union fractures – and spinal fusion - where the bone repair process is impaired. The company has developed a unique allogeneic osteoblastic cell product (ALLOB®) derived from bone marrow which is currently tested in three Phase I/IIa proof of concept clinical trials. The purpose of the study was to directly compare ALLOB® vs. non-differentiated mesenchymal stromal cells (MSC) for their in vitro osteogenic characteristics and their in vivo osteogenic potential in order to determine which cellular type would be the most adapted for bone repair.

Declaration of Conflict of interest: All authors have no potential conflict of interest to declare.

METHODS

1. Culture and expansion of MSC and ALLOB® cells (n=7 Bone Marrow)

Bone marrow aspirate from iliac crest

In vitro culture process

MSC

and

ALLOB®

2. In vitro characterization (n=7)

Cells were characterized in vitro by morphology, immunophenotype (FACS), gene expression (qRT-PCR) and differentiation potential.

3. In vivo assessment of efficacy (n=6)

Subcutaneous injection over the calvaria of nude mice of

- MSC cells
- ALLOB® cells
- excipient (control -)

Bone formation evaluated by

- Radiology (Faxitron®)
- Histomorphometry

RESULTS

1. Cells immunophenotype, ALP activity and morphology (n=7)

<table>
<thead>
<tr>
<th>Cell population</th>
<th>Mesenchymal markers</th>
<th>Hematopoietic markers</th>
<th>Osteoblastic markers</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>FACS analysis</td>
<td>CD73/CD90/CD105</td>
<td>CD3</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>CD45</td>
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<td></td>
<td></td>
<td></td>
<td>ALP</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td>Enz. act. analysis</td>
</tr>
<tr>
<td>MSC</td>
<td>99 ± 1 (n=7)</td>
<td>3 ± 1 (n=7)</td>
<td>2 ± 0 (n=7)</td>
</tr>
<tr>
<td>ALLOB®</td>
<td>100 ± 0 (n=7)</td>
<td>7 ± 0 (n=7)</td>
<td>1 ± 1 (n=7)</td>
</tr>
</tbody>
</table>

ALLOB® also expressed significantly higher levels of Sox9 (fold change (FC) >5), Ctsk (FC > 200), Bglap (FC > 16), Bmp2 (FC > 120) compared to MSC.

2. Analysis of chondro- and osteoblastic markers gene expression (n=7)

3. Bone Formation in NMRI-Nude mice

Histomorphometrical evaluation of the bone formation

Radiological evaluation of the bone formation

Control

MSC

ALLOB®

CONCLUSIONS

ALLOB® displays superior osteogenic capacity over BM-MSCs in vitro and in vivo, and is therefore a good clinical candidate.