Comparison of Efficiency of Self-renewal and Differentiation Potential in Tendon-derived Mesenchymal Stem Cells Isolated by Magnetic Activated Cell Sorting Method or Colony-Picking Method

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My disclosure is in the Final AOFAS Mobile App.
I have no potential conflicts with this presentation.
Tendon stem/progenitor cells (TSPCs)

Identification of TSPCs
- expression of tendon cell marker
- characteristics of stem cells

Human TSPCs formed tendon-like tissue in vivo 8 weeks after transplantation with HA/TCP (left) and Matrigel (right).

Tendon consists of …
- Extracellular matrix (ECM)
- Tenocyte

Limitation of isolation

Introduction

1. Heterogeneous
2. Contamination of tenocyte

Conventional method for the isolation of Tendon-derived stem cells (Colony picking method)

Colony picking

Colony vs Tenocyte

OS: osteogenic differentiation
AD: adipogenic differentiation
CH: chondrogenic differentiation
To evaluate the efficacy of mesenchymal stem cell isolation by the magnetic-activated cell sorting (MACS) method in tendon tissue-derived cells compared to the colony picking method for isolation of MSCs by picking colony-forming cells.
Materials and Methods

- **Human tendon**
  - Flexor Digitorum Longus (FDL) tendon
  - Flexor Hallucis Longus (FHL) tendon

- **Isolation methods of Tendon-derived stem cells**
  - Conventional colony picking method
  - Magnetic-Activated Cell Sorting (MACS) method
Isolation of tendon-derived mesenchymal stem cells

<table>
<thead>
<tr>
<th>CD90+ cells</th>
<th>CD90</th>
<th>CD105+ cells</th>
<th>CD105</th>
</tr>
</thead>
<tbody>
<tr>
<td>56.51%</td>
<td>97.83%</td>
<td>99.97%</td>
<td>86.19%</td>
</tr>
</tbody>
</table>

**TDSCs (early passage, n=5)**

| CD90+        | 56.5 ± 7.1 % |
| CD105+       | 86.2 ± 1.9 % |
Colony forming capacity

* Large colony (L-Colony): > 2.5mm
### Osteogenic Differentiation Capacity

#### Alkaline Phosphatase Staining

<table>
<thead>
<tr>
<th>Colony 1</th>
<th>Colony 2</th>
<th>Colony 3</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image1.png" alt="Colony 1" /></td>
<td><img src="image2.png" alt="Colony 2" /></td>
<td><img src="image3.png" alt="Colony 3" /></td>
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</table>

#### CD90+ and CD105+

<table>
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</thead>
<tbody>
<tr>
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<td><img src="image5.png" alt="CD105+" /></td>
<td><img src="image6.png" alt="CD90+" /></td>
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#### Alizarin Red S Staining

<table>
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<tbody>
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<td><img src="image8.png" alt="Colony 2" /></td>
<td><img src="image9.png" alt="Colony 3" /></td>
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#### CD90+ and CD105+

<table>
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<tr>
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<th>Colony 3</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image10.png" alt="CD90+" /></td>
<td><img src="image11.png" alt="CD105+" /></td>
<td><img src="image12.png" alt="CD90+" /></td>
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</tbody>
</table>
Adipogenic differentiation capacity

Oil Red O staining

<table>
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<th>Colony 3</th>
<th>CD90+</th>
<th>CD105+</th>
</tr>
</thead>
</table>

Absorbance at 500nm

- Colony-1
- Colony-2
- Colony-3
- CD90+
- CD105+
CD90+ or CD105+ cells isolated using MACS showed superior MSC characteristics in the self-renewal and multi-differentiation capacities compared with cells isolated using the colony picking method.


