MSC in clinics: Liver transplantation

Pr Olivier Detry
Dpt of Abdominal Surgery & Transplantation
CHU Liège, University of Liège, Belgium

olivier.detry@transplantation.be
INFUSION OF THIRD-PARTY MESENCHYMAL STEM CELLS (MSC) AFTER KIDNEY AND LIVER TRANSPLANTATION: A PHASE I-II, OPEN-LABEL, CLINICAL STUDY

(EudraCT 2011-001822-81 & NCT01429038)

O Detry, MH Delbouille, C Lechanteur, J Somja, A Deroover, L Weekers, P Delvenne, M Meurisse, E Baudoux, Y Beguin

Dpts of Abdominal Surgery & Transplantation, Pathology, Nephrology and Hematology

CHU Liège, GIGA-R, University of Liège, Belgium
LCGT

• Hematology department (Pr Yves Beguin)
• best clinical pratice to GMP
• Clinical grade, ready to use, third-party MSC
• Several clinical trials

• Clinical grade Tregs for future trials
Active MSC Clinical Trials

**HSCT**

1. **TJB0703**: Infusion of MSC as treatment for steroid resistant grade II to IV acute GVHD or poor graft function
2. **TJB0603**: Randomized double-blind study of mesenchymal stem cells (MSC) in patients undergoing matched unrelated allogeneic bone marrow or peripheral blood stem cell transplantation - A European multicentre study
3. **TJB0909**: Co-transplantation of MSC and HLA-mismatched allogeneic hematopoietic cell after nonmyeloablative conditioning: a phase II randomized double-blind study.
4. **TJB0905**: A pilot study to assess the feasibility of unrelated UCB transplantation with coinfusion of third-party MSC after myeloablative or non-myeloablative conditioning in adult patients with hematological malignancies
Active MSC Clinical Trials

Non-HSCT

1. **TJT1106**: Infusion of third-party mesenchymal stem cells after renal or liver transplantation: a phase I-II, open-label, clinical study

2. **TJT1123**: Mesenchymal stem cell therapy for the treatment of severe or refractory inflammatory and/or autoimmune disorders
<table>
<thead>
<tr>
<th>#</th>
<th>Status</th>
<th>Study Title</th>
<th>Conditions</th>
<th>Interventions</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Recruiting</td>
<td>MSC and HSC Coinfusion in Mismatched Minitransplants</td>
<td>Leukemia, Myeloid, Acute; Leukemia, Lymphoblastic, Acute; Leukemia, Myelocytic, Chronic; Myeloproliferative Disorders; Myelodysplastic Syndromes; Multiple Myeloma; Leukemia, Lymphocytic, Chronic; Hodgkin's Disease; Lymphoma, Non-Hodgkin</td>
<td>Biological: Mesenchymal stem cells; Other: Isotonic solution</td>
</tr>
<tr>
<td>2</td>
<td>Recruiting</td>
<td>Mesenchymal Stem Cells After Renal or Liver Transplantation</td>
<td>Liver Failure; Kidney Failure</td>
<td>Biological: Mesenchymal Stem Cells</td>
</tr>
<tr>
<td>3</td>
<td>Recruiting</td>
<td>Mesenchymal Stem Cell Infusion as Treatment for Steroid-Resistant Acute Graft Versus Host Disease (GVHD) or Poor Graft Function</td>
<td>Graft-versus-host Disease; Poor Graft Function</td>
<td>Biological: Mesenchymal stem cells</td>
</tr>
<tr>
<td>4</td>
<td>Not yet recruiting</td>
<td>Treatment of Atrophic Nonunion Fractures by Autologous Mesenchymal Stem Cell Percutaneous Grafting</td>
<td>Nonunion Fracture</td>
<td>Biological: Mesenchymal stem cells; Other: Culture medium without MSC.</td>
</tr>
<tr>
<td>5</td>
<td>Recruiting</td>
<td>Mesenchymal Stem Cell Therapy for the Treatment of Severe or Refractory Inflammatory and/or Autoimmune Disorders</td>
<td>Crohn’s Disease</td>
<td>Biological: Mesenchymal Stem Cells (MSC)</td>
</tr>
<tr>
<td>6</td>
<td>Completed</td>
<td>Mesenchymal Stem Cell Infusion as Prevention for Graft Rejection and Graft-versus-host Disease</td>
<td>Hematological Malignancies</td>
<td>Procedure: Mesenchymal stem cell infusion</td>
</tr>
</tbody>
</table>
Mesenchymal Stem Cells

MSC: technical aspects
MSC production

- Collect 30-60 ml BM healthy allogeneic volunteers
- Day 0: culture initiation (MNC obtained by ficoll isolation):
  DMEM-Glu + 10% FBS + 1% Pen/Strep
- 2X/week: medium exchange
- Day 14: 1st passage (trypsinisation et replating)
- Day 21: 2nd passage
- Day 28: Harvest and freezing
MSC Production

EBMT procedure

MNC culture initiation
(DMEM + FBS + Pen/Strep)

Day 0

Day 14 P1

Day 21 P2

Day 28 harvest and freezing in aliquots

Cell feeding 2x/week

Quality Controls
MSC Production

EBMT procedure

MNC culture initiation
(DMEM + FBS + Pen/Strep)

Day 0

Day 14 P1

Day 21 P2

Day 28 harvest and freezing in aliquots

Cell feeding 2x/week

Quality Controls

MSC
Release Criteria

- **Donor**: Serology, clinical examination and consent
- **MSC Product**: Sterility, Endotoxins and mycoplasma free, Phenotype:
  - **CD105+**, **CD90+**, **CD73+**
  - **CD34-**, **CD45-**, **CD14-** **CD3-**
  - Fibroblastic morphology
  - No aggregates
  - Normal karyotypes
  - Immunosuppressive *in vitro*
PBMC (100,000 or 50,000) were stimulated (S-PBMC) with anti-αCD3/CD28 microbeads during 4 days with or without irradiated (25 Gy) MSC (10/1 or 5/1 PBMC/MSC ratios) added at the beginning of the culture. Proliferation was assessed by analysis of the cell cycle by flow cytometry. Result are expressed as the percentage of cells present in S+G2M phases (A) and as the percentage of inhibition compared to the stimulated PBMC condition alone (B).
MSC Bank in Liège (12-2006 to 03-2014)

63 cultures completed → 440 units produced

140 available
- 50 awaiting validation
- 16 non compliant
- 234 infused

146 CHU Liège
88 other hospitals

31 placebo units available
50 placebo units produced
1 placebo unit used for R&D
18 placebo units infused

- UCL St Luc Brussel
- UZ Brussel
- KUL Leuven
- UZG Gent
- UZA Antwerpen
- ZNA Stuivenberg
- AZ St Jan Brugge
- UZ Gasthuisberg
- Mont-Godinne Yvoir
- ...
INFUSION OF THIRD-PARTY MESENCHYMAL STEM CELLS (MSC) AFTER KIDNEY AND LIVER TRANSPLANTATION: A PHASE I-II, OPEN-LABEL, CLINICAL STUDY

(EudraCT 2011-001822-81 & NCT01429038)

O Detry, MH Delbouille, C Lechanteur, J Somja, A Deroover, L Weekers, P Delvenne, M Meurisse, E Baudoux, Y Beguin

Dpts of Abdominal Surgery & Transplantation, Pathology, Nephrology and Hematology

CHU Liège, GIGA-R, University of Liège, Belgium
Background

• MSC may have an immunosuppressive effect
• MSC may be used in GVHD after SCTx
• MSC may have an effect in organ regeneration
Background

• MSC may have an immunosuppressive effect
• MSC may be used in GVHD after SCTx
• MSC may have an effect in organ regeneration

• Role after organ transplantation ???
  - safety?
  - efficacy?
Objectives

• Primary endpoint: safety for LT & KT recipients
  - tolerability of infusion
  - infections (bacterial, viral, fungi)
  - cancers (PTLD, others)
  - patient and graft survivals
Objectives

• Primary endpoint: safety for LT & KT recipients
  - tolerability of infusion
  - infections (bacterial, viral, fungi)
  - cancers (PTLD, others)
  - patient and graft survivals

• Secondary endpoint 1: immunosuppression
  - rejection rate
  - biopsy
  - blood immune profile

• Secondary endpoint 2: graft function & biopsy
M&M

- Cadaveric liver & kidney recipients
- Classical immunosuppressive management
- Dose: 1.5 to $3 \times 10^6$/kg MSC
- Central IV injection at day 3 +/-2 post Tx
M&M: liver 1

• Liver transplantation
  - 2 groups: -10 MSC +
    -10 MSC -

  - no randomisation, no double-blind
  - regular immunosuppression (Tac-MMF-steroids)
  - protocol biopsy at month 6 in both groups

• MSC group:
  - tapering of Tac from month 6 to 9, then biopsy
  - tapering MMF from month 9 to 12
M&M: liver 2

• Inclusions:
  - liver candidates between 18 to 75y

• Exclusions:
  - history of K except HCC within Milan
  - active infection (D and R), including HCV, HIV
  - EBV negative
  - reTx, combined Tx, LRLTx
  - autoimmune
  - intubation
  - clinical problem at the time of injection
M&M: liver 3

- Tacrolimus: 8-12 for one month, 5-8 after one month
- MMF: 2x500mg/d
- Steroids:
  - Solumedrol: 500 mg d0, 125 mg d1, 80 mg d2, 40 mg d3
  - Medrol:
    - 32mg d4 to 6, 16mg d7 to 9,
    - 8mg d10 to 12, then 4 mg until day 30
- Antibiotics:
  - cefuroxime 3x1.5g or piperacillin-tazobactam for 5 days
  - co-trimoxazole 500 mg po 1/d for three months
- CMV prophylaxis if D+/R-
M&M: kidney 1

- Kidney transplantation
  - 2 groups: -10 MSC +
    -10 MSC –

- regular immunosuppression (antiIL2-Tac-MMF-steroids)
  - biopsy at month 3 in both groups
  - weaning of steroids
M&M: kidney 2

• Inclusions:
  - kidney candidates between 18 to 75y

• Exclusions:
  - history of K
  - active infection (D and R), including HCV, HIV
  - EBV negative
  - reTx, combined Tx, LRLTx
  - presumed impossibility to wean steroids
  - intubation
  - clinical problem at the time of injection
  - PRA >50%
M&M: kidney 3

- Anti-IL2 day 0 & 4
- Tac: 12-15 for one week, 8-12 for three months
- MMF: 2x1000mg/d
- Steroids:
  - Solumedrol: 500 mg d0, 125 mg d1
  - Medrol:
    16mg d2 to d21
    12mg d22 to d42
    8mg d43 to 63
    6 mg d64 to 84
    4mg d85 to 90
- Antibiotics:
  cefuroxime 3x1.5g or piperacillin-tazobactam for 1 day
  co-trimoxazole 500 mg po 1/d for three months
  CMV if D+/R-
M&M

• Blood: FACS, Tregs, Ig, anti-HLA

• Biopsies:
  - Histology
  - Immunohistology: C4d, CD3, CD4, CD8, CD20, CD138, CD68, CD1a, FoxP3, CMV, EBV, Hbs

• Banking of serum & biopsies
Update

• Started in early 2012

• Liver Transplantation
  - 10 MSC treated, 10 controls
    (last February 2014)

• Kidney transplantation
  - 4 MSC treated, 4 controls
# Liver recipients

<table>
<thead>
<tr>
<th></th>
<th>MSC+ (n=10)</th>
<th>MSC- (n=10)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age (years)</strong></td>
<td>62.5 (47-74)</td>
<td>58 (52-69)</td>
<td>0.516</td>
</tr>
<tr>
<td><strong>Male/Female</strong></td>
<td>8/2</td>
<td>7/3</td>
<td>1</td>
</tr>
<tr>
<td><strong>Lab MELD</strong></td>
<td>16.5 (6-29)</td>
<td>15 (8-38)</td>
<td>0.491</td>
</tr>
<tr>
<td><strong>BMI</strong></td>
<td>25.7 (20.9-38.2)</td>
<td>25.6 (22.2-33.0)</td>
<td>0.541</td>
</tr>
<tr>
<td><strong>Liver disease</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Post alcoholic cirrhosis</td>
<td>5</td>
<td>5</td>
<td></td>
</tr>
<tr>
<td>NASH</td>
<td>3</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>HCC</td>
<td>2</td>
<td>5</td>
<td></td>
</tr>
</tbody>
</table>

Median (Ranges) or n (Mann Whitney or Fischer test)
# Liver donors & Transplantations

<table>
<thead>
<tr>
<th></th>
<th>MSC+ group (n=10)</th>
<th>Control group (n=10)</th>
<th>( P )</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>57 (17-77)</td>
<td>54 (18-79)</td>
<td>0.985</td>
</tr>
<tr>
<td>Male/Female</td>
<td>4/6</td>
<td>6/4</td>
<td>0.656</td>
</tr>
<tr>
<td>CPR (Y/N)</td>
<td>4/6</td>
<td>3/7</td>
<td>1</td>
</tr>
<tr>
<td>Donor type (DBD/DCD)</td>
<td>4/6</td>
<td>5/5</td>
<td>1</td>
</tr>
<tr>
<td>BMI (kg/m(^2))</td>
<td>24 (21-31)</td>
<td>25 (22-29)</td>
<td>0.510</td>
</tr>
<tr>
<td>Intensive care stay (days)</td>
<td>4 (1-75)</td>
<td>6.5 (2-29)</td>
<td>0.401</td>
</tr>
<tr>
<td>Urinary output (mL/h)</td>
<td>82 (7-160)</td>
<td>127.5 (47-357)</td>
<td>0.096</td>
</tr>
<tr>
<td>Pressors (Y/N)</td>
<td>5/5</td>
<td>6/4</td>
<td>1</td>
</tr>
<tr>
<td>Na (mmol/L)</td>
<td>144 (133-155)</td>
<td>144.5 (141-160)</td>
<td>0.445</td>
</tr>
<tr>
<td>Total bilirubin (mg/dL)</td>
<td>0.35 (0.30-1.59)</td>
<td>0.32 (0.15-0.85)</td>
<td>0.668</td>
</tr>
<tr>
<td>AST (U/L)</td>
<td>38 (23-190)</td>
<td>48.5 (26-91)</td>
<td>0.615</td>
</tr>
<tr>
<td>GGT (U/L)</td>
<td>59.5 (14-256)</td>
<td>68 (12-144)</td>
<td>0.888</td>
</tr>
<tr>
<td>CIT (min)</td>
<td>229 (149-800)</td>
<td>345 (181-713)</td>
<td>0.386</td>
</tr>
<tr>
<td>TIT (min)</td>
<td>317.5 (186-831)</td>
<td>402.5 (216-754)</td>
<td>0.519</td>
</tr>
</tbody>
</table>

Median (Ranges) or \( n \) (Mann Whitney or Fischer test)
MSC injection in LT recipients

<table>
<thead>
<tr>
<th></th>
<th>Per protocol</th>
<th>Study (Median) (IQR; Ranges)</th>
</tr>
</thead>
<tbody>
<tr>
<td>MSC Injection day</td>
<td>day 3 +/- 2</td>
<td>3 (3-3.25; 2-5)</td>
</tr>
<tr>
<td>Dose MSC (10^6/kg)</td>
<td>1.5-3</td>
<td>2.1 (2.0-2.4; 1.9-2.7)</td>
</tr>
<tr>
<td>Injection volume (ml)</td>
<td></td>
<td>342 (322-469; 306-614)</td>
</tr>
<tr>
<td>Injection duration (min)</td>
<td></td>
<td>25 (16.2-40; 11-60)</td>
</tr>
</tbody>
</table>
## Infusional toxicity

<table>
<thead>
<tr>
<th></th>
<th>Pre Infusion</th>
<th>15 min</th>
<th>End of infusion</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body temperature (°C)</td>
<td>36.0 (35.4-37.7)</td>
<td>36.3 (35-36.9)</td>
<td>36.2 (35.5-37)</td>
<td>0.869</td>
</tr>
<tr>
<td>Mean PA (mmHg)</td>
<td>103.3 (87-124)</td>
<td>107 (84-119.5)</td>
<td>106 (94-115)</td>
<td>0.830</td>
</tr>
<tr>
<td>NI O2 saturation</td>
<td>99 (93-100)</td>
<td>100 (92-100)</td>
<td>97.5 (93-100)</td>
<td>0.670</td>
</tr>
</tbody>
</table>

Median (Ranges) (Friedman test & ANOVA)

- No hepatic artery or portal vein thrombosis
- No sign of pulmonary embolism
- No post infusional intubation
- No anaphylactic reaction, no skin reaction
- No increase of 30 days mortality
- No increase in opportunistic infection rate
Issues

• MSC have to be thawed and infused within one hour
  - problems for intra operative infusion
  - 24/7 open GMP lab

• Next step?
  - 2 injections ?

• Multicenter blinded phase 3 study ?

• Any idea or collaboration is welcome

• Active MISOT participation?
Thanks to:

• Hematology & LTCG
  - Pr Y Beguin, Pr F Baron, Dr E Baudoux, Mrs C Lechanteur

• Surgery & Transplantation
  - Pr O Detry, Pr A Deroover, Dr N Meurisse, Mrs MH Delbouille, Dr Vandermeulen

• Nephrology
  - Dr L Weekers, Dr C Bonvoisin, Dr F Jouret, Dr P Erpicum

• Pathology
  - Pr P Delvenne, Dr J Somja Dr N Blethard
Sponsors
ALLOGENEIC MSC

Immunosuppressive therapy

Bone marrow collection (volunteer)

Culture (3-4 weeks)

Freezing & banking

Thawing & infusion

6 clinical trials
4 in bone marrow transplantation
1 in liver/kidney transplantation
1 in auto-immune diseases