Mesenchymal Stromal Cells for Acute Respiratory distress syndrome and severe sepsis

Adomas Bukauskas

Hematology oncology and transfusiology center
Vilnius University Santariskiu Klinikos
ARDS: Rationale

• **ARDS** is **major** cause of **acute respiratory failure** in critically ill patients.

• **Mortality** associated with ARDS **20-40%** (etiology pneumonia, sepsis, aspiration, trauma)

• Current treatments: **supportive care**
  • Lung protective ventilation,
  • Fluid conservative strategy,
  • Prone positioning.
  • **No pharmacological therapies** from preclinical models have yet been translated to effective clinical treatment options.
### Table 3. The Berlin Definition of Acute Respiratory Distress Syndrome

<table>
<thead>
<tr>
<th>Acute Respiratory Distress Syndrome</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Timing</strong></td>
</tr>
<tr>
<td><strong>Chest imaging</strong>&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td><strong>Origin of edema</strong></td>
</tr>
<tr>
<td><strong>Oxygenation</strong>&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Mild</td>
</tr>
<tr>
<td>Moderate</td>
</tr>
<tr>
<td>Severe</td>
</tr>
</tbody>
</table>

Abbreviations: CPAP, continuous positive airway pressure; FIO₂, fraction of inspired oxygen; PaO₂, partial pressure of arterial oxygen; PEEP, positive end-expiratory pressure.

<sup>a</sup>Chest radiograph or computed tomography scan.

<sup>b</sup>If altitude is higher than 1000 m, the correction factor should be calculated as follows: $[\text{PaO}_2/\text{FiO}_2 \times (\text{barometric pressure}/760)]$.

<sup>c</sup>This may be delivered noninvasively in the mild acute respiratory distress syndrome group.
Pathophysiology

M. Rojas et al, Mesenchymal Stem Cells: A Promising Therapy for the Acute Respiratory Distress Syndrome
Mechanisms of MSC effect in lung injury

• Ability to actively respond to the local environment
  • **Engraftment**
    • Engraftment of MSCs into the lung epithelium is documented - rare event in the context of lung injury with reports of less than 5% engraftment occurring, it is likely this does not represent the primary mechanism of their effect.
  • **Paracrine factors**
    • *Keratinocyte growth factor (KGF)* - essential for the restoration of alveolar epithelium permeability and alveolar fluid clearance after injury by rescuing the activity of the epithelial sodium channel.
    • *Angiopoietin-1* - responsible for the MSC protective effects on type II alveolar epithelial cell permeability in an in vitro model
    • The antimicrobial effect of MSCs is partially explained by their ability to enhance phagocytosis by cells of the innate immune system. Additional antimicrobial activity is exerted by MSCs through the secretion of antimicrobial peptides and proteins
  • **Nanotubule formation and microvesicle secretion**
    • the ability of MSCs to transfer mitochondria to diverse cell types including endothelial cells
Pathophysiology

M. Rojas et al, Mesenchymal Stem Cells: A Promising Therapy for the Acute Respiratory Distress Syndrome
2 clinical trials

Treatment of acute respiratory distress syndrome with allogeneic adipose-derived mesenchymal stem cells: a randomized, placebo-controlled pilot study.

Mesenchymal stem (stromal) cells for treatment of ARDS: a phase 1 clinical trial.
Mesenchymal stem (stromal) cells for treatment of ARDS: a phase 1 clinical trial

Jennifer G Wilson, Kathleen D Liu, Hanjing Zhuo, Lizette Caballero, Melanie McMillan, Xiaohui Fang, Katherine Cosgrove, Rosemary Vojnik, Carolyn S Calfee, Jae-Woo Lee, Angela J Rogers, Joseph Levitt, Jean Wiener-Kronish, Ednan K Bajwa, Andrew Leavitt, David McKenna, B Taylor Thompson, Michael A Matthay
Methods

• The STem cells for ARDS Treatment (START) trial
• Multicentre (3 study sites), open-label, phase 1 clinical trial.
• July 8, 2013, - Jan 13, 2014.
### Inclusion criteria

1. Positive pressure ventilation by an endotracheal or tracheal tube with a \( \text{PaO}_2 : \text{FiO}_2 \) \(< 200 \text{ mm Hg} \) with at least 8 cm H\(_2\)O positive end-expiratory airway pressure
2. Bilateral infiltrates consistent with pulmonary oedema on frontal chest radiograph
3. No clinical evidence of left atrial hypertension, or if measured, a pulmonary arterial occlusion pressure \( \leq 18 \text{ mm Hg} \)
4. Criteria 1–3 must all be present within a 24 h time period and at the time of enrolment

### Exclusion criteria

1. Age younger than 18 years
2. \( > 96 \text{ h since first meeting acute respiratory distress syndrome criteria per the Berlin definition} \)
3. Pregnant or breastfeeding
4. Prisoner
5. Presence of any active malignancy (other than non-melanoma skin cancer) that required treatment within the past 2 years
6. Any other irreversible disease or condition for which 6-month mortality is estimated to be \( > 50\% \)
7. Moderate to severe liver failure (Child-Pugh score \( > 12 \))
8. Severe chronic respiratory disease with a \( \text{PaCO}_2 \) \( > 50 \text{ mm Hg} \) or the use of home oxygen
9. Patient, surrogate, or physician not committed to full support (exception: a patient will not be excluded if he or she would receive all supportive care except for attempts at resuscitation from cardiac arrest)
10. Major trauma in the previous 5 days
11. Lung transplant patient
12. No consent or inability to obtain consent
13. Moribund patient not expected to survive 24 h
14. WHO class III or IV pulmonary hypertension
15. Documented deep venous thrombosis or pulmonary embolism within past 3 months
16. No arterial line or no intent to place an arterial line
17. No intent or unwillingness to follow lung protective ventilation strategy or fluid management protocol
18. Currently receiving extracorporeal life support or high-frequency oscillatory ventilation
Methods

- MSC initiated within 120 h within meeting Berlin criteria
- IAS pts or appropriate surrogate
- 2 h period of bedside observation of haemodynamic and respiratory parameters was initiated to ensure that the patient was stable before the MSC infusion.
- Stability:
  - transcutaneous oxygen saturation in the target range of 88–95% without any increase in ventilator settings
  - stable use of vasopressor if the patient required vasopressors for blood pressure support minimal increase

- Procedure:
  - 3 low dose MSCs (1 million cells/kg PBW);
  - 3 patients intermediate dose MSCs (5 million cells/kg PBW);
  - 3 patients were assigned to receive high dose MSCs (10 million cells/kg PBW).
  - The total volume of the MSC infusion was 100 mL regardless of dose. The viability ranged from 50–63% (mean 56%).
  - The cells were infused via gravity over roughly 60–80 min
Aims

• Primary:
  • Safety

• Secondary:
  • LIS,
  • SOFA score
  • Acute Physiology and Chronic Health Evaluation (APACHE) score,
  • Duration of mechanical ventilation,
  • Ventilator-free days,
  • Duration of vasopressor and intensive care unit [ICU]-free days,
  • Biomarker values.
## Results

<table>
<thead>
<tr>
<th>Age (years)</th>
<th>Sex</th>
<th>APACHE III</th>
<th>Primary cause of ARDS</th>
<th>Tidal volume (mL/kg PBW)</th>
<th>Plateau pressure (cm H$_2$O)</th>
<th>PEEP (cm H$_2$O)</th>
<th>PaO$_2$/FiO$_2$ (mmHg)</th>
<th>Lung injury score</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 million cells/kg PBW</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Patient 1</td>
<td>29</td>
<td>Female</td>
<td>81</td>
<td>Pre-eclampsia</td>
<td>7.0</td>
<td>28</td>
<td>10</td>
<td>173</td>
</tr>
<tr>
<td>Patient 2</td>
<td>86</td>
<td>Female</td>
<td>121</td>
<td>Pneumonia</td>
<td>6.6</td>
<td>31</td>
<td>10</td>
<td>101</td>
</tr>
<tr>
<td>Patient 3</td>
<td>59</td>
<td>Female</td>
<td>130</td>
<td>Aspiration</td>
<td>6.0</td>
<td>25</td>
<td>10</td>
<td>168</td>
</tr>
<tr>
<td>5 million cells/kg PBW</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Patient 4</td>
<td>67</td>
<td>Female</td>
<td>133</td>
<td>Aspiration</td>
<td>6.3</td>
<td>21</td>
<td>10</td>
<td>105</td>
</tr>
<tr>
<td>Patient 5</td>
<td>62</td>
<td>Female</td>
<td>109</td>
<td>Pneumonia</td>
<td>5.6</td>
<td>20</td>
<td>14</td>
<td>111</td>
</tr>
<tr>
<td>Patient 6</td>
<td>46</td>
<td>Female</td>
<td>83</td>
<td>Aspiration</td>
<td>6.0</td>
<td>19</td>
<td>10</td>
<td>153</td>
</tr>
<tr>
<td>10 million cells/kg PBW</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Patient 7</td>
<td>52</td>
<td>Male</td>
<td>121</td>
<td>Pneumonia</td>
<td>7.1</td>
<td>23</td>
<td>10</td>
<td>154</td>
</tr>
<tr>
<td>Patient 8</td>
<td>55</td>
<td>Female</td>
<td>127</td>
<td>Sepsis (biliary)</td>
<td>5.9</td>
<td>34</td>
<td>10</td>
<td>194</td>
</tr>
<tr>
<td>Patient 9</td>
<td>38</td>
<td>Male</td>
<td>68</td>
<td>Pneumonia</td>
<td>6.0</td>
<td>Not measured</td>
<td>8</td>
<td>118</td>
</tr>
</tbody>
</table>

APACHE=Acute Physiology and Chronic Health Evaluation. ARDS=acute respiratory distress syndrome. PBW=predicted bodyweight. PEEP=positive end-expiratory pressure.
Results

- No prespecified infusion-associated AE
- Specifically, no significant changes in heart rate, mean arterial pressure, or oxygen saturation were reported in any of the three dosing groups during the infusion or in the immediate post-infusion period
Results

Figure 2: Lung injury score (LIS)
Mean (SD) LIS for each dosing group at baseline, 6 h from start of mesenchymal stem-cell infusion, and study days 1, 2, and 3. LIS is calculated from four variables: (1) number of affected quadrants on chest radiograph; (2) severity of hypoxia as measured by PaO₂:FiO₂; (3) level of positive end-expiratory pressure; and (4) the static compliance of respiratory system.²⁷

Figure 3: Sequential Organ Failure Assessment (SOFA) score
Mean (SD) SOFA score for each dosing group at baseline, 6 h from start of mesenchymal stem cell infusion, and study days 1, 2, and 3. The SOFA score quantifies the severity of organ dysfunction in six systems (respiratory, coagulation, hepatic, cardiovascular, renal, and neurological), and predicts outcomes in critically ill patients.²⁸²⁹
## Results

<table>
<thead>
<tr>
<th></th>
<th>Duration of mechanical ventilation (days)</th>
<th>Ventilator-free days (up to day 28)</th>
<th>Oxygenation index (day 3)</th>
<th>Duration of vasopressor use (days)</th>
<th>Intensive care unit-free days (up to day 28)</th>
<th>Vital status and day of discharge</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Low dose</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>5</td>
<td>24</td>
<td>2.33</td>
<td>0</td>
<td>24</td>
<td>Alive, day 8</td>
</tr>
<tr>
<td>2</td>
<td>10</td>
<td>0</td>
<td>13.36</td>
<td>20</td>
<td>0</td>
<td>Dead, day 9</td>
</tr>
<tr>
<td>3</td>
<td>11</td>
<td>18</td>
<td>5.78</td>
<td>4</td>
<td>14</td>
<td>Alive, day 22</td>
</tr>
<tr>
<td><strong>Intermediate dose</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>7</td>
<td>22</td>
<td>4.63</td>
<td>0</td>
<td>21</td>
<td>Alive, day 34</td>
</tr>
<tr>
<td>5</td>
<td>31</td>
<td>0</td>
<td>5.36</td>
<td>2</td>
<td>0</td>
<td>Dead, day 31</td>
</tr>
<tr>
<td>6</td>
<td>3</td>
<td>27</td>
<td>&quot;</td>
<td>3</td>
<td>26</td>
<td>Alive, day 5</td>
</tr>
<tr>
<td><strong>High dose</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>3</td>
<td>26</td>
<td>10</td>
<td>0</td>
<td>22</td>
<td>Alive, day 7</td>
</tr>
<tr>
<td>8</td>
<td>17</td>
<td>12</td>
<td>&quot;</td>
<td>0</td>
<td>9</td>
<td>Alive, day 25</td>
</tr>
<tr>
<td>9</td>
<td>19</td>
<td>20</td>
<td>6.99</td>
<td>0</td>
<td>18</td>
<td>Alive, day 14</td>
</tr>
</tbody>
</table>

Oxygenation index=FIO₂ x mean airway pressure/PaO₂, *Extubated.

*Table 3: Secondary respiratory and systemic results by dosing cohort*
Results

- Median concentrations of all four biomarkers declined between baseline and day 3.
- No significant differences in the magnitude of decline among groups for any of the biomarkers were noted.
Discussion

• SAFE: all three doses of MSCs are safe in patients with moderate-to-severe ARDS
  • no infusion-associated adverse events, immediate clinical instability, or dose-limiting toxicity at any of the doses tested
  • none of the SAE were related to MSC infusion.
• Mortality 22% (MSC) vs 32% (historical control)
• More is better
  • The favourable changes observed in LIS and SOFA score with the high dose of MSCs (10 million cells/kg PBW) compared with both reduced doses are consistent with the hypothesis that increased doses of MSCs might provide increased clinical benefit.
• Without a matched control group, we cannot conclude that the recorded changes in biomarkers were related to MSC therapy.
• First and foremost, with only nine patients, we can neither generalise our phase 1 experience, nor draw conclusions about either the efficacy or long-term safety of MSCs for ARDS.
Future directions

• Safe
• Well tolerated
• No SAE related to MSC up to 6 months
• A randomised, double-blind, placebo-controlled phase 2 clinical trial of 10 million MSCs/kg PBW in 60 patients with moderate- to-severe ARDS, with a primary focus on safety and secondary outcomes including respiratory, systemic, and biological endpoints.
12 adult pts meeting the Berlin definition of ARDS (PaO2/FiO2 ratio of < 200)

MSCs vs placebo (saline) 1 :1.

Dose – single iv dose of 1 × 10⁶ cells/kg of body weight.

Side effects were monitored after treatment.

Treatment of acute respiratory distress syndrome with allogeneic adipose-derived mesenchymal stem cells: a randomized, placebo-controlled pilot study.


Table 3 Hospital indices by treatment group

<table>
<thead>
<tr>
<th></th>
<th>MSCs group</th>
<th>Placebo group</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Days in hospital</td>
<td>33.7 ± 6.3</td>
<td>27.8 ± 5.0</td>
<td>0.10</td>
</tr>
<tr>
<td>ICU-free days at study day 28</td>
<td>4.0 ± 8.2</td>
<td>4.3 ± 5.7</td>
<td>0.94</td>
</tr>
<tr>
<td>Ventilator-free days at study day 28</td>
<td>11.2 ± 11.5</td>
<td>7.3 ± 7.8</td>
<td>0.52</td>
</tr>
</tbody>
</table>

Data are presented as mean ± SD. p values were calculated using Student's t-test.
• Randomization >48 hours > one dose of $1 \times 10^6$ cells/kg body weight or saline as a single intravenous infusion over 60 minutes.

• Study drugs were well tolerated.

• No adverse events were recorded during infusions. One patient from each group presented with diarrhea one day after study drug treatment and resolved within 48 hours.

• One patient in the MSCs group developed rash in the chest area after the infusion and resolved spontaneously over 24 hours.

• During the study period, one patient in the MSCs group died of multiple organ failure.

• Deaths occurred in two patients in the placebo group with one multiple organ failure and the other sepsis.

• None of the deaths were considered to be related to the study drugs by the clinical investigators and were consistent with the patients’ existing disease processes.

• All the remaining patients completed the 28-day follow-up period. There were no other adverse events or serious adverse events.
<table>
<thead>
<tr>
<th>Rank</th>
<th>Status</th>
<th>Study</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Recruiting</td>
<td>Human Mesenchymal Stem Cells For Acute Respiratory Distress Syndrome</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Condition:</strong> Acute Respiratory Distress Syndrome</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Intervention:</strong> Biological: Allogeneic Bone Marrow-Derived Human Mesenchymal Stem Cells</td>
</tr>
<tr>
<td>2</td>
<td>Recruiting</td>
<td>Human Mesenchymal Stem Cells For Acute Respiratory Distress Syndrome (START)</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Condition:</strong> Respiratory Distress Syndrome, Adult</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Interventions:</strong> Biological: Allogeneic Bone Marrow-Derived Human Mesenchymal Stem Cells; Biological: Plasma-Lyte A</td>
</tr>
<tr>
<td>3</td>
<td>Recruiting</td>
<td>Treatment of Severe Acute Respiratory Distress Syndrome With Allogeneic Bone Marrow-derived Mesenchymal Stromal Cells</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Condition:</strong> Acute Respiratory Distress Syndrome, Adult</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Intervention:</strong> Biological: Mesenchymal stromal cells</td>
</tr>
<tr>
<td>4</td>
<td>Recruiting</td>
<td>Human Umbilical-Cord-Derived Mesenchymal Stem Cell Therapy in Acute Lung Injury</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Conditions:</strong> Acute Lung Injury; Acute Respiratory Distress Syndrome</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Intervention:</strong> Biological: UCMSC group</td>
</tr>
<tr>
<td>5</td>
<td>Unknown</td>
<td>Adipose-derived Mesenchymal Stem Cells in Acute Respiratory Distress Syndrome</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Condition:</strong> ARDS</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Interventions:</strong> Drug: Mesenchymal stem cells; Drug: Placebo</td>
</tr>
<tr>
<td>6</td>
<td>Recruiting</td>
<td>Mesenchymal Stem Cell In Patients With Acute Severe Respiratory Failure</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Condition:</strong> Respiratory Distress Syndrome, Adult</td>
</tr>
<tr>
<td></td>
<td></td>
<td><strong>Intervention:</strong> Biological: Mesenchymal Stem Cell</td>
</tr>
</tbody>
</table>
2220 The Results of the Russian Clinical Trial of Mesenchymal Stromal Cells (MSCs) in Severe Neutropenic Patients (pts) with Septic Shock (SS) (RUMCESS trial)

Lymphocytes, Lymphocyte Activation and Immunodeficiency, including HIV and Other Infections
Program: Oral and Poster Abstracts
Session: 203. Lymphocytes, Lymphocyte Activation and Immunodeficiency, including HIV and Other Infections: Poster II

Sunday, December 6, 2015, 6:00 PM-8:00 PM
Hall A, Level 2 (Orange County Convention Center)

Gennadii M. Galstian, MD, PhD¹*, Elena N. Parovichnikova, MD, PhD²*, Polina M. Makarova, MD¹*, Larisa A. Kuzmina, MD, PhD³*, Vera V. Troitskaya, MD, PhD²*, Eduard Gemdzhian, PhD⁴*, Nina I. Drize, PhD⁵ and Valeri G. Savchenko, MD, PhD, Prof.²

¹ICU, National Research Center for Hematology, Moscow, Russia
²National Research Center for Hematology, Moscow, Russia
³Department of bone marrow transplantation, National Research Center for Hematology, Moscow, Russia
⁴Biostatistics, National Research Center for Hematology, Moscow, Russia
⁵Lab for Physiology of Hematopoiesis, National Research Center for Hematology, Moscow, Russia
Severe sepsis

- No study has investigated the effects of MSC therapy on the survival of pts with sepsis and SS, especially severe-neutropenic pts.
- Aim: Efficacy of the administration of MSCs for the treatment of SS in neutropenic pts.
- Patients and Methods: prospective, single-center, randomized Russian clinical trial of MSCs in severe neutropenic pts with SS (RUMCESS) (NCT 01849237) since December 2012. Neutropenic pts (WBC < 0.5x10⁹/l) with SS were enrolled on to the study.
  - The pts were randomly assigned (1:1) to receive either conventional therapy (CT) of SS (CT group), or CT plus donor MSCs at a dose of 10⁶/kg intravenously within the first 10 hours after SS onset (CT+MSCs group).
  - All pts were admitted and treated in the ICU of the National Research Center for Hematology (Moscow).
  - The Acute Physiology and Chronic Health Evaluation (APACHE) II score and
Severe sepsis

- **Results:** 27 neutropenic pts with SS, 13 CT vs 14 CT+MSCs. There were no statistically significant differences in the demographic variables between groups.
- The CT group included 7 males, 6 females, aged 33-81 yrs, median 55 yrs.
- The CT+MSCs group included 6 males, 8 females, aged 30-75 yrs, median 48 yrs.
- Hematological disorders were also similar in the two groups: AML (4), NHL (4), HL (1), MM (3), MDS (1) in the CT group, and AML (5), NHL (7), MM (1) in the CT+MSCs group.
- In all pts, except for one with MDS, neutropenia developed after chemotherapy.
- In **8/13 pts** in the CT group and **9/14 pts** in the CT+MSCs group blood cultures were found positive, mostly gram-negative.
Baseline APACHE II scores (34.2 [95% CI 28.3-43.1] and 32.2 [95% CI 26.2-37.5] in the CT- and CT+MSC-groups, respectively), and SOFA scores (17.9 [95% CI 13.5-22.2] and 15.1 [95% CI 11.0-19.2] respectively), were similar in the two groups.

**28-day survival** rates were 15% (2 out of 13 pts) in the CT group and 57% (8 out of 14 pts) in the CT+MSCs group (P=0.04) (Figure 1).

The significant increase in 28 days OS of the pts in CT+MCSs group was associated with SOFA score decrease, which was started in three days after onset of SS.

Despite higher 28-day survival rates only 3 pts treated with CT+MSCs remained alive after 3 months, and 5 of 8 pts from the CT+MSCs-group who survived 28 days died later because of sepsis-related organ dysfunction.

**Conclusions:** Administration of MSCs in the first hours of SS might improve short-term survival in neutropenic pts, but **does not prevent death from sepsis-related organ dysfunction in the long term.** Perhaps repeated administration of MSC is required.
Mesenchymal stem cells as a therapeutic tool to treat sepsis

Eleuterio Lombardo, Tom van der Poll, Olga DelaRosa, Wilfried Dalemans
Idea

• Sepsis is a clinical syndrome caused by a deregulated host response to an infection.

• Sepsis is the most frequent cause of death in hospitalized patients.

• Sepsis will remain an important clinical problem in the future, especially in light of the ageing population and emerging antibiotic resistance.

• Based on their immunomodulatory properties, adult mesenchymal stem or stromal cells (MSCs) can be a novel therapeutic tool to treat sepsis.
Epidemiology

• Sepsis a leading cause of death, and the most frequent cause of death in non-coronary intensive care units (ICUs) in the developed world.

• In the United States the yearly incidence of severe sepsis is estimated at 300 cases per 100000 person-years population, which accounts for 10% of all ICU admissions.

• Mayr et al have recently reported that the mortality of severe sepsis and septic shock lies between 25%-50%, with the extent and number of organ failures as the strongest predictors of an adverse outcome.

Epidemiology

• The most common sources of sepsis are: pneumonia, intra-abdominal-, urinary tract- and soft tissue infections

• Blood cultures are positive in 1/3, and up to 1/3 are culture negative from all body sites.

• The most commonly isolated Gram-positive bacterial pathogens are Staphylococcus aureus and Streptococcus pneumoniae, and the most common Gram-negative pathogens are Escherichia coli, Klebsiella spp, and Pseudomonas aeruginosa.

• A recent study encompassing 14000 ICU patients in 75 countries found that 62% of positive isolates were Gram-negative bacteria, vs 47% Gram-positive and 19% fungal

Pathophysiology

- Sepsis occurs when the body’s response to infection injures the host’s tissues and organs.
- Immune cells can sense pathogens via so-called pattern-recognition receptors (PRRs), which recognize conserved motifs expressed by microorganisms called pathogen-associated molecular patterns or PAMPs.
- Four classes of PRRs have been identified: Toll-like receptors (TLRs), C-type lectin receptors, RIG-I-like receptors and NOD-like receptors.
- Activation of PRRs by PAMPs causes upregulation of inflammatory gene transcription and initiation of innate immunity, a response aimed at eliminating the invading pathogen.
- However, when bacteria overcome the ability of the innate immune system to clear the infection, resulting in progression to sepsis, the interactions between pathogens and PRRs advances into a deregulated response that no longer benefits the host.
- During such injurious host response inflammation can be perpetuated by stimulation of PRRs by so-called danger-associated molecular patterns (DAMPs or alarmins), which are endogenous molecules released by injured or dying cells.
- Alarmins are also released during sterile injury such as after trauma or severe pancreatitis, which contributes to the concept that the pathogenesis of multiple organ failure in sepsis and non-infectious critical illness is not fundamentally different.
• Cytokines are an important component of the “hyperinflammatory” response to severe infection. Experimental sepsis induced by systemic challenge with high bacterial doses is associated with enhanced release of multiple cytokines, and elimination or inhibition of several of these proinflammatory mediators [including tumor necrosis factor (TNF)-α, interleukin (IL)-1β, IL-12, IL-17, IL-18, interferon-γ, and macrophage migration inhibitory factor] improves survival in these models.

• However and importantly, these systemic challenge models do not adequately mimic the clinical syndrome of sepsis. Many trials evaluating the efficacy of proinflammatory cytokine inhibition, especially targeting TNF-α and IL-1, or other anti-inflammatory strategies have failed. Other proinflammatory mediators implicated in sepsis pathogenesis include high mobility group box 1 (HMGB1) and S100 proteins.
• Activation of the complement systems forms a fundamental part of the innate immune response to infection.

• Sepsis is associated with systemic activation of the complement system, which can be harmful in the setting of fulminant sepsis. Indeed, neutralization or genetic absence of complement factor C5a and its receptors results in increased survival during abdominal sepsis or endotoxemia in mice.

• Other hallmark features of the sepsis host response include activation of the coagulation system and vascular dysfunction. The most severe manifestation of coagulopathy is the syndrome of disseminated intravascular coagulation, with an estimated incidence between 30%-50% in severe sepsis, caused by tissue factor-driven activation of coagulation with concurrent impairment of anticoagulant and fibrinolytic mechanisms.

• Organ dysfunction in sepsis is at least in part caused by tissue hypoperfusion, secondary to hypotension, microvascular thrombosis and/or dysfunction of the vascular endothelium with loss of barrier function. Mitochondrial dysfunction and altered cellular bioenergetics have been implicated in sepsis-induced organ dysfunction, although further research is warranted to establish a causal relationship.
MSC

- MSCs are considered a promising tool for cell therapy, in particular for inflammatory diseases, based on their immunomodulatory properties and paracrine effects through trophic factors with anti-fibrotic, anti-apoptotic or pro-angiogenic properties.
- MSCs regulate the function of a broad range of immune cells and are activated by inflammatory mediators released from activated immune cells (i.e., IFNγ, IL1β and TNFα).
- The mechanisms involved in the immunoregulatory activity of MSCs are still under investigation but rely on both cell contact-dependent mechanisms (i.e., Jagged1-Notch1 interaction, Fas-Fas-L interaction) and paracrine effects through the release of soluble factors including hepatocyte growth factor, prostaglandin-E2 (PGE2), TGF-β1, nitric oxide (NO), IL-10, IL-6, heme oxygenase-1 (HO-1), HLA-G5 or the enzymatic activity of indoleamine 2,3-dioxygenase.
- In addition to the direct effect of these soluble factors, MSCs may also modulate immune responses through the generation of immune cells with regulatory phenotype, including regulatory T cells or anti-inflammatory macrophages.
• MSCs have also been reported to show antimicrobial activities against different pathogens upon activation with inflammatory cytokines.

• Noteworthy in the context of sepsis, the functionality of MSCs can also be modulated by activators of TLRs.

• It has been described that MSCs can be polarized in vitro towards either anti-inflammatory or pro-inflammatory phenotypes, depending on the TLR ligand time/concentration used for activation.

• Furthermore, it has been recently described that interaction of gastrointestinal bacteria (Salmonella typhimurium or Lactobacillus acidophilus) with MSCs increased their capacity to inhibit T lymphocyte proliferation in vitro through a PGE2-dependent mechanism, indicating that bacteria may also enhance the immunomodulating properties of MSCs.

• MSCs can sense inflammatory signals through the expression of cytokine/chemokine receptors and integrins, and subsequently migrate to sites of inflammation. Moreover, homing of systemically administered MSCs to lymphoid organs (draining lymph nodes and spleen) and the subsequent generation of functional Tregs have also been reported.

• MSCs do not long-term engraft at the inflammation site and cells seem to be cleared shortly after administration. This suggests that transient effects through soluble factors and cell-to-cell contacts play a main role in MSC-mediated initial controlling and balancing of local inflammation.
• Allogeneic MSCs are regarded as a preferred source for treatment as they would allow treatment with a ready to use, off-the-shelf product, available for a large number of patients, specially, in acute life threatening indications like sepsis in which isolation and expansion of autologous MSCs is not an option.

• In that context, MSCs are considered immune privileged as they express constitutively only low levels of cell-surface HLA class I molecules and lack expression of HLA class II, CD40, CD80 and CD86 which would lead to reduced activation of the innate and adaptive immune responses.

• This immune privilege of MSCs therefore supports the feasibility of allogeneic treatments without the requirement of suppression of host immunity. However the immunogenic features of MSCs are currently under review as there is some evidence of immunogenicity in experimental animal models that coincides with immunomodulatory effects by MSCs.
Conclusions

• The use of MSCs in experimental animal models of sepsis has reported strong evidence of the therapeutic potential of MSC therapy in this indication. These studies have been mainly focused on the effects of MSCs on the pro-inflammatory phase of sepsis, while the effects of MSCs on the subsequent anti-inflammatory/immune exhaustion phase of the disease has not been elucidated so far and will need further investigation.

• The mechanisms by which MSCs improve survival in sepsis models rely on the collective effects of their immunomodulatory and anti-microbial properties: MSC treatment modulates inflammation in septic mice by a mechanism that requires the reprogramming of macrophages towards a more anti-inflammatory phenotype (release of anti-inflammatory IL10), resulting in reduced levels of pro-inflammatory cytokines in blood and organs and attenuated infiltration of immune cells in infected tissues (monocytes and neutrophils).

• Moreover, MSCs show direct (release of LL-37 peptide) and indirect (increase of phagocytic properties of monocyte/macrophages and neutrophils) anti-microbial effects. The combined effect of reducing both the inflammatory response and the bacterial burden results in an improvement of organ function and higher survival rates.

• The promising results obtained in these, small animal, preclinical efficacy studies are encouraging and suggest that MSCs might be a therapeutic option to treat sepsis in patients. Importantly, efficacy of MSCs in large animal models that better replicate the inflammatory response, organ failure and disease in humans (e.g., sheep models) will be additionally relevant to support further testing of the therapeutic potential of allogeneic MSC treatment in humans.

• Such clinical trials should be prospective, controlled, and randomized so to guarantee a clear outcome of the MSC treatment effect. Moreover, taking into consideration the complexity and heterogeneity of sepsis and the poor results up to now in sepsis clinical trials, we believe that such trials should first be done in well defined and homogeneous sepsis patient populations.