Marrow Cellution™

Autologous Bone Marrow Aspiration & Cancellous Bone Graft Harvesting
Traditional Trocar Design & Technique

The path of least resistance is the physical pathway that provides the least resistance to motion by a given object or entity, among a set of alternative paths.

- Designed to perform a **single small volume pull (1-2mL)** from the distance most proximal from the entry of the needle.
- **Larger volumes** of bone marrow aspirate contain **higher amounts of peripheral blood** as the cannula is open ended.
- Aspirating after **retracting the needle exacerbates the problem** of peripheral blood contamination by exposing the open ended cannula to the resulting channel.
- **Side Port Fallacy:** Integration of side ports on traditional needles are ineffective due to the stronger forces associated with aspiration from distal end blocking side ports from within the lumen of the needle.
Traditional Bone Marrow Aspiration Techniques

Open End (Distal) Trocar with Side Port Fallacy

The regenerative qualities of bone marrow have been used for many decades and are considered the gold standard for stem cell harvesting.

Traditional open ended (distal) trocar designed aspiration needles operate optimally for small biopsy volumes of 1-2mL. After aspirating the first 1-2mL of bone marrow, peripheral blood will preferentially fill the vacated space, limiting the additional harvest of key stem and progenitor cells.

Further aspiration attempts diminish the number of total nucleated cells (TNC) in the aspirate drops dramatically due to the lower viscosity of blood following the path of least resistance through the distal end channel, minimizing efficiency of side channels.

Aspiration of larger quantities of bone marrow, typically required for most clinical indications, necessitate further manipulation and volume reduction processing steps such as, centrifugation systems or chemical gradient separation in a laboratory.
Marrow Cellution™ Solution

Marrow Cellution™ Overcomes Limitations & Maximizes Cell Yield

The unique patent pending techniques of implementing a closed end catheter through an introducer sheath overcomes distal end peripheral blood contamination.

The short sharp trocar introducer allows for easy access through soft tissue and cortical bone. A blunt trocar is then introduced to make access for closed end side port aspiration cannula. The design minimizes trauma to cancellous bone and marrow, thereby mitigating pooling of peripheral blood.

The patent pending design of the closed end catheter forces aspiration of marrow from lateral marrow space. The manual rotation of the handles allows the cannula to be raised to a desired position in a new level of undisturbed marrow for subsequent aspiration aliquots.

The Marrow Cellution™ is able to collect up to 10mL of high quality marrow equivalent or superior to other systems that require additional manipulation steps such as centrifugation or chemical separation in a laboratory.
Marrow Cellution™

Overcome Limitations & Maximize Cell Yield

- Marrow Cellution is a novel bone marrow access and retrieval device that incorporates unique features designed to minimize the limitations of traditional trocar needles.

- Aspirate flow is collected exclusively laterally as the tip of the aspiration cannula is closed allowing marrow collection perpendicular to and around the channel created by the tip of the device.

- Marrow Cellution achieves multiple small volumes of high quality bone marrow aspirate collected from various sites distributed within the marrow cavity.

- A single puncture with Marrow Cellution is functionally equivalent to repeated puncture sites with a traditional trocar needle collecting small aspirate volumes, but with substantial savings of time, effort, reduced patient trauma, morbidity and risk of infection.

Patent Pending Design
Four channel, closed tipped, aspirating cannula prevents exposure of the needle tip to the channel filled with peripheral blood created by the needle as it is being retracted from the bone space.
Overcoming Limitations

Overcome Limitations & Maximize Cell Yield

The innovative Marrow Cellution™ System allows for specific aspiration to eliminate peripheral blood contamination and thereby significantly improving cellular yield performance.

**PERFORMANCE**
- High Quality – Low Volume
- Higher CFU counts per mL
- Additional steps not required
- No Anticoagulant Contamination

**INNOVATIVE**
- Reduces peripheral blood aspiration
- Closed-end aspiration design
- Cannula via sheath technology
- Novel patent pending design

**SPECIFIC**
- Minimally invasive
- Minimizes OR Time
- Maximizes Sterility Conditions
- Low volume
- High yield
- Reduces Biologic Utilization Costs

Aspiration of larger quantities of bone marrow, typically required for most clinical indications, necessitate further manipulation and volume processing steps such as, centrifugation systems or chemical gradient separation in a laboratory.

The Marrow Cellution™ System is able to collect up to 10mL from each puncture site of high quality marrow equivalent or superior to other techniques that require additional manipulation steps such as centrifugation or chemical separation in a laboratory.
Marrow Cellution

Essential Healing Factors

Impaired Angiogenesis Results in Impaired Healing.

Creating a rich microenvironment with vascular sufficiency is a critical, well established first step in bone formation.
Autologous Bone Marrow Aspiration & Bone Graft Harvesting

Marrow Cellution

Overcome Limitations & Maximize Cell Yields

- Maximizes Cell Yield
- Minimally Invasive
- Centrifugation Not Required
- Never Leaves the Sterile Field
- Reduces Blood Contamination
- Regulatory Compliant
- Reduces Donor Site Morbidity
- 100% Natural

Unsurpassed Cell Collection

Cancellous Graft Collection

Fusion vs. Failure
“The mechanism of action in bone healing points to the hierarchical role of creating a vascular network before bone can be formed”

Creating a microenvironment with vascular sufficiency is a critical first step in bone formation since impaired angiogenesis results in impaired bone formation.

**Bone Healing Biology**

**Bone Grafting Procedure**

<table>
<thead>
<tr>
<th>Bone Grafting Procedure</th>
<th>=</th>
<th>Autologous Bone Graft Harvest &amp; Transplantation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Marrow Cellution</td>
<td>=</td>
<td>Minimally Invasive Bone Graft Composition (Liquid &amp; Cancellous)</td>
</tr>
</tbody>
</table>
Marrow Cellution
Subchondral Bone Healing

Seeding
Loading
Shaping
Positioning
Consolidation

Marrow Cellution: Striking Advantages
Autologous Cell Collection & Cancellous Bone Grafting

The functional design of the Marrow Cellution™ System includes two unique features: a **Closed Needle Tip** to prevent aspiration of excess blood from the entry channel and a **Handle With Threaded Guide** for controlled movement of the aspiration cannula within the marrow space.

The MC-RAN-8C Marrow Cellution™ System provides the additional benefit to **Percutaneously Harvest Bone Graft** in the same minimally invasive procedure. Thereby, reducing donor site morbidity.
**Marrow Cellution™**

**Marrow Cellution™** Bone Marrow Aspiration System (MC-RAN-11C).

Allows for measured and controlled aspiration over a large geography within the marrow space, while **restricting peripheral blood infiltration**.

**Marrow Cellution™** Bone Marrow Aspiration- & Autologous Bone Harvesting System (MC-RAN-8C).

Allows for the combination of high quality **bone marrow aspirate** and percutaneously harvested **cancellous bone autograft**.

"This is potentially a giant step in bone marrow processing. This needle will usher in a new age in bone marrow aspiration."

Dr. Joseph Purita, M.D.
Orthopedic Surgeon, Boca Raton/FL
Marrow Cellution™ Product Details

**MC-RAN-11C**
Item #: 74219-06M
Effective Length: 3.5" (9cm)

Components:
- 11 Gauge Introducer Cannula & Sharp Stylet
- 11 Gauge Introducer Blunt Stylet
- 14 Gauge Aspiration Cannula
- 10mL Syringe

**MC-RAN-11C STS (OBESE PTS.)**
Item #: 74219-07M
Effective Length: 4.5" (11.4cm)

Components:
- 11 Gauge Introducer Cannula & Sharp Stylet
- 11 Gauge Introducer Blunt Stylet
- 14 Gauge Aspiration Cannula
- 10mL Syringe

**MC-RAN-8C**
Item #: 74266-01M
Effective Length: 3.5" (9cm)

Components:
- all MC-RAN-11C components and
- 8G x 4" Swaged Tip Introducer Needle
- Measurement Probe
- Cancellous Bone Dowel Extraction Tool

**MC-RAN-8C STS (OBESE PTS.)**
Item #: 74266-04M
Effective Length: 4.5" (11.4cm)

Components:
- all MC-RAN-11C components and
- 8G x 6" Swaged Tip Introducer Needle
- Measurement Probe
- Cancellous Bone Dowel Extraction Tool
Component Flushing (Rinsing) Instructions

- Withdraw approximately 5-7mL of Heparin Solution (2,000 units/mL) into 10mL syringe
- Remove Stylets from Introducer Needle and Aspiration Cannula with distal end of needle inside sterile bowl
- Connect Heparin-filled syringe to the shorter Introducer needle and inject Heparin until needle is fully rinsed.
- Aspirate Heparin back into syringe and disconnect from needle.
- Repeat for the longer aspiration needle.
- Rinse each stylet, short introducer sharp and blunt, longer aspiration stylet.
- With needle guards in place, rinse the outside of each needle by injecting Heparin into the open end of the guard.

2,000 Units/mL Heparin Flush Bath
Heparin Flush Protocol

Preparation of a Heparin Flush Bath
- Obtain a 5mL vial of 5,000 units Heparin per mL (25,000 units in total).
- Using syringe, empty the 5mL into a sterile bowl.
- Add 7.5mL of sterile saline to bowl.
- Bowl contains 12.5mL of 2,000 units Heparin per mL.
- Summary: $25,000 \text{ (Units)} / 12.5 \text{ (mL)} = 2,000 \text{ Units/mL}$

Alternate Preparation
- Obtain 10mL of 1,000 unit per mL Heparin (10,000 units in total).
- No dilution required.

It is important that the strength per mL of the heparin rinse is at least 1,000 but preferably 2,000 and that you have adequate volume to rinse all of the needles and syringes.
Bone Marrow Aspiration Process Steps

Marrow Cellution™

1. Heparin Flush: Rinse all components with heparin (2,000 Units/ml)
2. Insert Access Needle just past cortex into medullary space. Ensure longitudinal orientation.
3. Remove Sharp Stylet, attach Syringe, draw 1ml marrow to test proper localization of Access Needle tip
4. Remove Syringe, insert Blunt Stylet. Continue to advance Access Needle to desired depth
5. Rotate Guide Grip to skin level and remove Blunt Stylet
6. Remove Blunt Stylet, insert and attach Aspiration Cannula.
7. Attach Syringe, draw 1ml marrow
8. Rotate handle 180°-360° counter clockwise to raise Cannula tip
9. Draw 1ml marrow
10. Repeat steps 8 & 9 until desired volume is attained

Abbreviated Instructions Overview. Information for Healthcare Professionals only. Refer to package insert for complete Instructions for Use.

For further information please contact your local Marrow Cellution representative or consult www.aspire-medical.eu

Manufacturer: Ranfac Corp.
30 Doherty Avenue
Avon/MA 02322, USA
www.ranfac.com

EU-Representative: Aspire Medical Innovation GmbH
Einsteinstraße 167
D-81677 Munich
www.aspire-medical.eu
**Hospital for Special Surgery (HSS): Dr. Joseph Lane, Comparison**

**AAOS 2013 Poster: Journal of Orthopedic Trauma (submission)**

**Aspiration to Application without Centrifugation**

<table>
<thead>
<tr>
<th>Count</th>
<th>Harvest</th>
<th>Biomet</th>
<th>Harvest</th>
<th>Arteriocyte</th>
<th>Marrow Cellution</th>
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</thead>
<tbody>
<tr>
<td><strong>Series 1</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Aspirate: 60mls, Concentrate: 7mls</td>
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<td></td>
<td></td>
<td></td>
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<tr>
<td><strong>Series 2</strong></td>
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<td></td>
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<tr>
<td>Aspirate: 60mls, Concentrate: 7mls</td>
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<tr>
<td><strong>Snap Aspiration</strong></td>
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<td></td>
</tr>
<tr>
<td>Aspirate: 5mls</td>
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</tbody>
</table>

| Nucleated Cell Count (million/ml) | 101,48 | 90,81 | 90,80 | 38,17 | 80,0 |
| Absolute CFU-F Count | 7.100 | 806 | 8.888 | 3.600 | 38.800 |
| CFU-F/ml | 1.014 | 134 | 1.270 | 514 | 7.760 |

1ml of Marrow Cellution (CFU-F/ml=7,760) contains the same CFU-F Volume compared with the final output after “point of care” processing of aspirated marrow (60mls) reduced to 7mls of concentrate.
What is the importance of CFU-f counts compared to nucleated cell counts?

CFU-f

- There is no constant ratio between average marrow cellularity as measured by number of total nucleated cells per mL and the number of CFU-f. Hernigou et al in several authoritative studies linked clinical outcomes in non-union and osteonecrosis to the number of CFU-f cells in the graft.

- Controlling for volume, Hernigou et al. noted that 70% of the variation in CFU-f from patient to patient was due to variations in the quality of the marrow aspirate or idiosyncratic to the patient with the remaining variation being due to the number of nucleated cells per mL in the aspirate.

- Statistically, the only variable Hernigou reported to be significant was CFU-f and not nucleated cells per mL. Interestingly, CFU-f is found frequently in marrow and very rarely in peripheral blood.

- “Therefore, it seems reasonable to suggest that a graft needs to contain greater than 1000 progenitors/cm$^3$” (P. Hernigou).

References:
## Comparative Results

<table>
<thead>
<tr>
<th>System</th>
<th>Hernigou Cobe</th>
<th>Harvest SmartPReP™</th>
<th>Marrow Cellution™</th>
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<tbody>
<tr>
<td><strong>Aspiration Volume</strong></td>
<td>306mL (Mean)</td>
<td>120mL (Mean)</td>
<td>10mL or <strong>20mL</strong></td>
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<tr>
<td><strong>CFU-f in Aspirate</strong></td>
<td>612/mL (Mean)</td>
<td>485/mL (Mean)</td>
<td>2275/mL</td>
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<tr>
<td><strong>Concentrate Volume</strong></td>
<td>20mL (Mean)</td>
<td>15mL (Mean)</td>
<td>10mL or 20mL (Unchanged)</td>
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<tr>
<td><strong>Total CFU-f Delivered in Concentrate</strong></td>
<td>2,579/mL (Mean)</td>
<td>3,200/mL (Mean)</td>
<td>2,275/mL (Unchanged)</td>
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<tr>
<td></td>
<td>Range: 1,458 - 3,700 /ml</td>
<td>Range: 2,500 - 4,100 /ml</td>
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<tr>
<td><strong>Yield of CFU-f in Concentrate</strong></td>
<td>27.5%</td>
<td>82%</td>
<td>100%</td>
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<td><strong>CFU-f Delivered to Non-Union Site</strong></td>
<td>51,589 (Mean)</td>
<td>48,000 (Mean)</td>
<td>22,750 in 10mL</td>
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<tr>
<td></td>
<td>Range: 34,149 - 74,000</td>
<td>Range: 37,500 - 61,500</td>
<td><strong>45,500 in 20mL</strong></td>
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</table>

Source: Hernigou / Harvest Comparison with Marrow Cellution Data
Competitive Performance

CFU-f Cell Count Comparison

<table>
<thead>
<tr>
<th></th>
<th>CFU-f / ml</th>
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<tr>
<td>Marrow Cellution</td>
<td>3,290 (1)</td>
</tr>
<tr>
<td>Harvest BMAC</td>
<td>1,270 (2)</td>
</tr>
<tr>
<td>Arteriocyte Magellan</td>
<td>514 (3)</td>
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<tr>
<td>Biomet BioCUE</td>
<td>134 (4)</td>
</tr>
<tr>
<td>Traditional Trocar</td>
<td>356 (4)</td>
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References:
## CFU Counts

### Additional Field Samples

<table>
<thead>
<tr>
<th>Lab</th>
<th>Orientation</th>
<th>Volume (mL)</th>
<th>TNC (x$10^6$)</th>
<th>CFU-f / mL</th>
<th>Total CFU-f in Graft</th>
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</thead>
<tbody>
<tr>
<td>CBR</td>
<td>Anterior</td>
<td>8</td>
<td>29.72</td>
<td>1,039</td>
<td>8,316</td>
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<td>36.44</td>
<td>4,513</td>
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<td>UT</td>
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<td>42.00</td>
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<td>21.60</td>
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<td>24.00</td>
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<td>Posterior</td>
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<td>Franciscan</td>
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<td>34,000</td>
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<td>Franciscan</td>
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<td><strong>Average</strong></td>
<td><strong>2,070</strong></td>
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Source: Validation Samples via Independent Laboratories
# Quick Fact Sheet

## Marrow Cellution™ vs. Centrifugation Systems

<table>
<thead>
<tr>
<th>Feature</th>
<th>Marrow Cellution™</th>
<th>Centrifugation Systems</th>
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<tbody>
<tr>
<td>Time</td>
<td>5 minutes</td>
<td>45 minutes</td>
</tr>
<tr>
<td>Invasiveness</td>
<td>10mL</td>
<td>60mL – 240mL</td>
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<tr>
<td>Efficiency</td>
<td>100% Utilizable</td>
<td>85% Discarded</td>
</tr>
<tr>
<td></td>
<td></td>
<td>15% Utilizable</td>
</tr>
<tr>
<td>Contamination Risk</td>
<td>100% Sterile Field</td>
<td>Offsite Processing</td>
</tr>
<tr>
<td>Peripheral Blood Contamination</td>
<td>Minimal</td>
<td>High</td>
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<tr>
<td>Personnel Time and Training</td>
<td>None</td>
<td>Extensive</td>
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<tr>
<td>Technique Sensitivity</td>
<td>No</td>
<td>Extensive</td>
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<tr>
<td>Regulatory Compliance</td>
<td>Compliant</td>
<td>Advanced Therapy Drug</td>
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</table>
Regulatory Compliance Becoming Increasingly More Restrictive

Advanced Therapy Medicinal Product (ATMP)

Regulatory Review & ATMP Assessment

of 13 November 2007
on advanced therapy medicinal products and amending Directive 2001/83/EC
and Regulation (EC) No 726/2004
(Text with EEA relevance)
Definition of ATMP

Advanced Therapy Medicinal Products
- Gene Therapy Medicinal Products (GTMP)
- Somatic Cell Therapy Medicinal Products (SCTMP)
- Tissue Engineering Products (TEP)
- Combined ATMP’s and Medical Devices

Gene Therapy Medicinal Products and Somatic Cell Therapy Products defined in Annex 1 to Directive 2001/83/EC
- Tissue Engineering Products: “Tissue engineering is the regeneration of biological tissue through the use of cells, with the aid of supporting structures and/or biomolecules” Defined in Reg: EU No. 1394/2007
EU Regulation 1394 / 2007
Do I have an ATMP according to EU 1394/2007 Regulation?

**ATMP Definition:**
Contains or consists...

...of cells or tissues that have been subject to **substantial** manipulation so that biological characteristics, physiological functions or structural properties relevant for the intended clinical use have been altered,

or...

...of cells or tissues that are not intended to be used for the **same essential function(s)** in the **recipient** and the **donor** (Homologous Use);
Thank you
Marrow Cellution System

- Maximizes Cell Yield
- Regulatory Compliant
- Centrifugation Not Required
- Never Leaves the Sterile Field
- Reduces Blood Contamination
- Reduces Donor Site Morbidity

Marrow Cellution™ provides substantial savings in time, effort and expense. It reduces patient trauma, morbidity and risk of infection.