“Cellular heterogeneity as predictor for the differentiation and regenerative potential of bone marrow stromal cells”

The Danish 3R Symposium 2021

Ali Jamil, PhD student

Molecular Endocrinology Unit (KMEB),

Odense University Hospital / University of Southern Denmark

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Background

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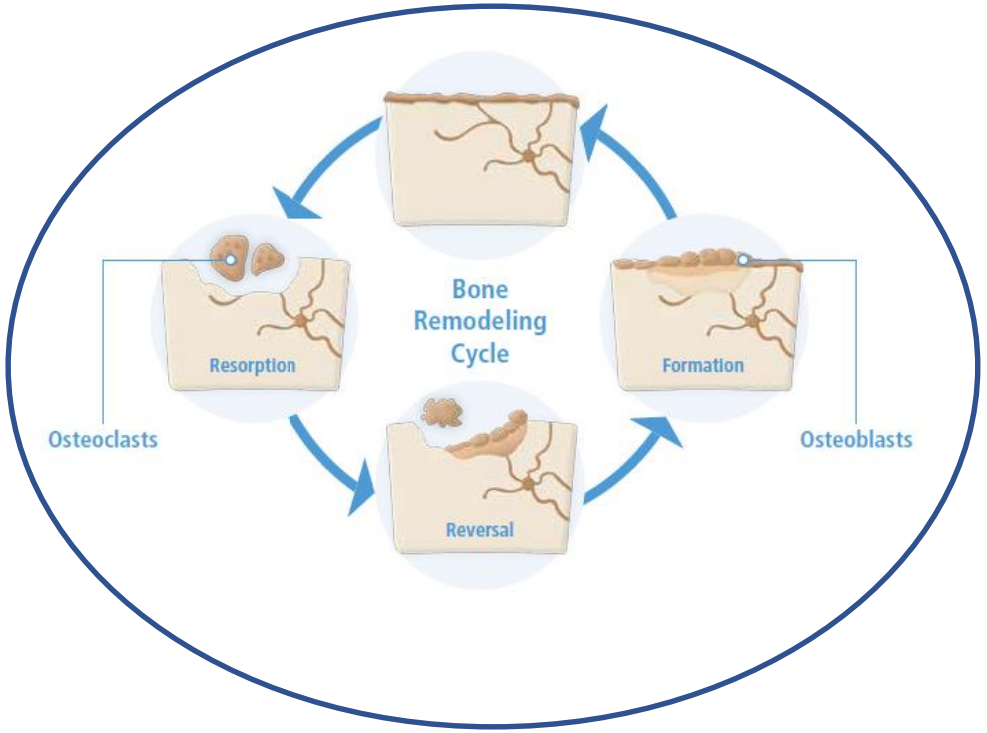
Background

Bone remodelling



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Background

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Background

Step 1: Remove

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Background

Step 1:
Remove

Step 2:
Pour new

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Background

Step 1: Remove

+

Step 2: Pour new

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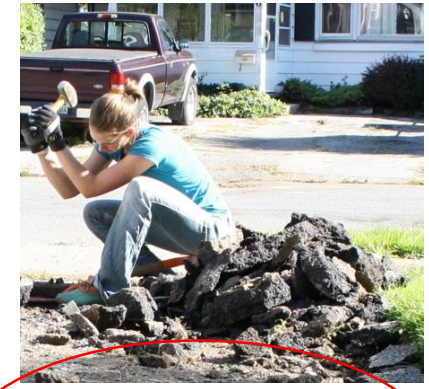
Background

Old bone

Step 1: Remove



Osteo**clast** cells

+

Step 2: Pour new

Osteo**blast** cells

New bone

Background

Old bone

Step 1: Remove

+

Osteo**clast** cells

Step 2: Pour new

Osteo**blast** cells

New bone

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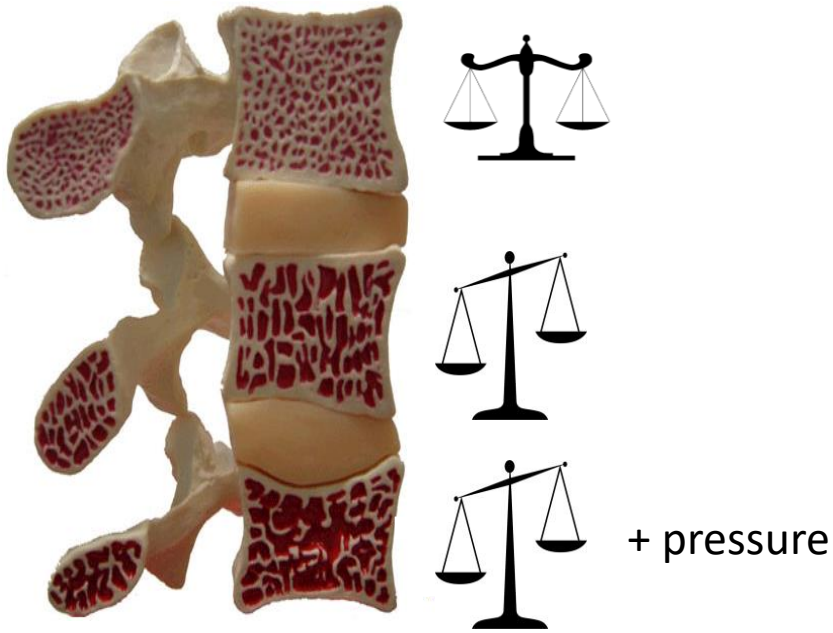
Background

Osteoporosis



+ pressure

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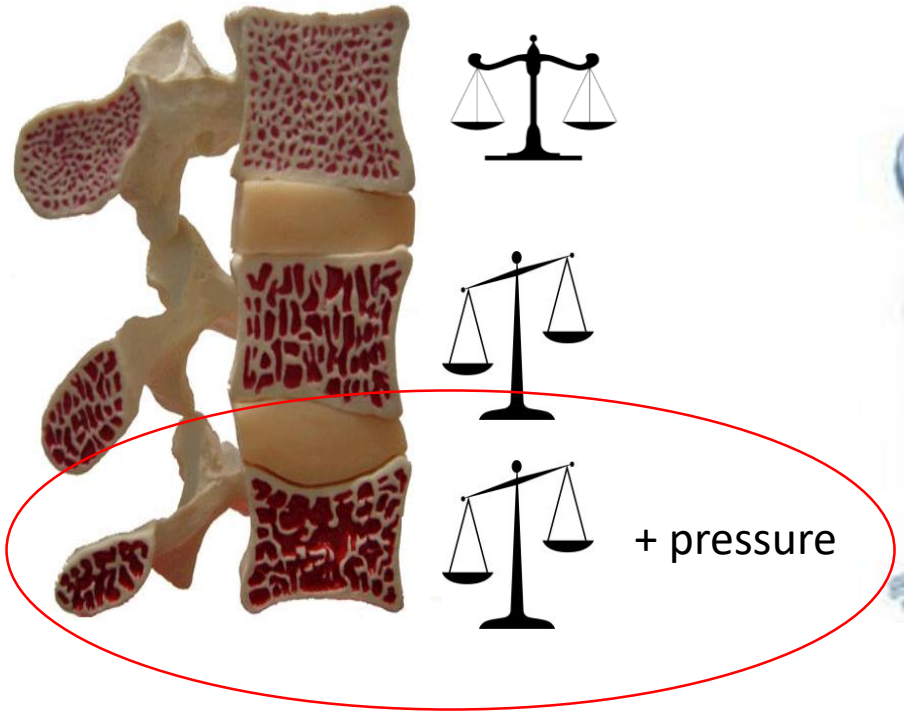
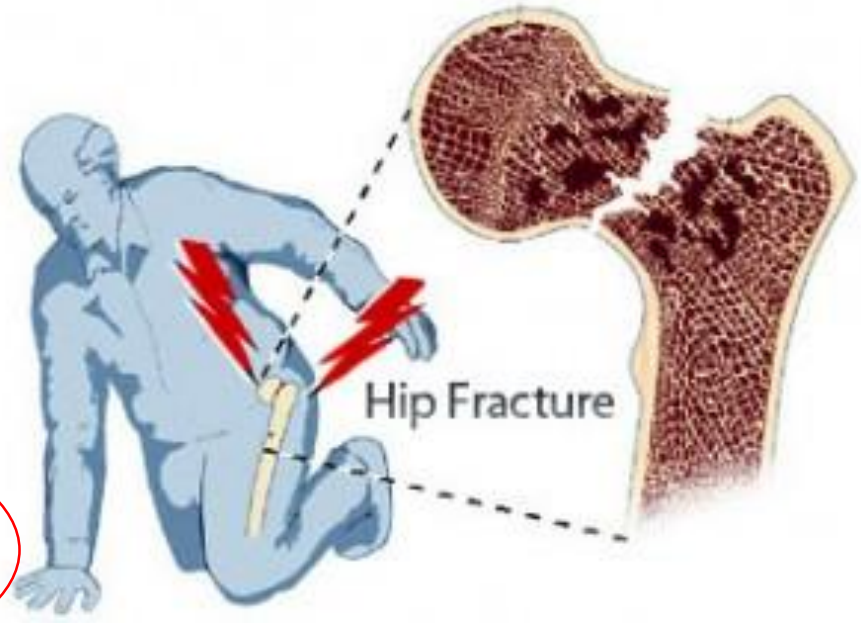
Background

Osteoporosis



+ pressure



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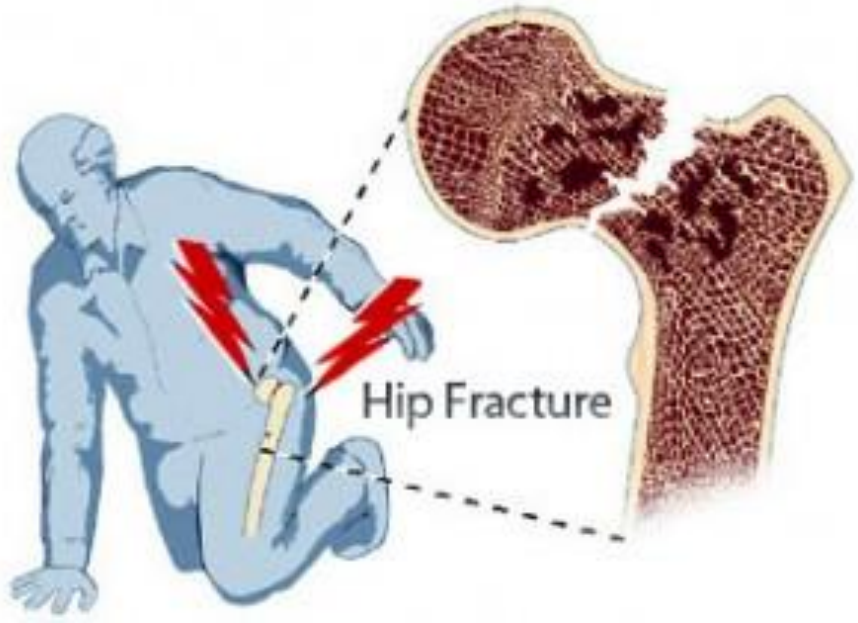
Background

Osteoporosis

+ pressure



Cell Transplantation

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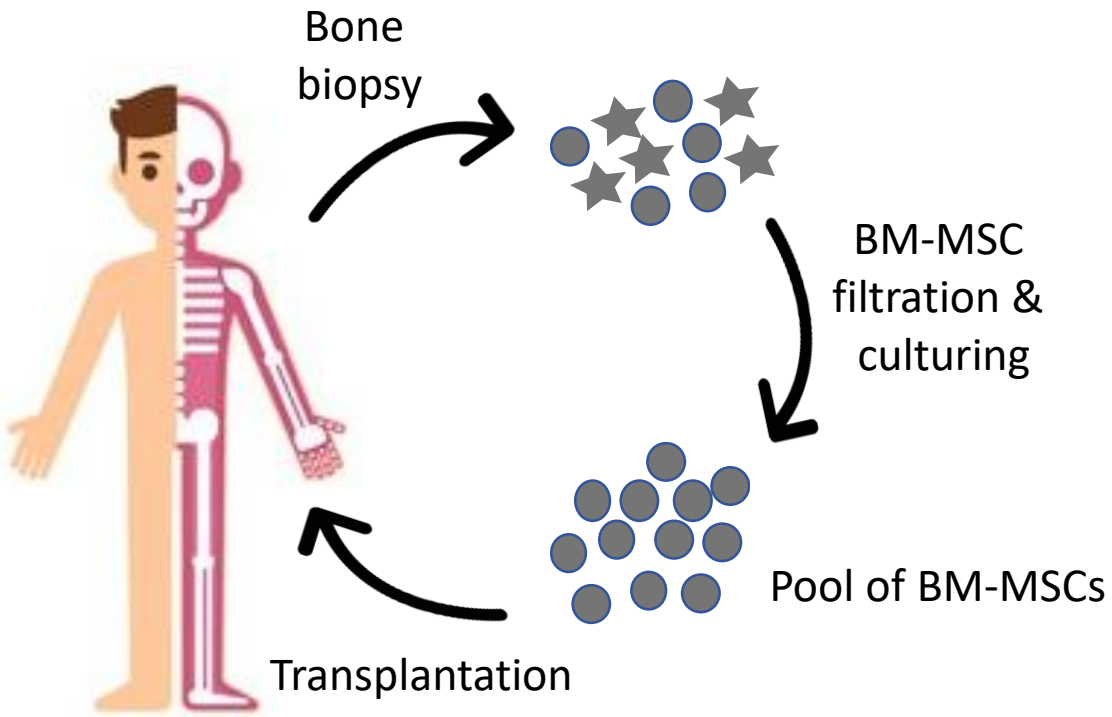
Background

Cell transplantation



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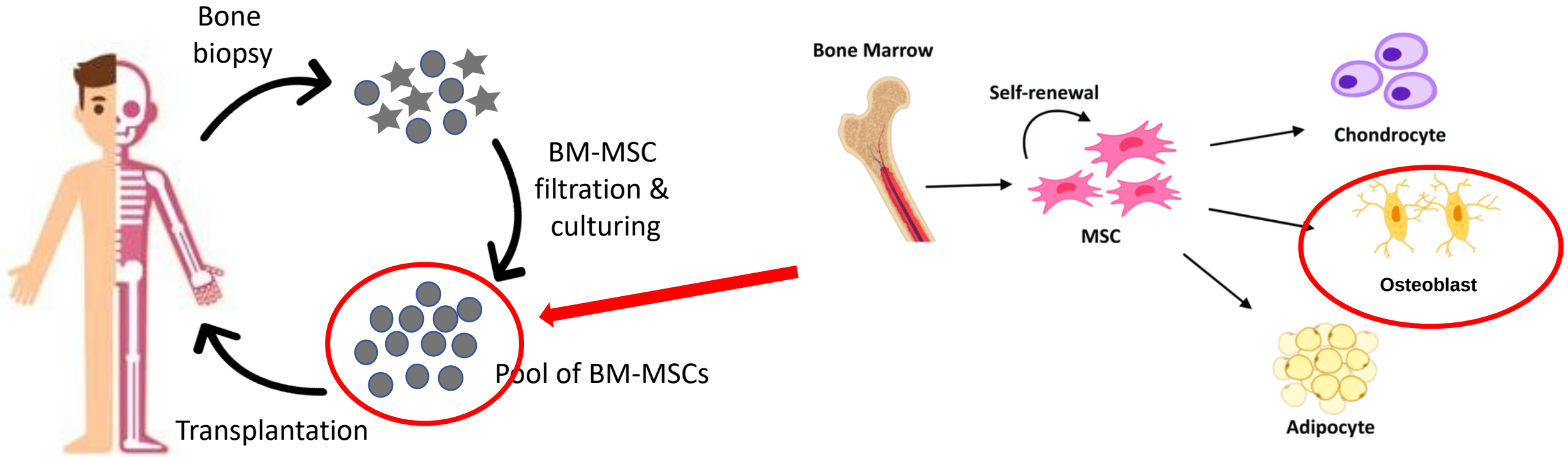
Background

Cell transplantation



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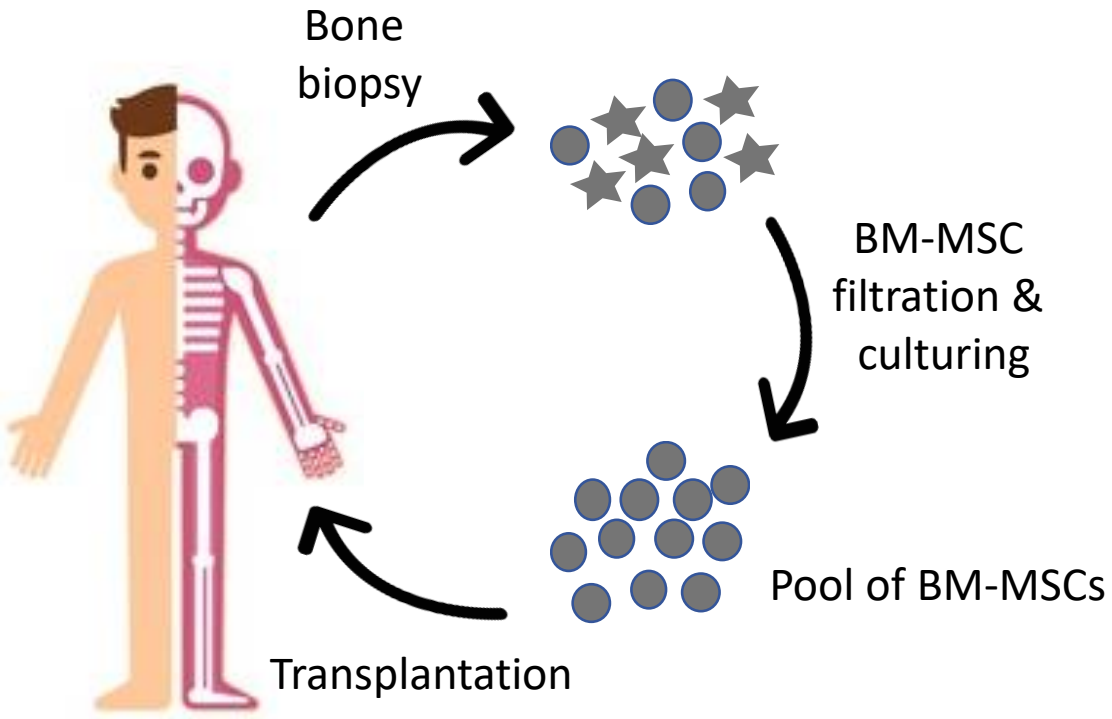
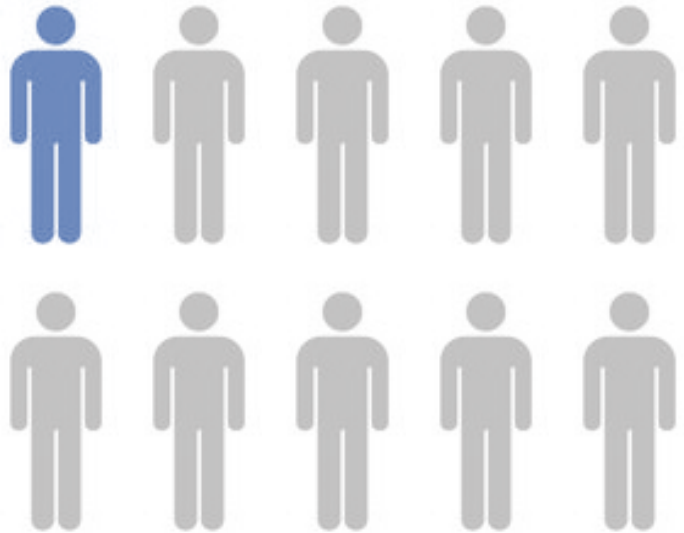
Background

Cell transplantation





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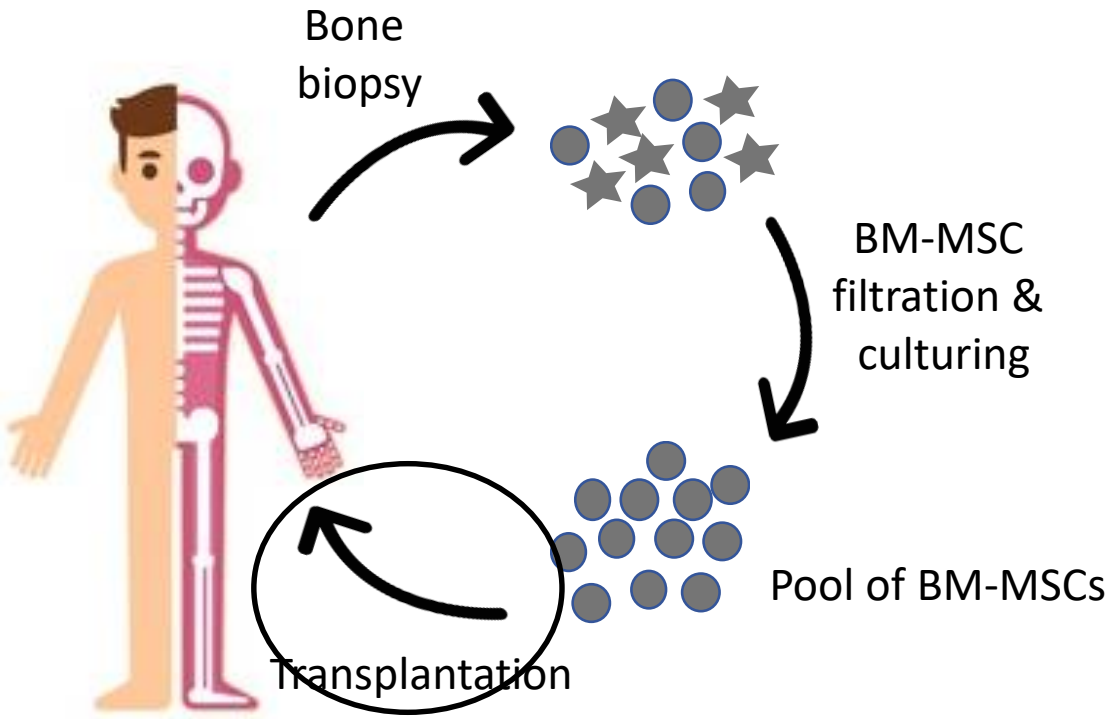
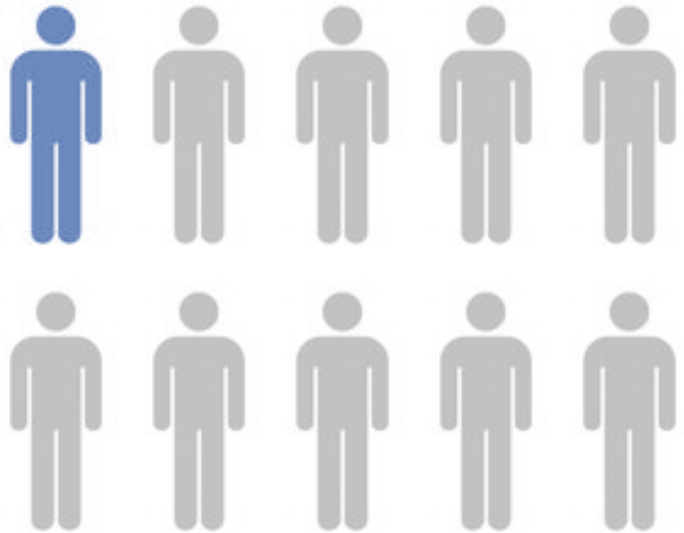
Background

Cell transplantation





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Background

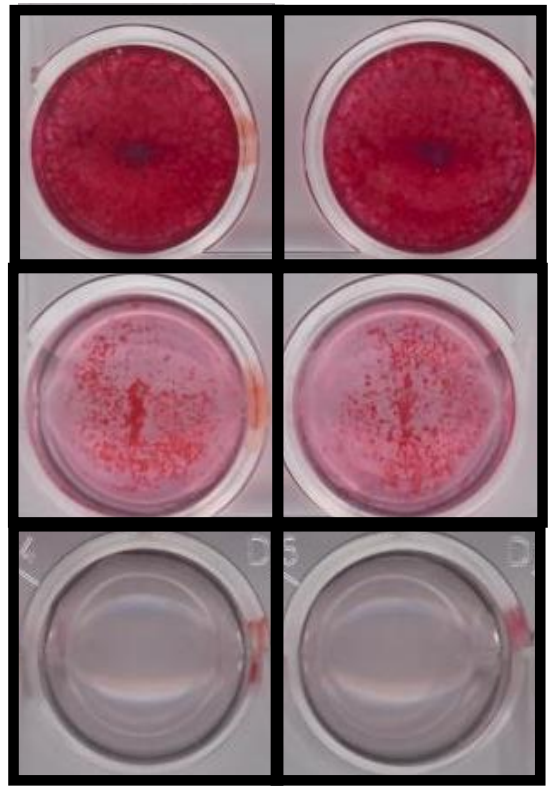
Cohort size = 56

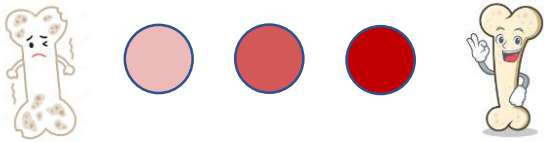
Donor examples:

Donor 1

Donor 2

Donor 3





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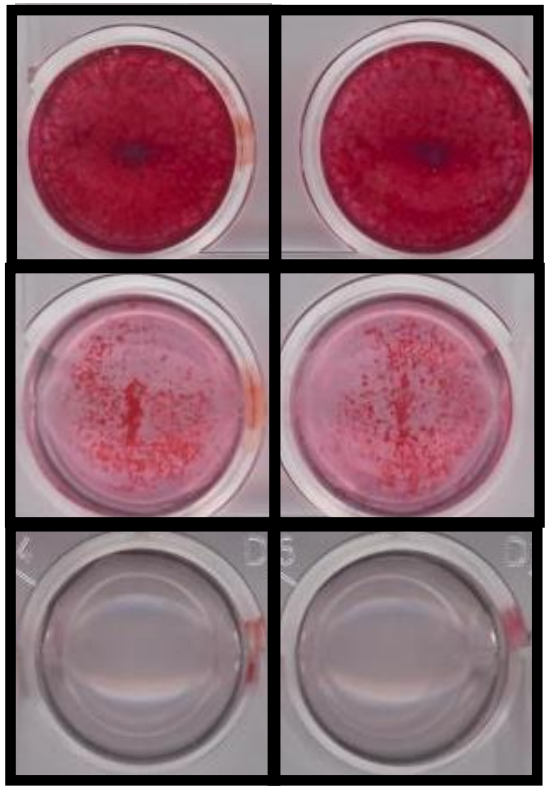
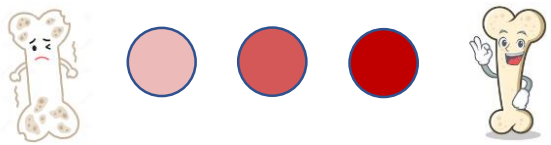
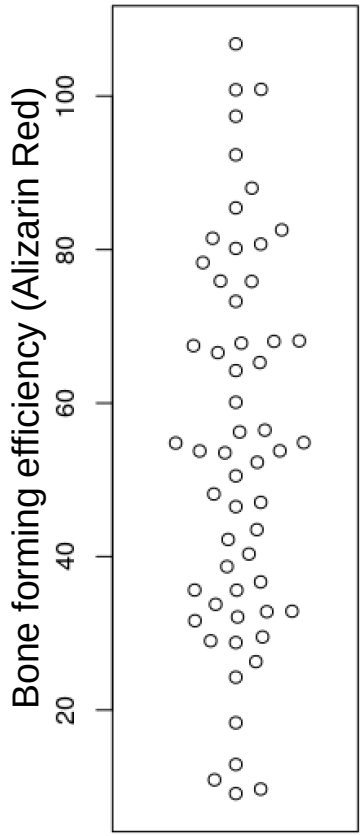




All donors:



Background

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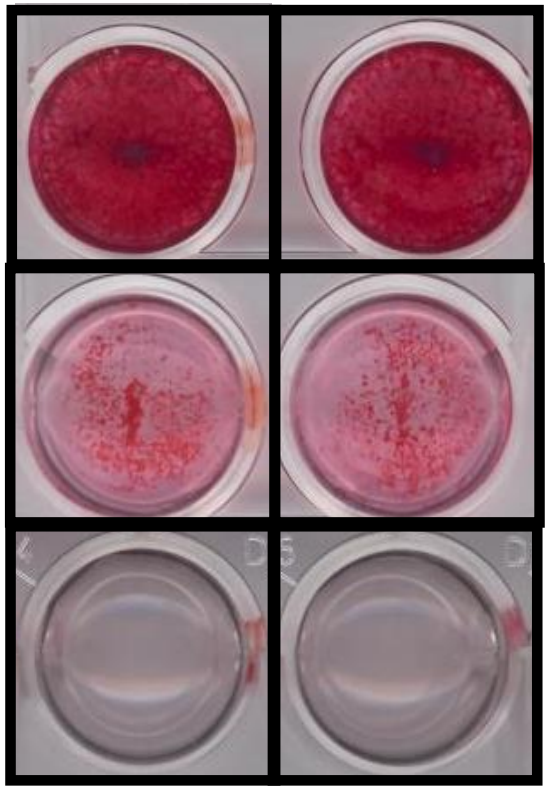
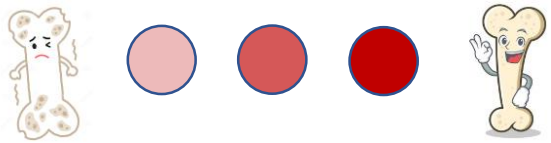




All donors:

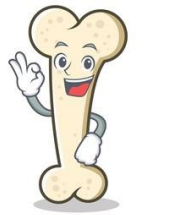
Gender

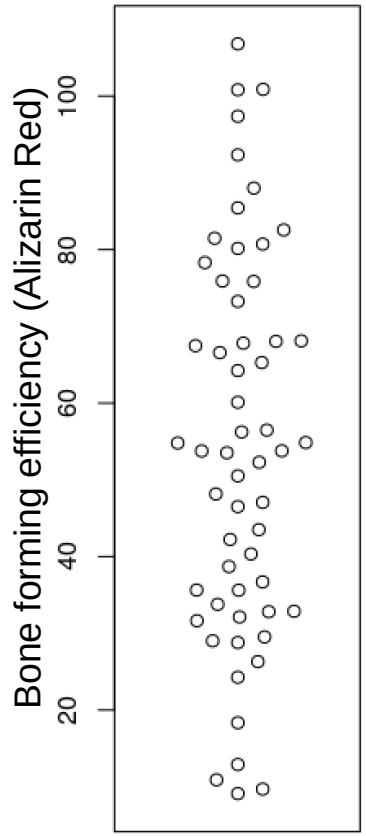
Age

BMI

Osteoporosis

D-vitamin





Background

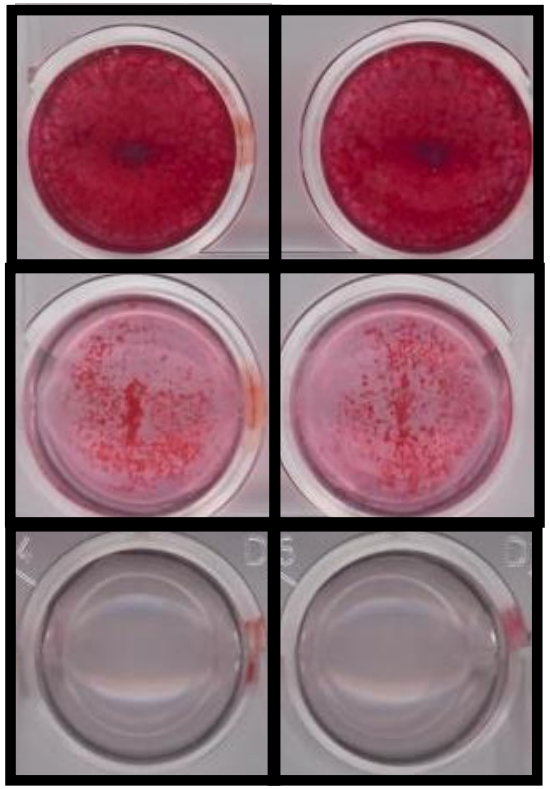
Cohort size = 56

Donor examples:

Donor 1

Donor 2

Donor 3



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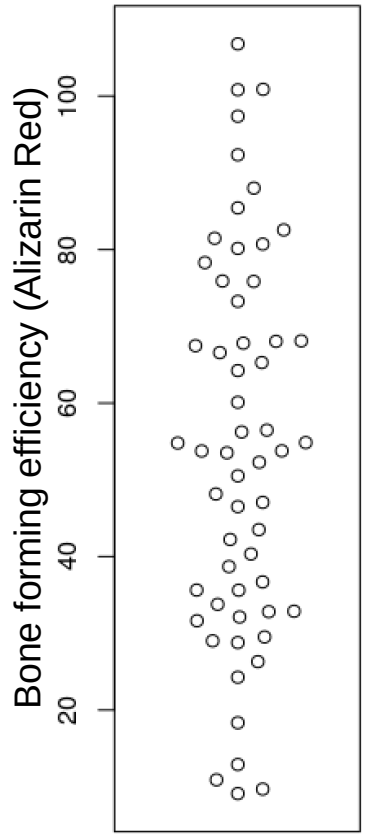

50% potency explained

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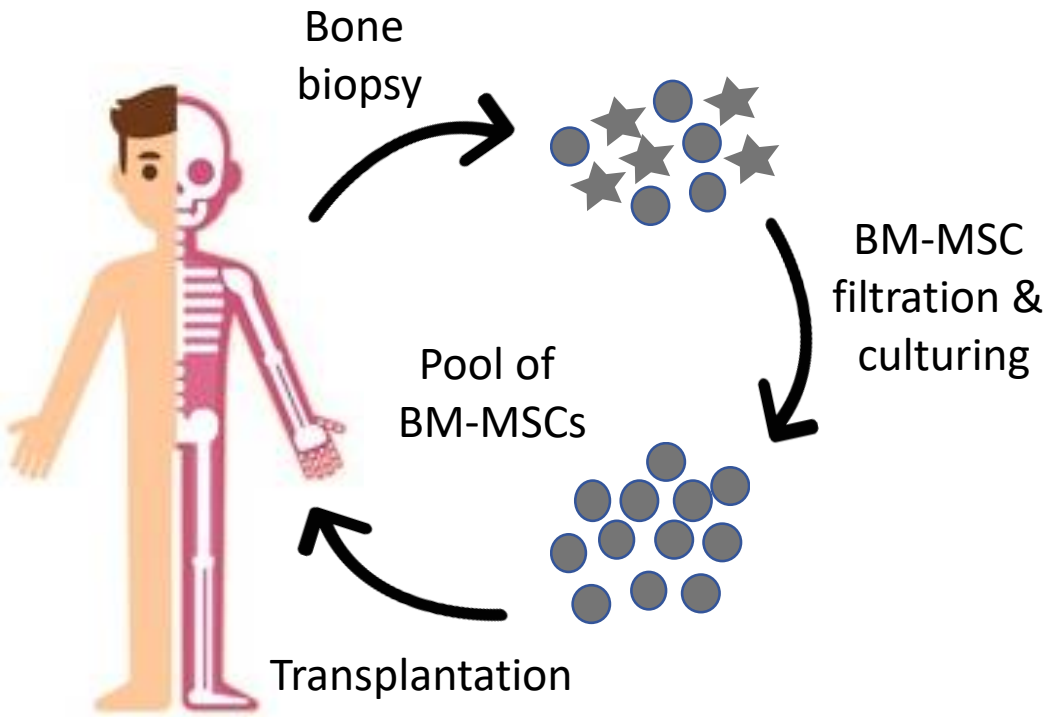
Hypothesis



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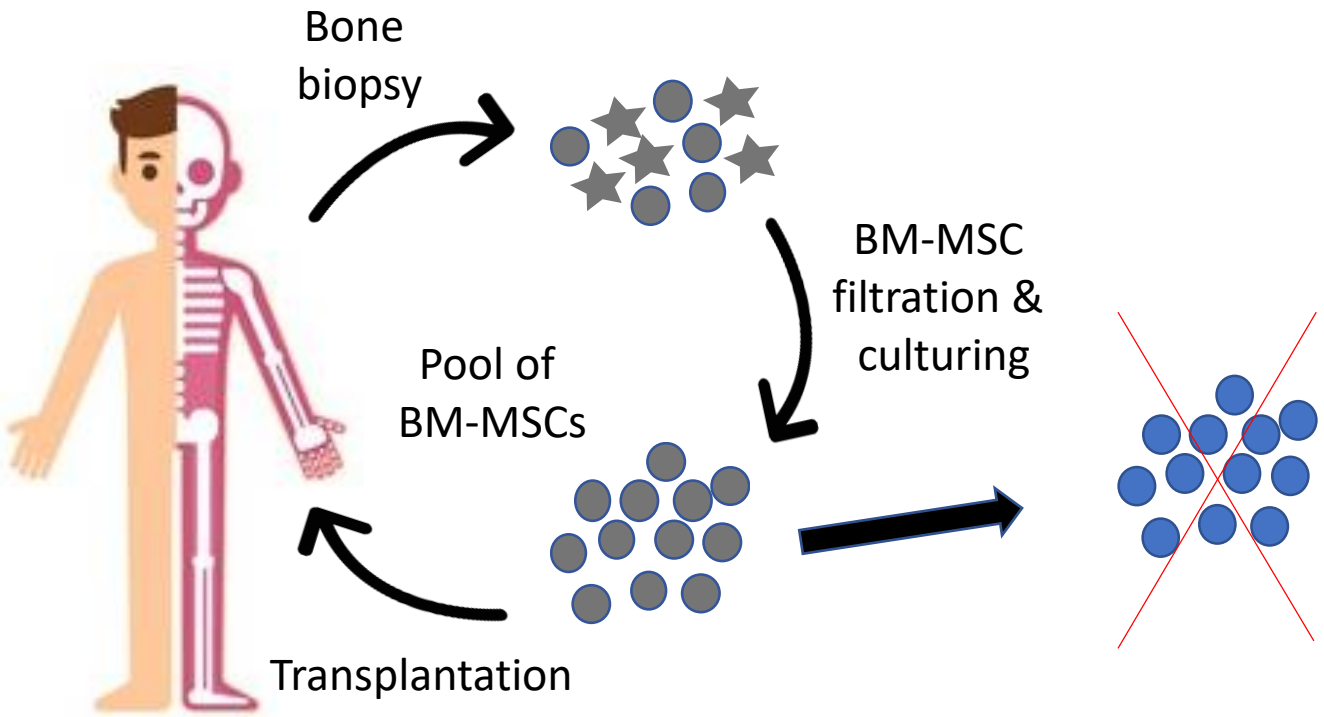
Hypothesis



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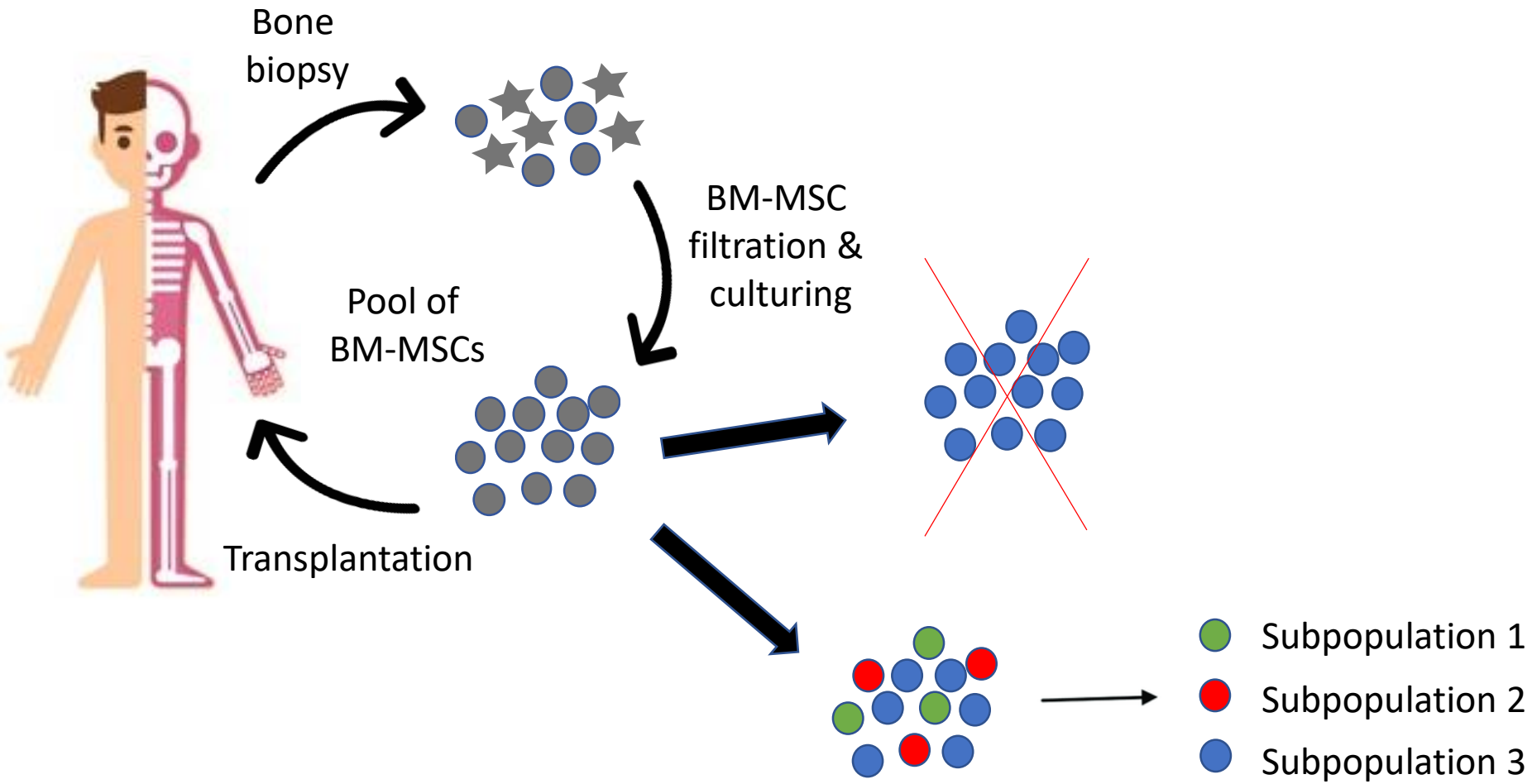
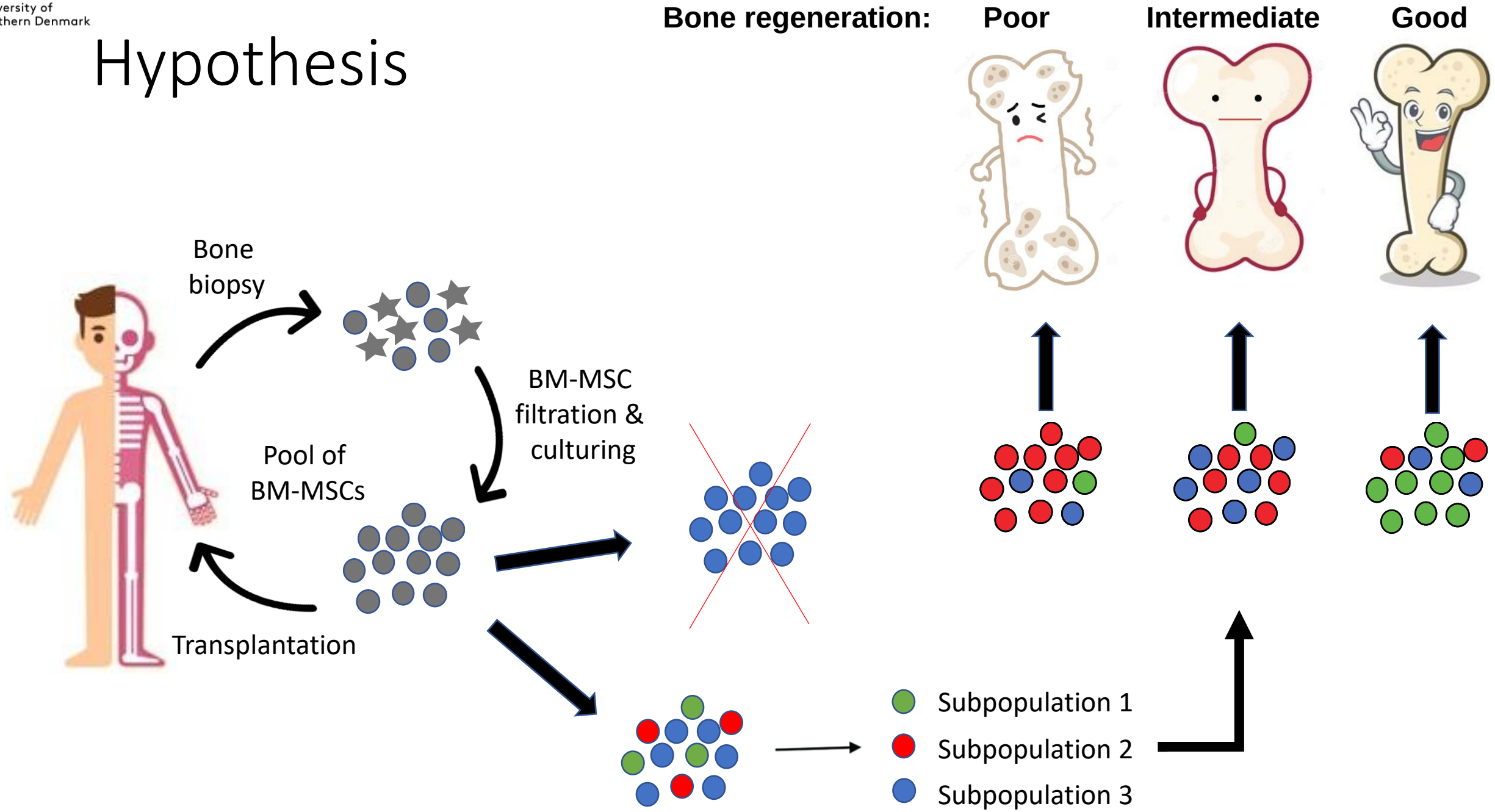
Hypothesis

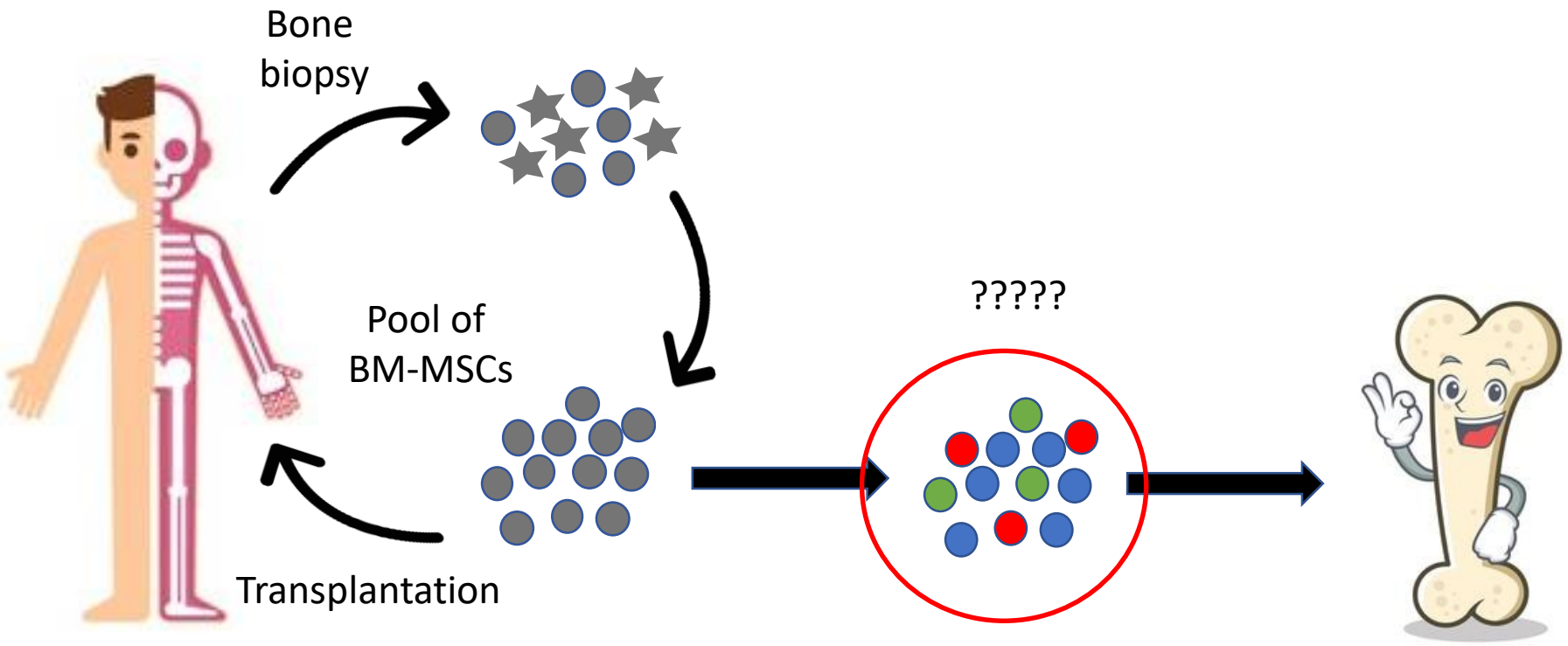


Hypothesis

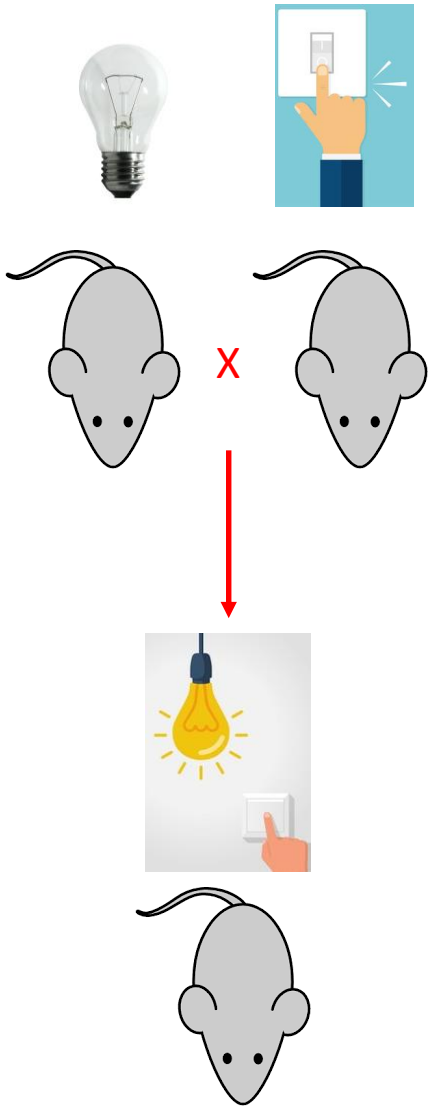

Bone regeneration:
Poor
Intermediate
Good
Bone biopsy
BM-MSC filtration & culturing
Pool of BM-MSCs
Transplantation
Subpopulation 1
Subpopulation 2
Subpopulation 3

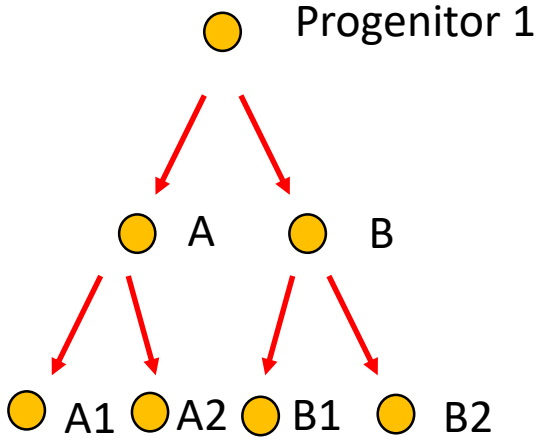
Overall aim

To investigate how cellular heterogeneity within patient-derived BM-MSCs affect osteogenic differentiation and bone tissue regeneration potential.

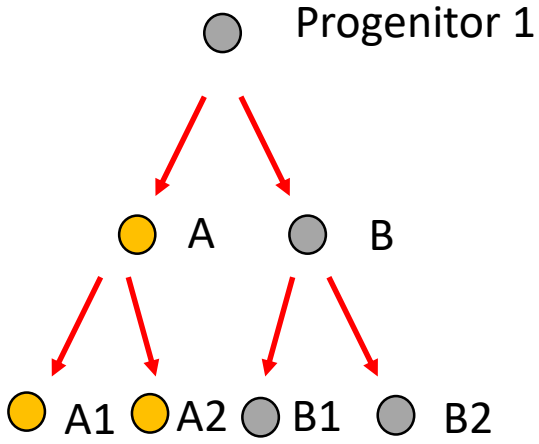


Lineage tracing

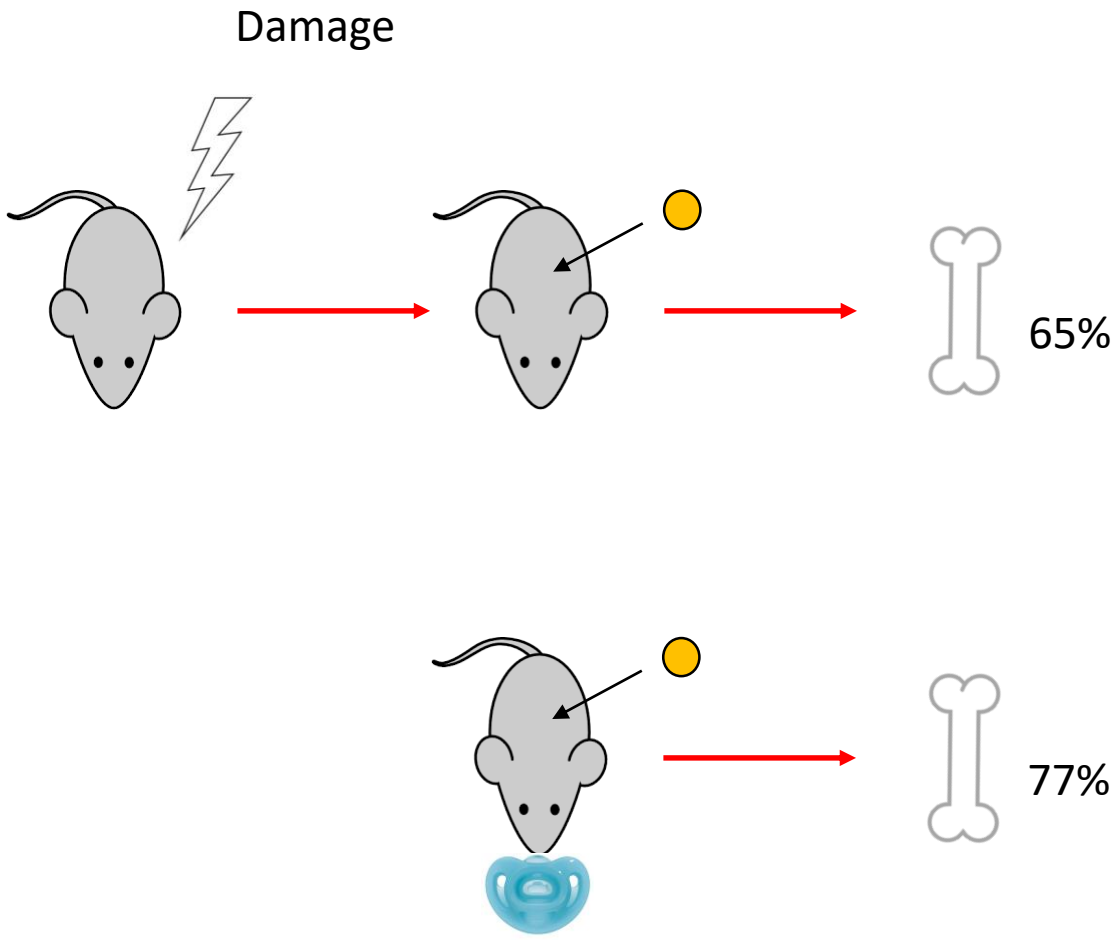




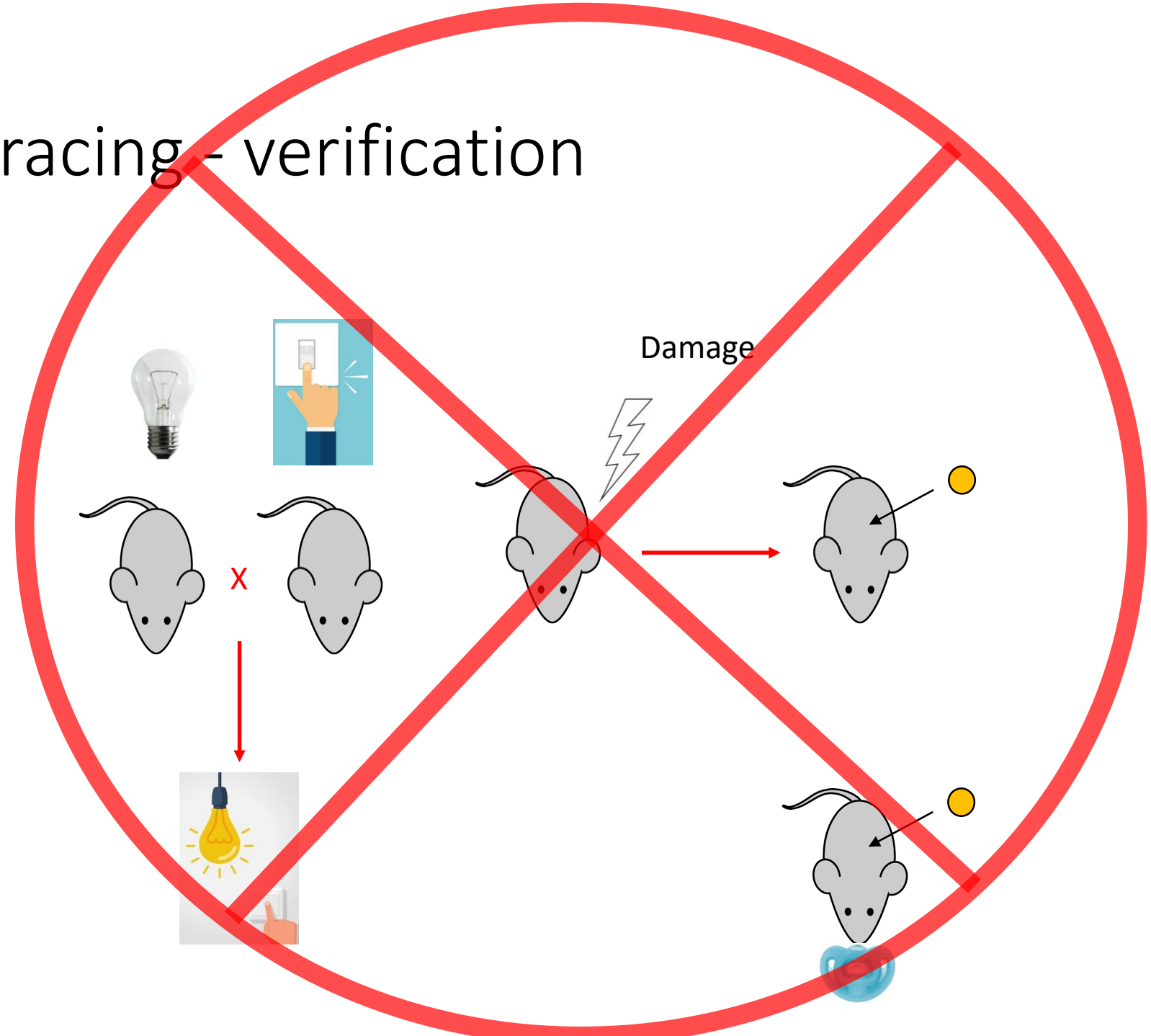
or



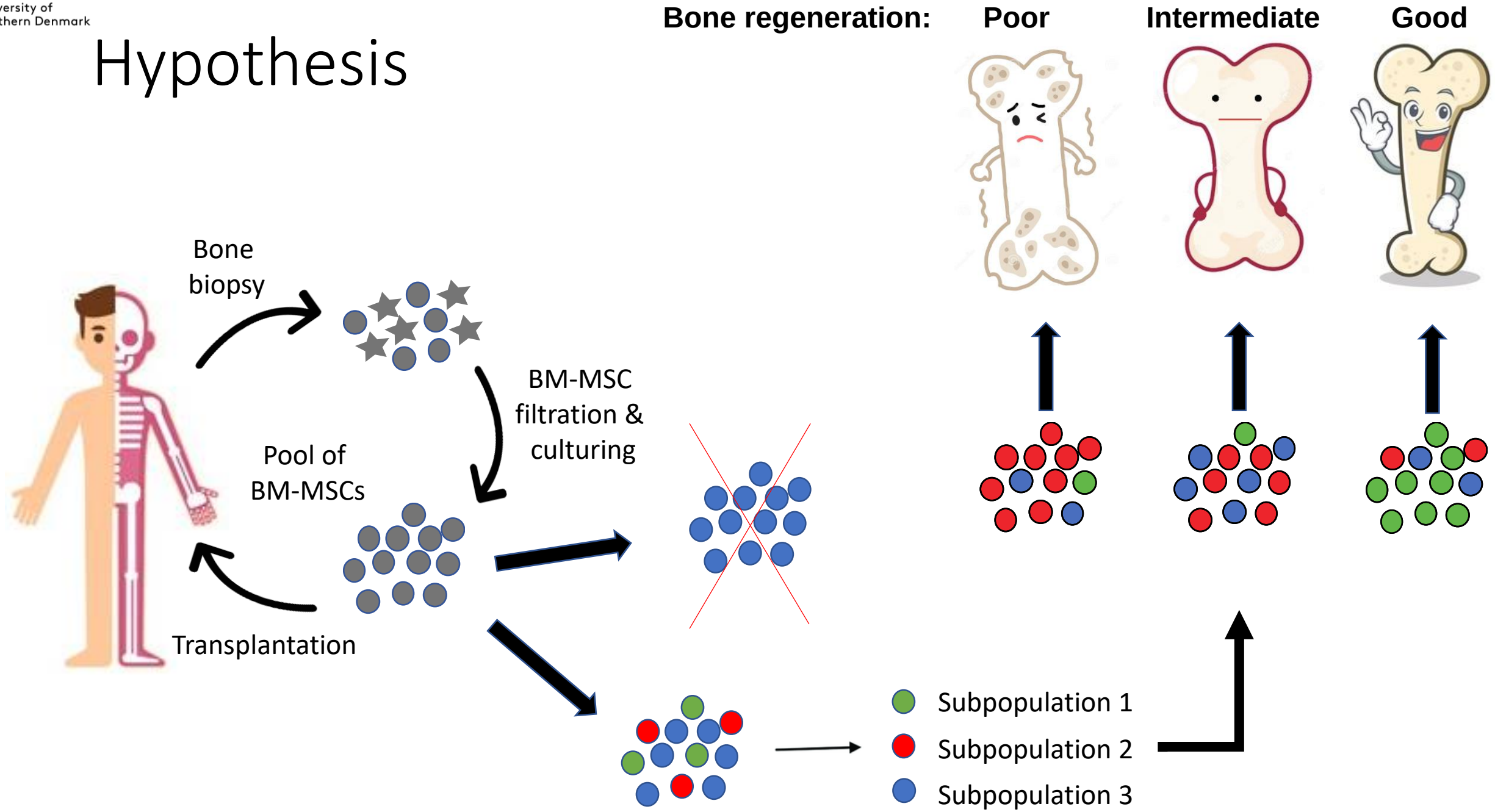
Lineage tracing - verification



Lineage tracing - verification



Hypothesis


Bone regeneration:
Poor
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BM-MSC filtration & culturing
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Subpopulation 1
Subpopulation 2
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scRNA-sequencing

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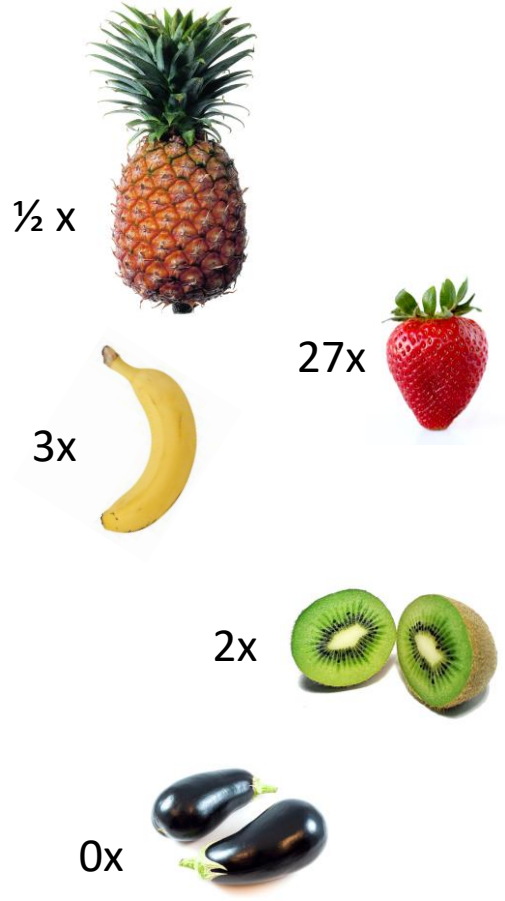
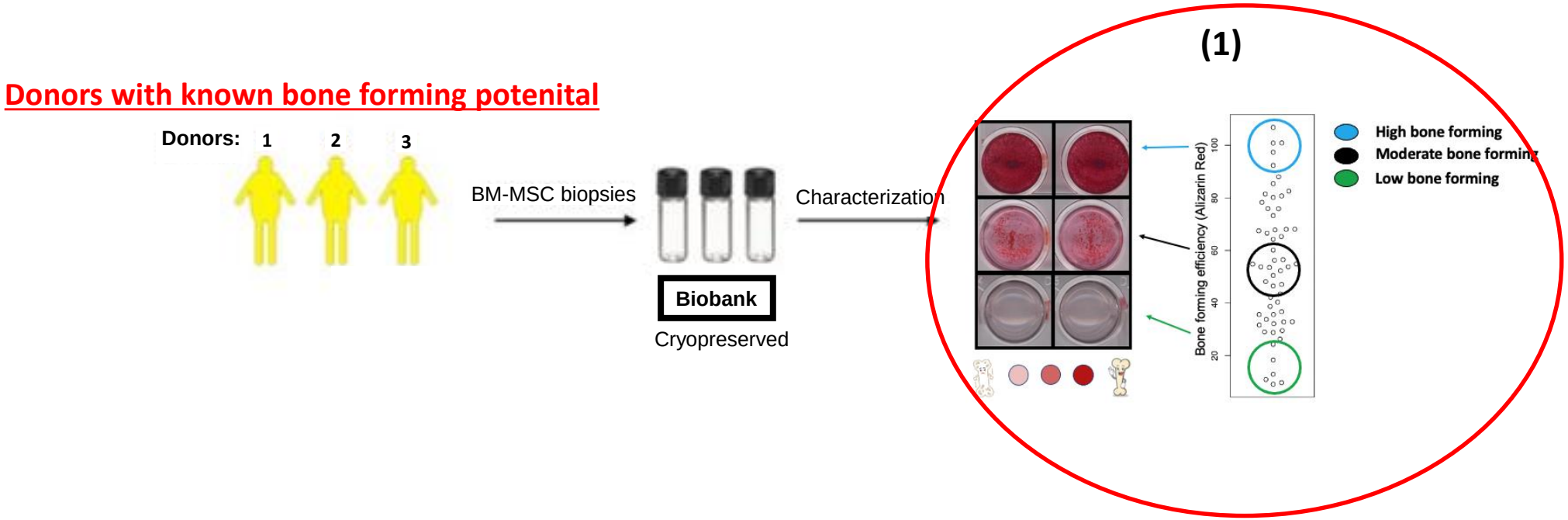
scRNA-sequencing



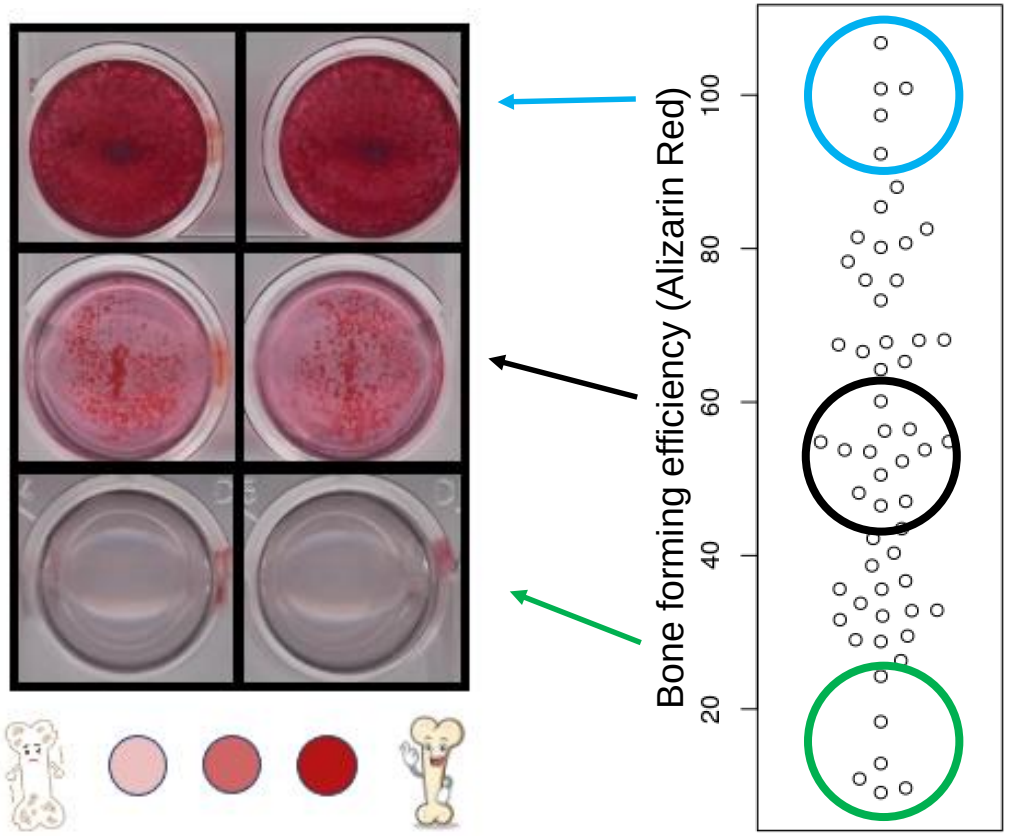
Study design - identification



Practicalities:

1) **Sample selection**

Study design - identification



High bone forming

Moderate bone forming

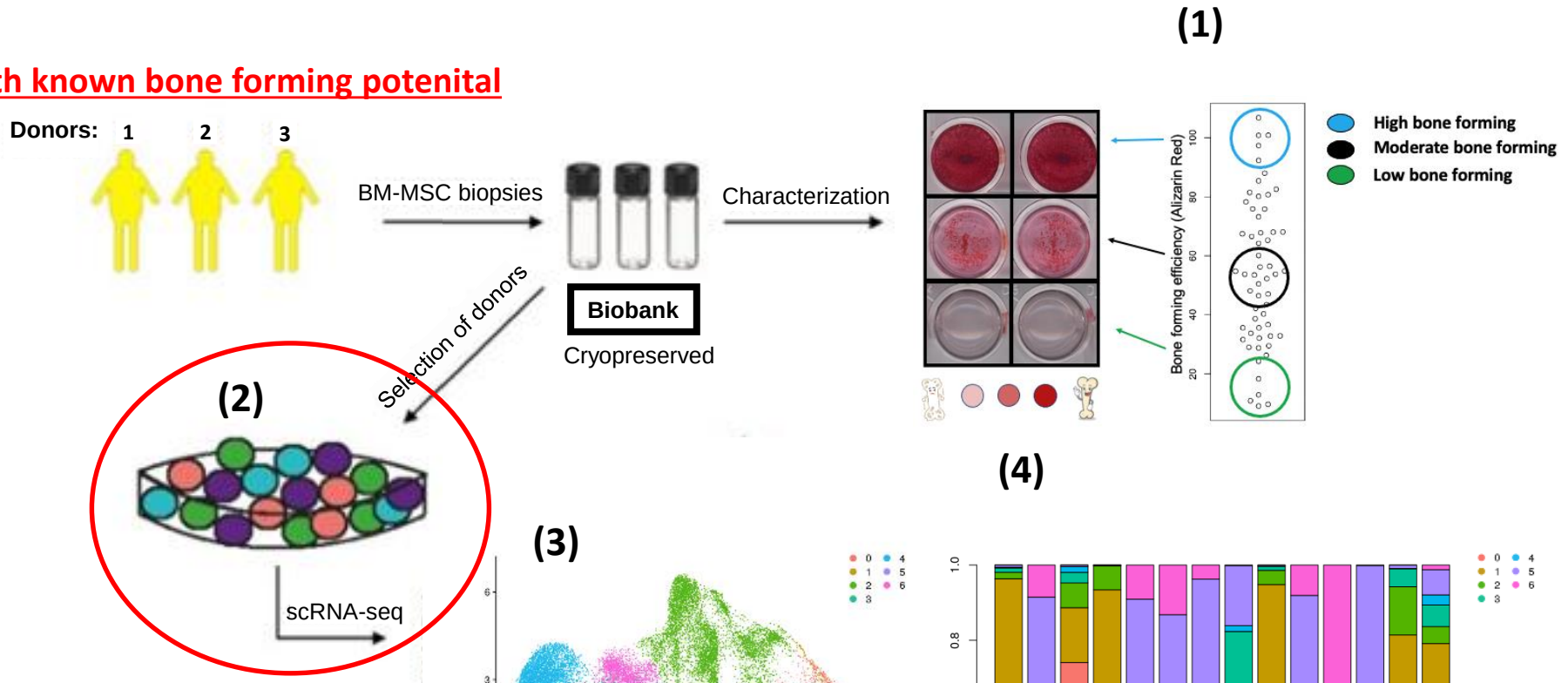
Low bone forming

Practicalities:

1) **Sample selection**

Study design - identification

Donors with known bone forming potenital

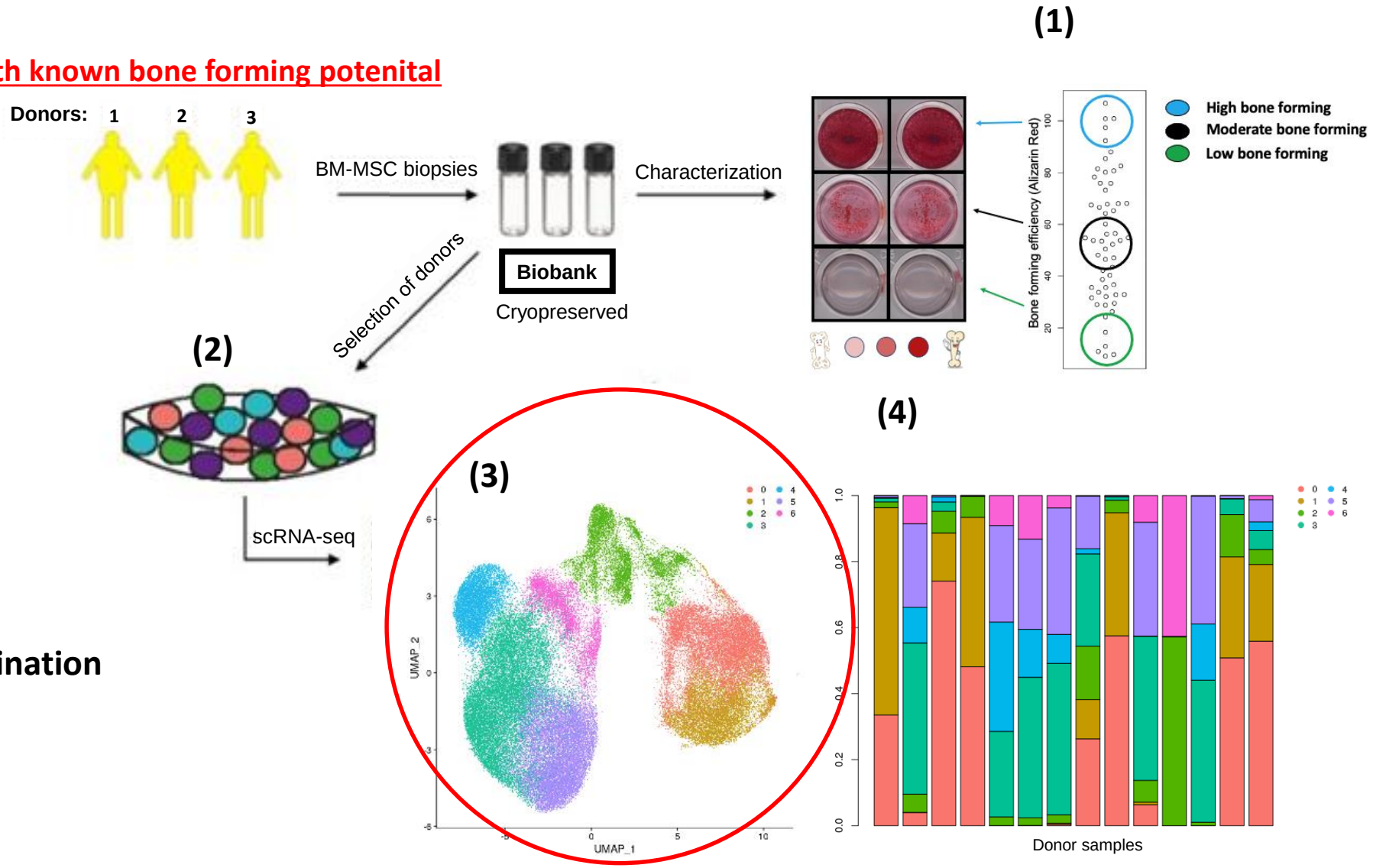


Practicalities:

1) **Sample selection**
2) **Single cell RNA-sequencing**

Study design - identification

Donors with known bone forming potenital

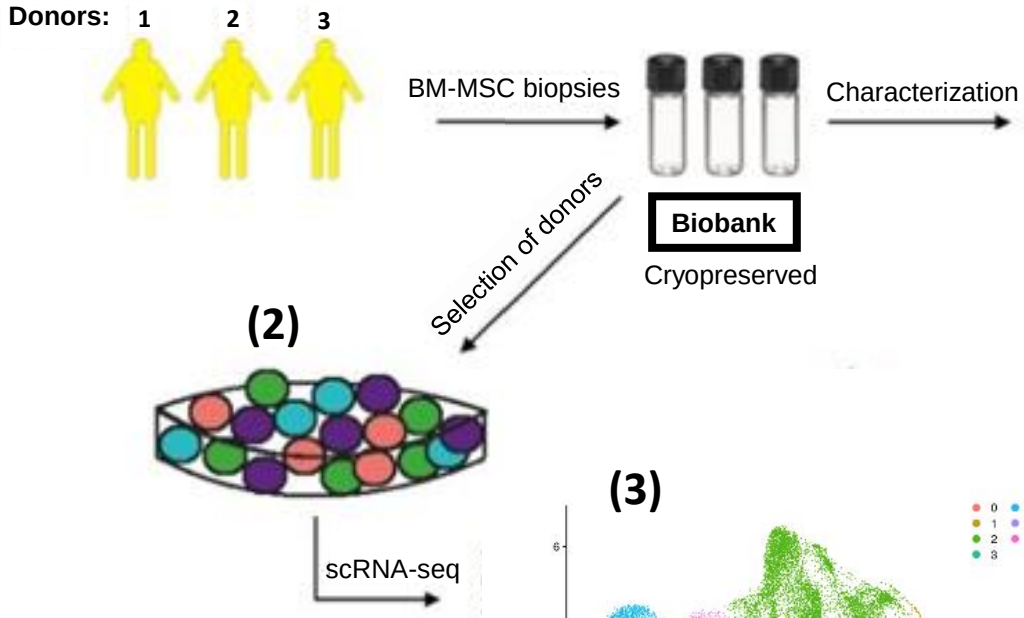


Practicalities:

1) **Sample selection**
2) **Single cell RNA-sequencing**
3) **BM-MSC subpopulation determination**

Study design - identification

Donors with known bone forming potenital



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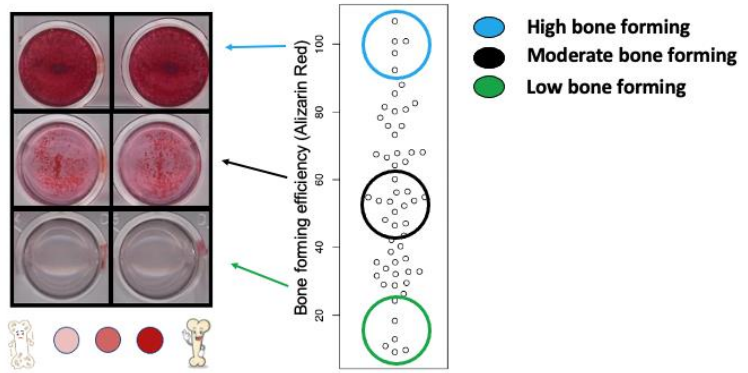

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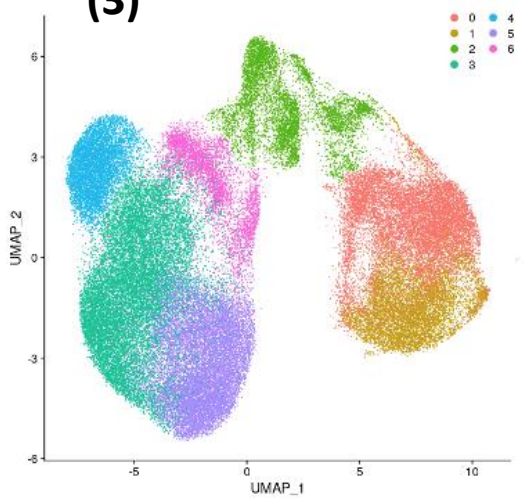

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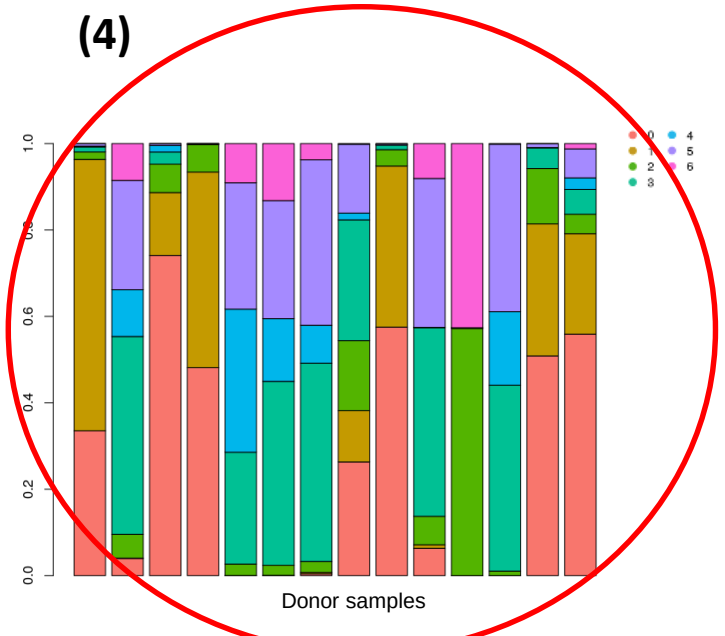

Practicalities:

1) **Sample selection**
2) **Single cell RNA-sequencing**
3) **BM-MSC subpopulation determination**
4) **BM-MSC subpopulation abundancies quantification**

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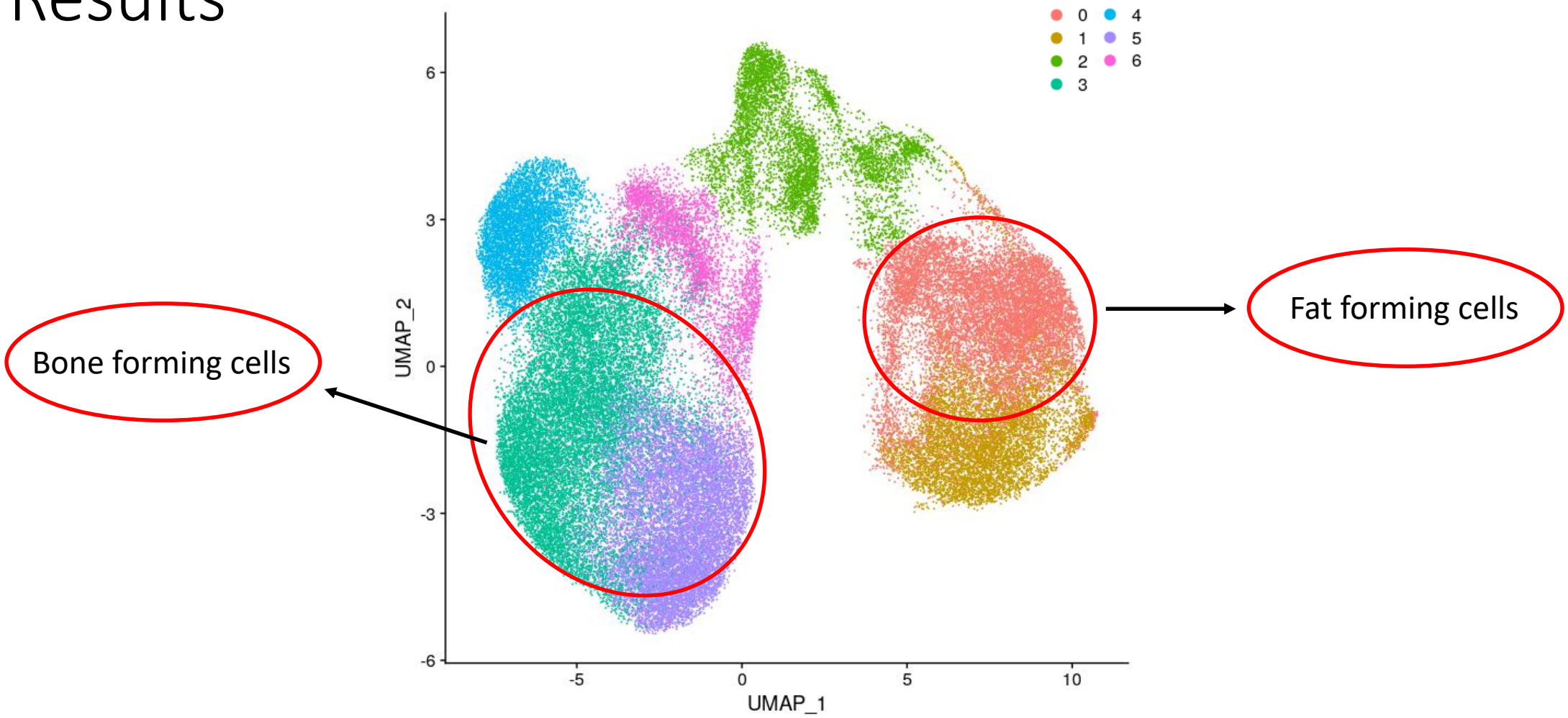
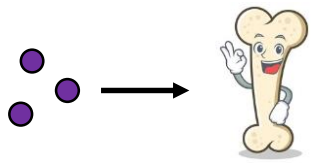
Results



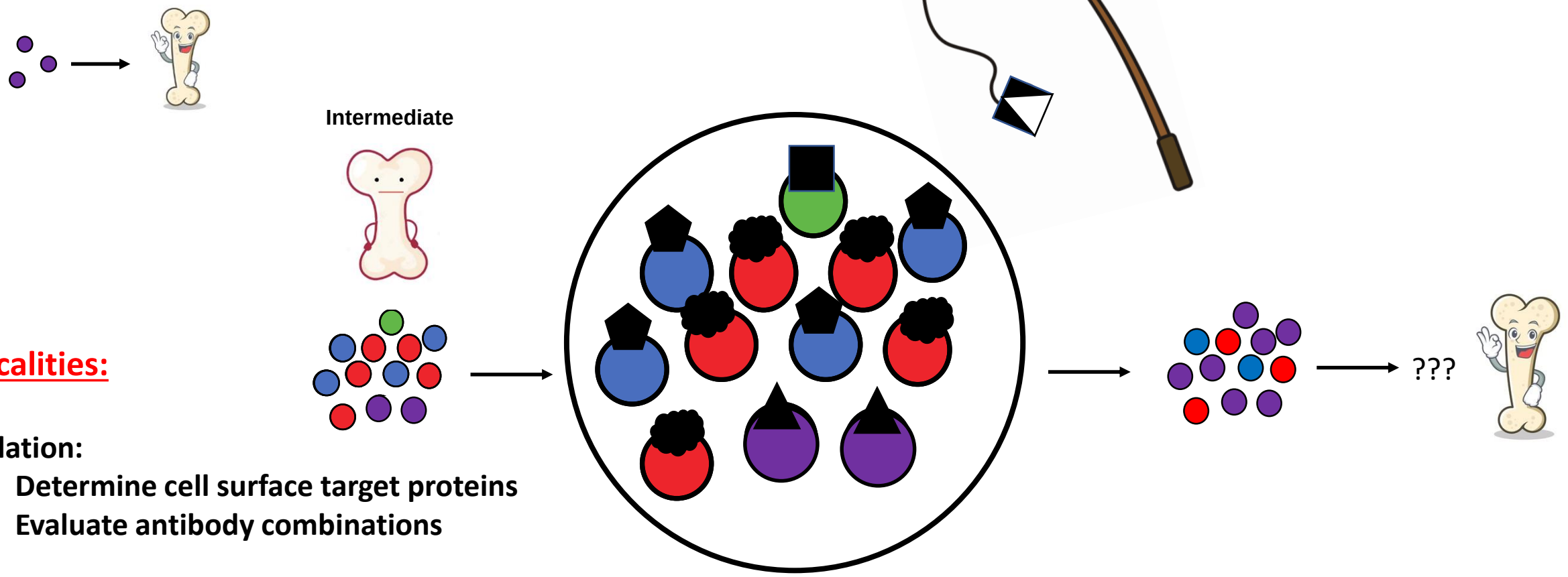
Study design - verification



Practicalities:

1) **Isolation:**
   - **Determine cell surface target proteins**
   - **Evaluate antibody combinations (A)**

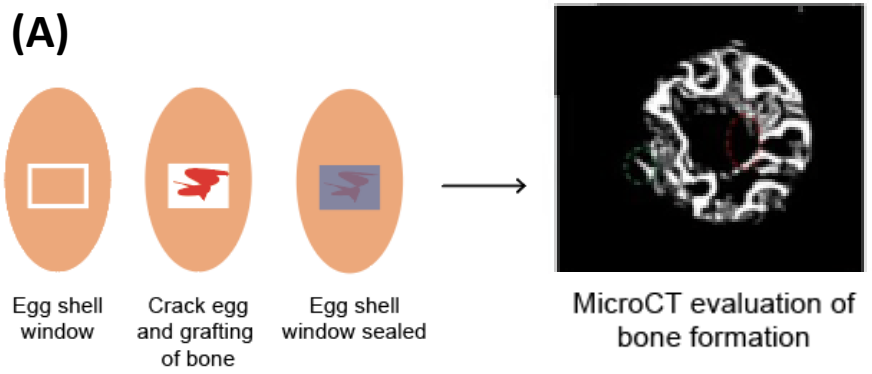
Study design - verification



Practicalities:

1) **Isolation:**
   - **Determine cell surface target proteins**
   - **Evaluate antibody combinations**

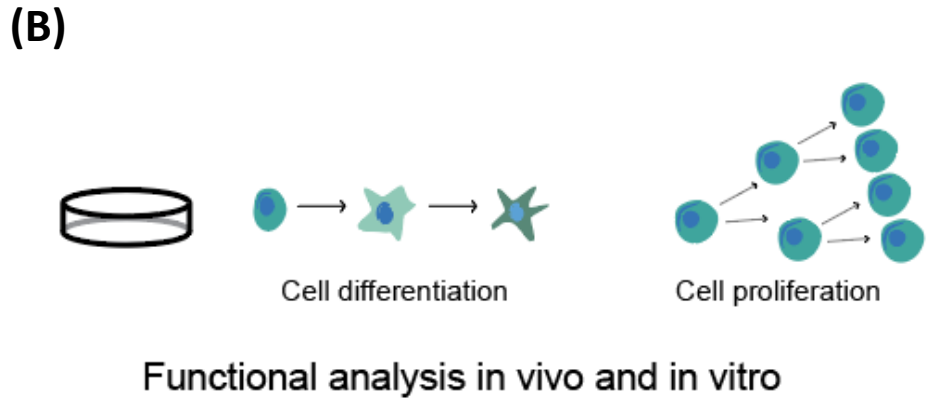
Study design - verification



Practicalities:

1) **Isolation:**
   - **Determine cell surface target proteins**
   - **Evaluate antibody combinations**

2) **Intrinsic osteogenic potential determination:**
   - ***In vivo* differentiation (A)**
   - ***In vitro* differentiation (B)**



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