Labeling of porcine mesenchymal stromal cells (MSCs) with Indium-111-oxine

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Abstract

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Protocol

Buffer Indium-111-oxine with Tris buffer to increase the pH of Indium-111-oxine to 7-8
Step 1.

- Indium-111-oxine was supplied by Mallinckrodt Medical B.V., Petten, The Netherlands

SAFETY INFORMATION
Always be careful when working with radioactive substances. Only allow trained personnel to perform this kind of work.

Centrifuge the MSCs suspended in culture medium for 5 minutes at 300 G (1224 rpm)
Step 2.

Remove and discard supernatant. Be careful not to touch the cell pellet
Step 3.

Resuspend the MSCs in 1 milliliters of Hanks buffered saline solution
Step 4.

- Hanks buffer used: Hank's Balanced Salt Solution 1x $CaCl_2$+ $MgCl_2$+ (Life Technologies Corp, Grand Island, NY, USA)

Place the MSCs-Hanks buffer suspension in a lead container and add ~40 megabecquerel (MBq) of buffered Indium-111-oxine
Step 5.

- Indium-111-oxine was supplied by Mallinckrodt Medical B.V., Petten, The Netherlands

Incubate for 20 minutes at room temperature (20 degrees Celcius)
Step 6.

After 20 minutes of incubation, centrifuge the MSC-Indium mixture for 5 minutes at 300 G (1224 rpm)
Step 7.

Measure the radiation coming from the vial containing the cell pellet and supernatant. Then remove the supernatant with a syringe and measure the signal coming from the cell pellet, syringe and supernatant seperately with a dose calibrator to calculate the labeling efficiency

Step 8.

- Divide the radioactive signal coming from the cell pellet by the signal coming from the complete vial to calculate the labeling efficiency.

- For instance: if the complete vial contains 10 MBq of radioactivity and the cell pellet contains 9 MBq of radioactivity, the labeling efficiency is (9 MBq / 10 MBq)*100 = 90%

Dissolve the pellet in 10 milliliters of Hanks buffered saline solution

Step 9.

- Hanks buffer used: Hank's Balanced Salt Solution 1x $CaCl_2$+ $MgCl_2$+ (Life Technologies Corp, Grand Island, NY, USA)

Centrifuge the pellet + 10 milliliters of Hanks buffered saline solution for 5 minutes at 300 G (1224 rpm)

Step 10.

Measure the radioactive signal coming from the vial containing the cell pellet and 10 milliliters of Hanks buffered saline solution. Then remove the supernatant with a syringe and measure the signal coming from the cell pellet, syringe and supernatant seperately with a dose calibrator to calculate the labeling efficiency

Step 11.

Repeat step 10, 11, and 12 a total of 3 to 4 times until a labeling efficiency of 90 - 95% is achieved

Step 12.

Add 10 milliliters of phosphate buffered saline to the cell pellet and dissolve the pellet. The cell suspension is now ready for administration

Step 13.