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# Cell seeding

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Works for me

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PMAT0001

## PROTOCOL CITATION

PMAT0001 2020. Cell seeding. [protocols.io](https://protocols.io)  
<https://protocols.io/view/cell-seeding-bnmbmc2n>

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43395

## MATERIALS

NAME	CATALOG #	VENDOR
1X Dulbecco's Phosphate Buffered Saline (DPBS)	14190094	Thermo Fisher Scientific
DMEM	11885	Invitrogen - Thermo Fisher

## BEFORE STARTING

- Aseptic techniques\*\*\*
- Wipe down hood and every item introduced into the hood with 70% ethanol

- 1 Take cells from the freezer and thaw for 00:02:00 in the 37 °C water bath. 2m
- 2 Take a T75 plate and place all needed items in ethanol-sterilized laminar flow hood.
- 3 Draw the needed amount of cells into a 50mL centrifuge tube.
- 4 Add about 7 mL of PBS into the centrifuge tube.
- 5 Spin the mixture in a centrifuge for 00:03:00 at 1500rpm. 3m

- 6 Decant supernatant and add PBS to the cell pellet once again.
- 7 Resuspend cell pellet with the PBS using a micropipette.
- 8 Repeat steps 5 to 7.
- 9 Add DMEM into centrifuge tube and resuspend cell pellet.
- 10 Add DMEM to the T75 plate (sufficient to cover the entire surface area, about 5 mL ).
- 11 Add 5 mL of the cell mixture into the T75 flask.
- 12 Incubate T75 flask at 37 °C , 5% CO<sub>2</sub>.