Standard Operating Procedure CellCompact 33

Macro perfusion SUB setup for prep of CellTank / CellCore spec as of 2013.

Purpose

Test in order to find relevant candidate of CellCore scaffolds for harboring the relevant cell line for perfusion cultivation in CellTank. In practice not needed to be performed under sterile conditions. You can easily perform 2-5 tests per day.



One CellVessel 21-0250 STR connected in series with one or more CellCompact Macro Perfusion SUBs and a peristaltic pump able to re-circulate 100 ml/min media.

Example

- Bioreactor was "CellCompact 33-0015-09" with 8 cm2 inlet surface area
- Scaffold sample #09 designed for suspension CHO cell line perfusion
- 150 ml liquid PBS media in STR 21-0250
- >100 mio mammalian cells total inoculated into STR 21-0250
- 100 ml/min flow set on hose pump
- Alternatively a bio mass sensor installed in STR

| Time minutes | Cell counts before - mio/ml | Cell counts after - mio/ml | Bio mass pF/cm |
|-----------------|-----------------------------|----------------------------|-------------------|
| 0 | ? | ? | Ş |
| 1 | ? | ? | Ş |
| 2 | ? | ? | Ş |
| 5 | ? | ? | Ş |
| 10 | ? | ? | Ş |
| 20 | ? | ? | ? |
| 40 | ? | ? | ? |
| 80 | ? | ? | ? |

Procedure:

- Set the stirrer table to a suitable rpm which insure cells are suspended such as 200-300 rpm.
- Use a stop watch for precise on/off operation of the hose pump
- Operate hose pump sequentially and take samples with a syringe in between pump operation.
- Clean the hose by drawing 5 ml into the syringe and return the volume to the STR

- Sample with the 10 ml syringe and draw slowly some ml from the port after the (MPB) and from the STR and fill sample volume into respective sample containers
- Lock the hose clamp after each sampling
- Return excess sample volume, what's not use for cell counting to the STR in order to reduce the liquid loss.
- The first couple of minutes the matrix will attract mostly all the cells passing through the MPB as to the electrostatic forces.
- Fill in the table and return this to CerCell

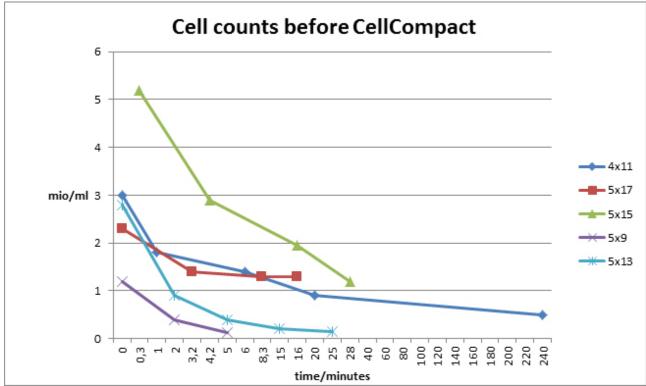


Table illustrate the time it takes for the cells to be filtered out of the re-circulation media and harboured in the scaffold.

Use

- NucleoCounter with 5,000 cell/ml detection limit
- Schärfe particle counter with 5,000 cell/ml detection limit
- Millipore handheld cell counter

END