

Transfection of Mouse Embryonic Fibroblasts

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Introduction:

Since it is really difficult to transfect MEFs, we just wanted to try whether or not this was possible using Metafectene Pro

Materials and methods:

- MEFs (passage 20)
- 6 well plates
- Glass coverslips
- DMEM / F10, 10% serum, Pen / Strep
- Optimem

Experimental procedures / transfection protocol:

-MEFs were cultured in 6 well plates with a glass non treated coverslip in each well.

-One day before transfection cells were seeded. Cells were transfected at 50% confluency.

Per well:

-4 microliter MetafectenePRO was mixed with 96 microliter Optimem.

-1 microgram of Plasmid (in this case containing GFP-CLIP170) was diluted in 99 microliter Optimem

-The MetafectenePRO/optimem and the DNA/optimem were mixed gently, and left at roomtemperature for 20 minutes

-Complexes were added to the well (containing 1,5 ml medium) in a drop wise manner

-After 4 hrs, the medium was refreshed

-Cells were imaged 24 hours after transfection

Results and discussion:

We imaged the cells 24 hrs after transfection. The transfection efficiency was around 50%. The cells looked normal.

Conclusion / summary:

Metafectene PRO seems to work very well !!

I recommend it to everyone who wants to transfect Mouse Embryonic Fibroblasts