

HeLa transfection efficiencies using Metafectene EASY

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Introduction: The aim of this report is to test Metafectene EASY transfection reagent on HeLa cells.

Cells and plasmids: HeLa cells were grown in DMEM supplemented with 10% FBS, 2 mM glutamine, 100 U/ml penicillin and 10 µg/ml streptomycin, in a 5% CO₂ incubator at 37°C. Cells were transfected with the mammalian expression vector pEGFP-C1.

Experimental procedures / transfection protocol: We prepared 250 µl of a cell suspension in serum- and antibiotic-free medium with a concentration of $7 \cdot 10^5$ cells/ml and we used different amounts of transfection reagent and/or DNA, or with different transfection reagent/DNA ratio as indicated below:

2 µl: 0,5 µg
1 µl: 1 µg
2,5 µl: 2,5 µg
5 µl: 5 µg

In brief, we pipetted Metafectene EASY in 50 µl of 1x PBS and mixed gently by pipetting. Then we added the DNA and gently mixed the sample in PS (Polystyrene) tubes. After a 15 min incubation at room temperature, the mixture was added to the 250 µl of cell suspension and cells were subsequently plated, and incubated at 37°C in a CO₂ containing atmosphere. 6 hours after plating, the transfection mixture was removed and replaced it with fresh complete medium. After additional 18 hours, transfection efficiency was determined by counting GFP-expressing cells using a fluorescence microscope.

Results:

Table 1.

Cells density(cells/well)	µl Metafectene EASY	µg DNA	Transfection efficiency
$1,8 \times 10^5$	2	0,5	15%
$1,8 \times 10^5$	1	1	21%
$1,8 \times 10^5$	2,5	2,5	36%
$1,8 \times 10^5$	5	5	49%

As indicated in Table 1, we were able to achieve a transfection efficiency of about 50% by plating a $7 \cdot 10^5$ cells/ml cell suspension using 5µg DNA: 5µl Metafectene EASY ratio.

Similar results were obtained using Metafectene PRO (at a different DNA:reagent ratio) following Biontex instructions, after 24 and 48 hours of transfection, as shown in table 3.

Table 3.

Transfection Reagent	Cells density(cells/well)	Metafectene EASY: DNA ratio (µl:µg)	Transfection efficiency 24h	Transfection efficiency 48h
Metafectene EASY	1,8 x 10 ⁵	5:5	49%	46%
Metafectene PRO	1,8 x 10 ⁵	2:0,5	50%	48%

Conclusion:

We were able to get similar transfection efficiencies (around 50%) with Metafectene Easy compared to Metafectene Pro. The great advantage of Metafectene easy is that can be used on cell suspensions thus after detaching cells with trypsin. Therefore this protocol allowed us to save at least one day as with Metafectene Easy we don't need to plate the cells, wait for them to attach (1-2 days) and then transfect them. Indeed, cells can be transfected immediately before plating in contrast to what happens with most transfection reagents including Metafectene Pro.