

Transfection with CHO- and Hek293- Cells

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Introduction:

cell lines = CHO
 Hek293

Both easy to transfect and Hek is human origin

Experimental procedures / transfection protocol:

optical cell density at timepoint of transfection: CHO 90% confluent and Hek 80% confluent

for 1x6well: 4ug DNA + 100ul optimem in tube 1

 16ul or 24ul METAFECTENE PRO + 100ul optimem in tube2

mix => incubate 20min => add to cells

change medium after 4h after transfection

measure the expression efficiency after 24 and 48h.

Results and discussion:

the ratio DNA: METAFECTENE PRO 1:7 was much better than 1:5

expression efficiency after 48h was much better than 24h

for a single GFP construct in pmax-vector (Amaxxa) with ratio 1:7:

after 24h in Hek = 50-60% positive cells	after 24h in CHO = 40-50% positive cells
after 48h in Hek = 90% positive cells	after 48h in CHO = 70% positive cells

for my gene of interest (7kb) in peGFP-C1 as GFP fusion construct (ratio 1:7):

after 24h in Hek = 15%	after 24h in CHO = 5%
after 48h in Hek = 30%	after 48h in CHO = 20%

for the same gene of interest (7kb) as RFP fusion construct (ratio 1:7):

after 24h in Hek = 15-20%	after 24h in CHO = 5%
after 48h in Hek = 40-50%	after 48h in CHO = 20%

for another gene of interest (3kb) in peGFP-C1 as GFP fusion construct (ratio 1:7):

after 24h in Hek = 30%	after 24h in CHO = 5%
after 48h in Hek = 60-70%	after 48h in CHO = 20%

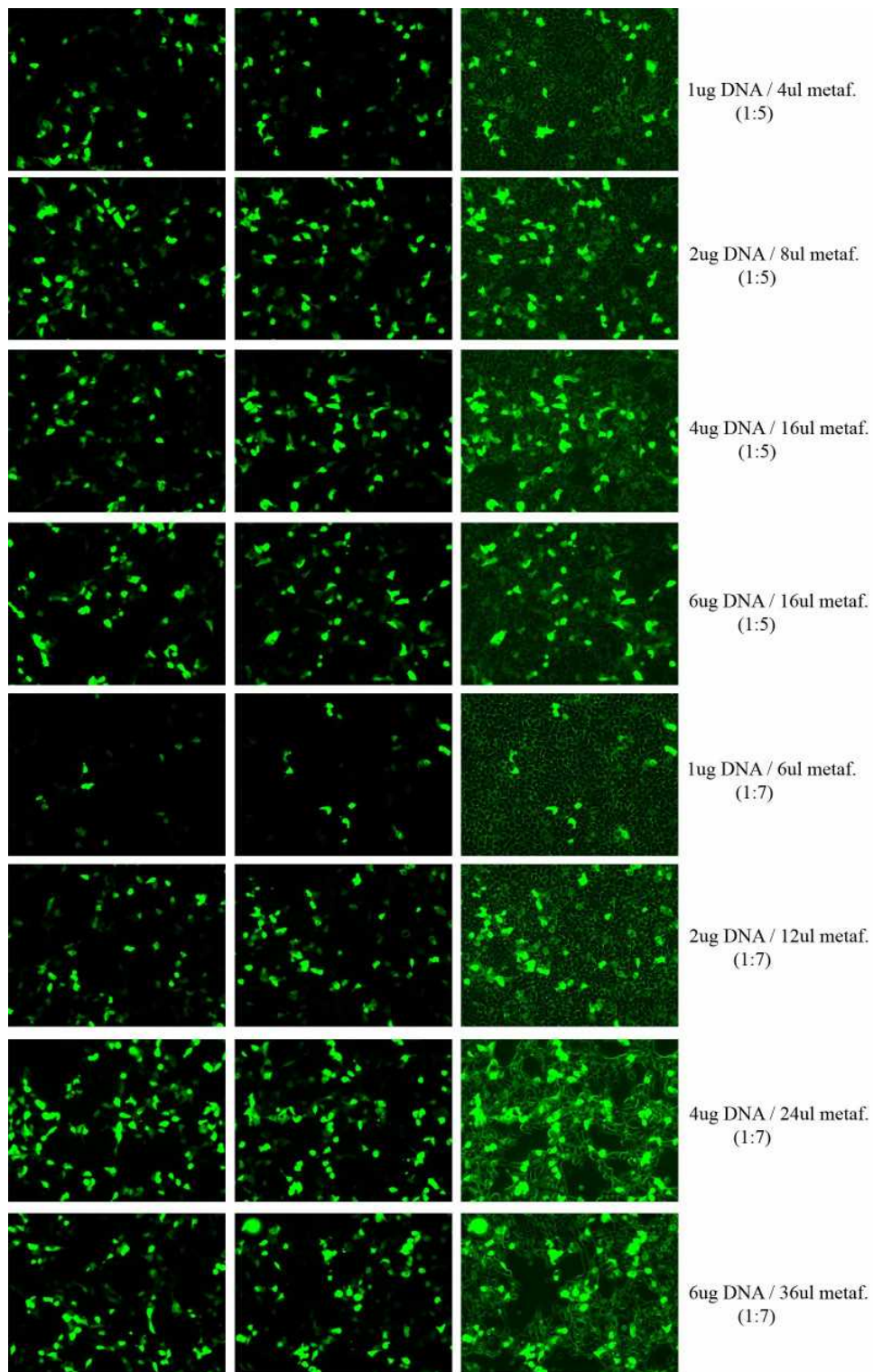
for another gene of interest (3kb) in peGFP-C1 as GFP fusion construct (ratio 1:7):

after 24h in Hek =40-50%	after 24h in CHO = 40%
after 48h in Hek = 70%	after 48h in CHO = 40%

Conclusion / summary:

METAFACTENE PRO is better than Fugene, Lipofectamine, Lipofectamine2000.
METAFACTENE PRO, PEI and Hifect is the same

Appendix: Tables and/or figures:



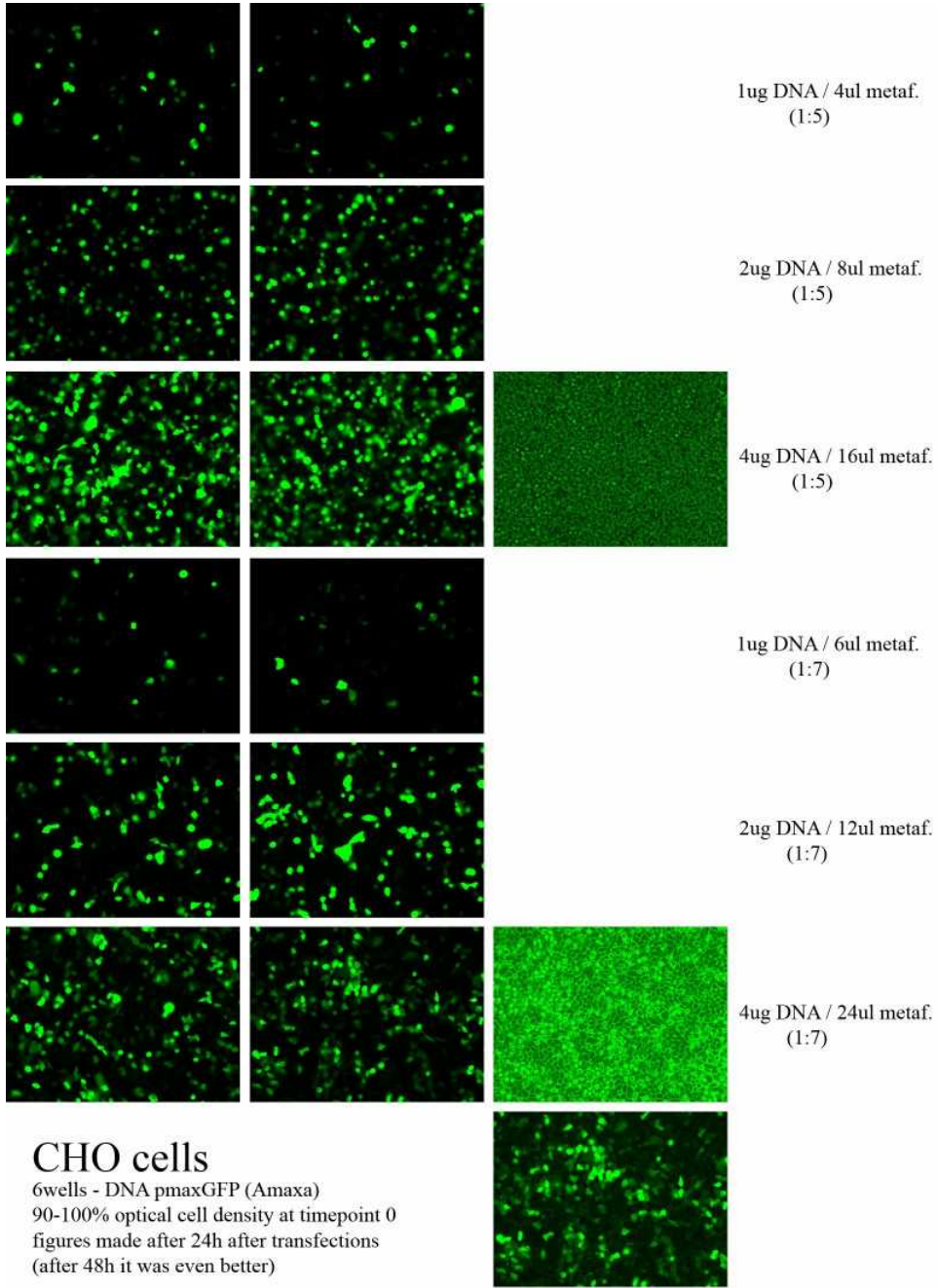
Hek293 cells

6wells - DNA pmaxGFP (Amaya)

90% optical cell density at timepoint 0

figures made after 24h after transfections

(after 48h it was even better=> up to 90% for cond 7+8; my impression; I haven't count)



CHO cells

6wells - DNA pmaxGFP (Amaxa)
 90-100% optical cell density at timepoint 0
 figures made after 24h after transfections
 (after 48h it was even better)