

Testing of different transfection reagents:

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cells tested:

- A431: epidermal carcinoma, monolayer, expressing EGF-R and c-erb-B2
 - HT 29: colon carcinoma, monolayer, expressing 17-1A
 - CHO: chinese hamster ovary
 - BHK: baby hamster kidney,
 - A293: human transformed kidney,
- test in 12 well plates with 8×10^4 cells/ml/well resulting in 50% confluency after one night

transfection substances tested:

- Dendrimer S
- METAFECTENE
- Lipid L

working procedures:

1,5 µg pcDNA3.1-mH-lacZ each

☞☞Dendrimer S:

0,75 µl = 1,5 µg DNA in 75 µl serumfree medium mixed with 5; 7,5; 10 µl Superfect
10 min at rt
add serum containing medium up to 400 µl and incubate for 5 h at 37°C, 5% CO₂
aspirate the content of the wells and add 1 ml serumcontaining medium for o/n incubation

☞☞METAFECTENE:

0,75 µl = 1,5 µg DNA in 30 µl serumfree medium mixed with 2,5; 5; 7,5; 10 µl
compound B in 30 µl serumfree medium
15 min at rt
add serum containing medium up to 400 µl and incubate for 5h at 37°C, 5% CO₂
add 600 µl serum containing medium for o/n incubation

☞☞Lipid L:

0,75 µl = 1,5 µg DNA in 75 µl serumfree medium mixed with 2; 7,5; 12; 18 µl Lipid L in
75 µl serumfree medium
20 min at RT.
add serumfree medium up to 400 µl and incubate for 5 h at 37°C, 5% CO₂
add 600 µl serum containing medium for o/n incubation

the next day the wells were washed with 1 ml PBS and stained with X-gal, counting of stained cells

conclusion resulting from this single test (crude data see next page):

METAFECTENE is over all less toxic than the other reagents.

METAFECTENE seems to be the best transfection reagent for all tested cell lines.

results (% of blue cells):

A293								
Dendrimer S		METAFFECTENE	2,5 µl	31%	Lipid L	2 µl	29%	
	5µl		79% tox	5 µl		85%	7,5 µl	41% tox
	7,5 µl		71% tox	7,5 µl		86%	12 µl	57% tox
	10 µl		42% tox	10 µl		89%	18 µl	36% tox
A431								
Dendrimer S		METAFFECTENE	2,5 µl	2%	Lipid L	2 µl	0,5%	
	5µl		15%	5 µl		11%	7,5 µl	3%
	7,5 µl		15% tox	7,5 µl		13%	12 µl	2,5% tox
	10 µl		16% tox	10 µl		17%	18 µl	0,7% tox
HT 29								
Dendrimer S		METAFFECTENE	2,5 µl	0,5%	Lipid L	2 µl	0,2%	
	5µl		1%	5 µl		0,6%	7,5 µl	0%
	7,5 µl		0,3% tox	7,5 µl		0,6%	12 µl	0,1% tox
	10 µl		0,8% tox	10 µl		0,7%	18 µl	0% tox
CHO								
Dendrimer S		METAFFECTENE	2,5 µl	65%	Lipid L	2 µl	47%	
	5µl		71%	5 µl		77%	7,5 µl	2% tox
	7,5 µl		75%	7,5 µl		81%	12 µl	3% tox
	10 µl		63% tox	10 µl		91%	18 µl	0,2% tox
BHK								
Dendrimer S		METAFFECTENE	2,5 µl	59%	Lipid L	2 µl	12%	
	5µl		86%	5 µl		93%	7,5 µl	89% tox
	7,5 µl		83% tox	7,5 µl		95%	12 µl	7% tox
	10 µl		78% tox	10 µl		94% tox	18 µl	3% tox