

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 8x10⁴ cells/ml/well resulting in 50% confluence after one night

transfection substances tested:

- Dendrimer S
- METAFECTENE
- Lipid L

working procedures:

1,5 µg pcDNA3.1-mH-lacZ each

~~xx~~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

~~xx~~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

~~xx~~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):

METAFFECTENE is over all less toxic than the other reagents.

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

results (% of blue cells):

A293							
Dendrimer S		METAFFECTENE		Lipid L			
5µl	79% tox	2,5 µl	31%	2 µl	29%		
7,5 µl	71% tox	5 µl	85%	7,5 µl	41% tox		
10 µl	42% tox	7,5 µl	86%	12 µl	57% tox		
		10 µl	89%	18 µl	36% tox		

A431							
Dendrimer S		METAFFECTENE		Lipid L			
5µl	15%	2,5 µl	2%	2 µl	0,5%		
7,5 µl	15% tox	5 µl	11%	7,5 µl	3%		
10 µl	16% tox	7,5 µl	13%	12 µl	2,5% tox		
		10 µl	17%	18 µl	0,7% tox		

HT 29							
Dendrimer S		METAFFECTENE		Lipid L			
5µl	1%	2,5 µl	0,5%	2 µl	0,2%		
7,5 µl	0,3% tox	5 µl	0,6%	7,5 µl	0%		
10 µl	0,8% tox	7,5 µl	0,6%	12 µl	0,1% tox		
		10 µl	0,7%	18 µl	0% tox		

CHO							
Dendrimer S		METAFFECTENE		Lipid L			
5µl	71%	2,5 µl	65%	2 µl	47%		
7,5 µl	75%	5 µl	77%	7,5 µl	2% tox		
10 µl	63% tox	7,5 µl	81%	12 µl	3% tox		
		10 µl	91%	18 µl	0,2% tox		

BHK							
Dendrimer S		METAFFECTENE		Lipid L			
5µl	86%	2,5 µl	59%	2 µl	12%		
7,5 µl	83% tox	5 µl	93%	7,5 µl	89% tox		
10 µl	78% tox	7,5 µl	95%	12 µl	7% tox		
		10 µl	94% tox	18 µl	3% tox		