## LIPOFECTION

- Dissolve Tfx<sup>TM</sup> -10, -20 or -50 in 400  $\mu$ l nuclease-free water. Vortex and incubate at 65 °C for 1 min.

Store at -20 °C.

- Culture cells to 80% confluency on 24-well plate.
- Prepare complete medium without serum.
  Add DNA to medium, mix well
  Add Tfx<sup>TM</sup> at ratio Tfx<sup>TM</sup>: DNA = 3:1 according to table:

	Amount of DNA per well			
	<u>0.25 µg</u>	0.50 <u>µ</u> g	0.75 µg	1.00 µg
medium	1.4 ml	1.4 ml	1.4 ml	1.4 ml
DNA	1.8 <i>μ</i> g	3.5 <i>µ</i> q	5.3 <i>μ</i> g	7.0 μg
$Tf x^{TM}$	7.9 μl	15.8 μl	23.6 <i>μ</i> l	$31.5 \mu$

(ratio  $Tfx^{TM} : DNA = 2 : 1 \text{ or } 4 : 1 \text{ can also be tried}$ )

- Incubate medium/DNA/Tf $x^{TM}$  lipofection mix at RT for 15 min.
- Incubate cells in 200  $\mu$ l lipofection mix at 37 °C for 30 min.
- Add 1 ml complete medium (with serum).
- Incubate cells for 24-48 h.
- A LacZ reporter plasmid can be used to measure transfection efficiency.

NOTE

Optimal conditions for ES cells:  $0.75-1.00 \mu g$  of DNA per well

 $Tfx^{TM} - 50 : DNA = 3:1$