jetPRIME® Transfection Reagent

Short Protocol – DNA Transfection

Day 0: Cell Seeding

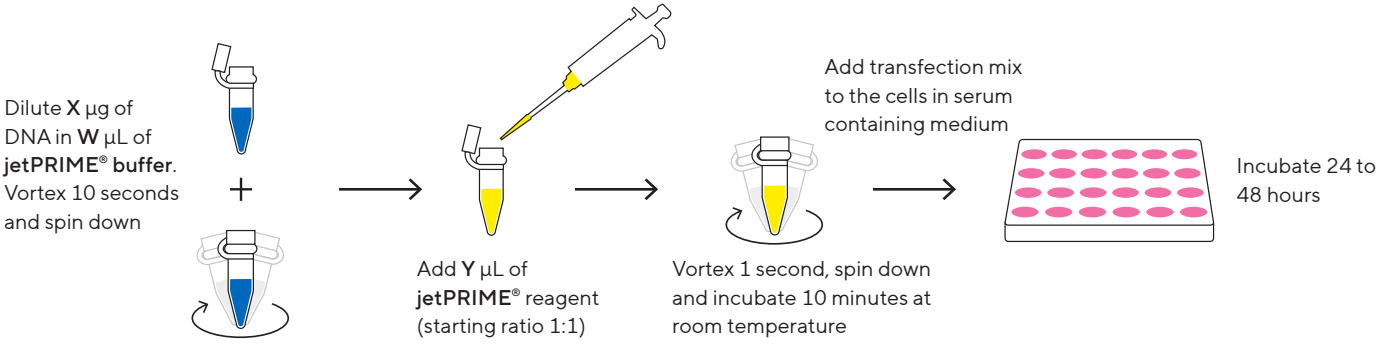
- Seed cells in **V** mL of cell growth medium according to the table below

Culture vessel	Number of cells*	V = volume of medium during transfection
96-well	7,500 – 10,000	0.1 mL
24-well	50,000 – 80,000	0.5 mL
12-well	80,000 – 150,000	1 mL
6-well/35 mm	150,000 – 250,000	2 mL
100 mm/flask 75 cm²	$1 \times 10^6 - 2 \times 10^6$	10 mL

Quantities per well, dish or flask.

Day 1: Transfection

- Perform transfection **in the presence of serum**
- Use **jetPRIME® buffer only**
- Transfect cells at **60 – 80% confluency**



Culture vessel	1. W = volume of jetPRIME® buffer [µL]	2. X = amount of DNA added [µg]	3. Y = volume of jetPRIME® reagent [µL]
96-well	10	0.1	0.2
24-well	50	0.5	1
12-well	75	0.8	1.6
6-well/35 mm	200	2	4
100 mm/flask 75 cm²	500	10	20

Quantities per well, dish or flask.

Day 2 – 3: Measure Gene Expression

See back page for optimization tips.

Download complete protocol on sartorius.com

Short Protocol – Optimization Tips (DNA)

Protocol Optimization

- Test different DNA amounts: X, 0.5X and 1.5X.
- Test different DNA/jetPRIME® ratios, 1:2 to 1:3.
- For cell specific protocols, visit www.sartorius.com.

Culture vessel	W = volume of jetPRIME® buffer [µL]	X = amount of DNA added [µg]	Y = volume of jetPRIME® reagent [µL]
96-well	10	0.05 – 0.20	0.10 – 0.60
24-well	50	0.25 – 0.75	0.50 – 2.25
12-well	75	0.4 – 1.2	0.8 – 3.6
6-well/35 mm	200	1 – 3	2 – 9
100 mm/flask 75 cm²	500	5 – 15	10 – 45

Quantities per well, dish or flask.

For HEK-293 and HeLa cells, you may decrease the DNA amount to 0.5X and use the 1:2 DNA/jetPRIME® ratio.

Tips to Increase Cell Viability of Sensitive Cells

- Replace medium 4 hours after transfection.
- Decrease DNA amount to 0.5X while maintaining the DNA/jetPRIME® ratio previously used.
- Analyze transfection at an earlier time point (24 hours after transfection instead of 48 hours for instance).
- Check that the target gene does not affect cell viability.

Good DNA Transfection Practices

- Store appropriately jetPRIME® (5 ± 3 °C).
- Ensure that cells have been passaged more than twice and less than 20 times prior to transfection.
- Discard overconfluent cells.
- Regularly check for mycoplasma contaminations.
- Use a reporter gene to set up and optimize transfection conditions.
- Serum quality may drastically affect transfection efficiency. When purchasing a new batch of serum or trypsin, check cell viability as well as transfection efficiency.

⚠ NOTE:

jetPRIME® is also recommended for DNA | siRNA co-transfection. Please refer to the complete protocol on www.sartorius.com.

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Status: 09 | 2024