

jetPRIME[®] transfection reagent

Short protocol - DNA Transfection

DAY 0: Cell seeding

→ Seed cells in **V** mL of serum containing medium according to the table below

Quantities per well, dish or flask

Culture vessel	Number of cells	V = volume of growth medium for cell seeding
96-well	7500 - 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 cm ²	1 x 10 ⁶ - 2 x 10 ⁶	10 mL

DAY 1: Transfection

→ Perform transfection **in the presence of serum**

→ Use **jetPRIME[®] buffer only**

→ Transfect cells at **60-80% confluency**

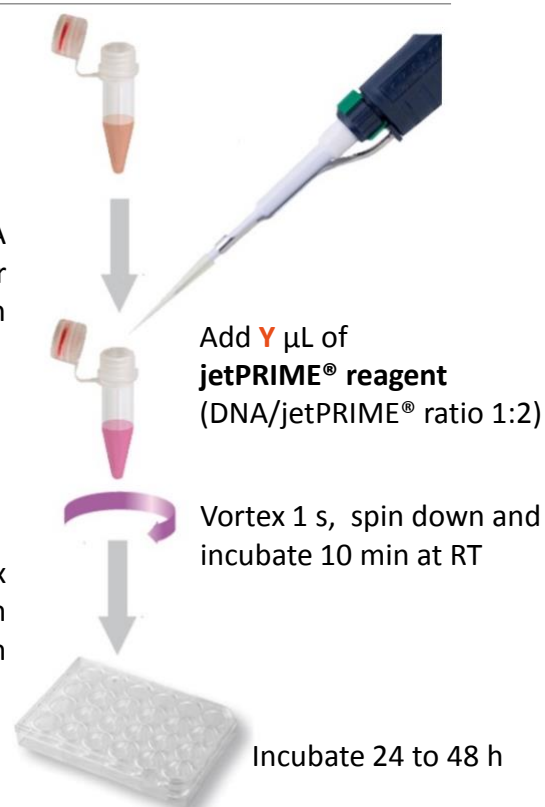


Watch the video «transfection using jetPRIME[®]» on YouTube!

<http://www.youtube.com/watch?v=G39wNXaZPX4>

Dilute **X** µg of DNA
in **W** µL of jetPRIME[®] buffer
Vortex 10 s and spin down

Add transfection mix
to the cells in serum
containing medium



Quantities per well, dish or flask

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

DAY 2-3: Measure gene expression

See back page for optimization tips

Download complete protocol on <http://www.polyplus-transfection.com/resources/product-literature/>

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Short protocol - Optimization Tips (DNA)

+ Protocol Optimization

- + Check our online Cell Transfection Database for cell specific protocols at: <http://www.polyplus-transfection.com/resources/cell-transfection-database/>
- + Test different DNA amounts: X, 0.5X and 1.5X.
- + Test different DNA/jetPRIME[®] ratios, 1:2 to 1:3.



Quantities per well, dish or flask

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

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 h after transfection.
- + Decrease DNA amount to 0.5 X while maintaining the DNA/jetPRIME[®] ratio previously used.
- + Analyze transfection at an earlier time point (24 h after transfection instead of 48 h 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 virus production and DNA/siRNA cotransfection, please refer to the complete protocol available online at: <http://www.polyplus-transfection.com/resources/product-literature/>