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

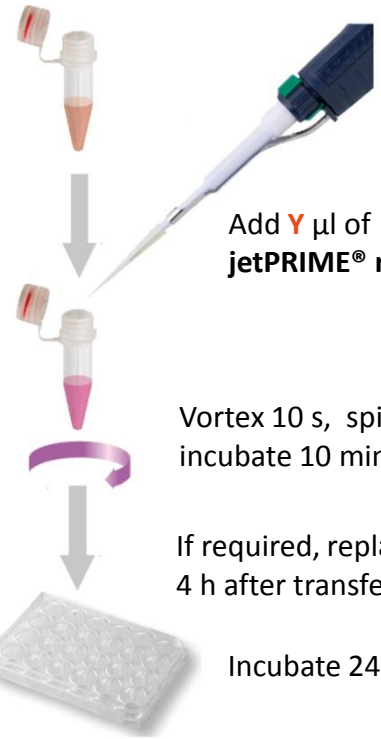
Quantities per well, dish or flask

Culture vessel	Number of cells	V volume of serum containing medium during transfection
24-well	50 000 – 80 000	0.5 ml
6-well / 35 mm	150 000 – 250 000	2 ml
100 mm / flask 75 cm²	1 000 000 – 2 000 000	10 ml

DAY 1: Transfection = 1:2 DNA to jetPRIME® reagent ratio

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

Dilute X µg of DNA
in W µl of jetPRIME® buffer
Vortex 10 s and spin down



Add Y µl of
jetPRIME® reagent

Vortex 10 s, spin down and incubate 10 min at RT

Add transfection mix to the cells in serum containing medium

If required, replace medium 4 h after transfection

Incubate 24 to 48 h

Watch the video «transfection using jetPRIME®» on YouTube!
http://www.youtube.com/watch?v=G39wNXaZPX4

Quantities per well, dish or flask

Culture vessel	W volume of jetPRIME® buffer	X amount of DNA added	Y volume of jetPRIME® reagent
24-well	50 µl	0.5 µg	1 µl
6-well / 35 mm	200 µl	2 µg	4 µl
100 mm / flask 75 cm²	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/

Polyplus transfection® THE DELIVERY EXPERTS

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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
24-well	50 µl	0.25 – 0.75 µg	0.5 – 2.25 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 after 4 h
- Decrease DNA amount to 0.5X
- 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® (4°C) and the DNA
- 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/

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