

Plasmid transfection protocol

FOR TECREA PRODUCTS TNP-250, TNP-500, TNP-1000 AND TNP-10000

Product information

Nanocin PLASMID is an innovative transfection reagent dedicated to the efficient and non-toxic transfection into a range of mammalian cells, including primary cells and other sensitive cells. **For research use only.**

Quality control

Each batch is tested using biophysical methods and by ensuring efficient delivery of GFP encoding plasmid into HEK293T cells, assessed by both microscopy and flow cytometry.

Shipping, storage and shelf life

Nanocin products are shipped at room temperature, stored at 2-8°C and are stable for at least one year. The expiry date is indicated on the tube label.

Safety

Nanocin PLASMID shows very low toxicity in a range of assays. See our MSDS for more details and handling instructions. https://www.tecrea.com/product/nanocin-plasmid/

Technical resources and scientific advice

Tecrea provides extensive technical support and scientific guidance for any experiments involving **Nanocin** products. Please contact us for more information. info@tecrea.com / Frequently asked questions

Helpful information

Save time and increase experiment efficiency with **Nanocin PLASMID's** rapid protocol (see next page).

Nanocin PLASMID products are effectively non-toxic, meaning they can facilitate multiple transfections.

Nanocin PLASMID products are for research use only but compatible with clinical development, meaning you can carry research from lab to clinic with confidence.

Contents and ordering

Unit size (mL)	Transfections*	Cat No.
0.25	50-75	TNP-250
0.5	100-150	TNP-500
1.0	200-300	TNP-1000
10.0	2000-3000	TNP-10000

*Approximate number based on 12-well plate

Related products

Product

Nanocin RNA

Nanocin PRO (for protein & peptide delivery)

Nanocin SM (for small molecule delivery)

Protocol overview



Cat No.

TNR-250, TNR-500, TNR-1000

TNPRO-250, TNPRO-500

TNSM-250, TNSM-500



Standard PLASMID TRANSFECTION PROTOCOL

Rapid PLASMID TRANSFECTION PROTOCOL

Use this protocol to transfect plasmid DNA into mammalian cells after the cells have recovered from splitting or seeding. The details here are for a 12-well plate format. For other formats, see table below. All volumes are given per well.

Set-up

- Seed and grow cells to 60-80% confluence (for low-confluence experiments see notes the table below*).
- Vortex Nanocin PLASMID reagent for 10seconds and centrifuge briefly.

START Transfection

Step 1. Prepare transfection mixture for 12-well plate (example): Dilute 1 μ g plasmid DNA in Transfection Buffer to a final volume of 47.5 μ l. <u>Mix thoroughly</u> by pipetting the full volume up and down 5-10 times. Place tube on ice. Add 2.5 μ l of **Nanocin PLASMID** reagent so volume totals 50 μ l. <u>Mix thoroughly</u>, pipetting the full volume up and down 5-10 times. Incubate for 20 minutes at room temperature.

Step 2. Transfect: Transfer tubes from ice to rack at room temperature. Add 950 μ l of pre-warmed growth medium to each tube prepared in step 1 so the volume total is 1000 μ l in each. <u>Mix thoroughly</u>. Remove old growth media from wells. Immediately add diluted transfection mixture by pipetting gently onto well walls.

Incubate plates as usual for 24-72 hours.

Use this rapid protocol to transfect plasmid into mammalian cells at the time of splitting or seeding. The rapid protocol saves at least one day of experiment time and reduces the process by several steps. The details here are for a 12-well plate format. For other formats, see table below. All volumes given are per well.

Set-up

• Vortex Nanocin PLASMID reagent for 10 seconds and centrifuge briefly.

START Transfection

Step 1. Prepare transfection mixture for 12-well plate (example): Dilute 1 μ g plasmid DNA in Transfection Buffer to a final volume of 47.5 μ l. <u>Mix thoroughly</u> by pipetting the full volume up and down 5-10 times. Place tube on ice. Add 2.5 μ l of **Nanocin PLASMID** reagent so volume totals 50 μ l, <u>Mix thoroughly</u>, pipetting the full volume up and down 5-10 times. Incubate for 20 minutes at room temperature. While the transfection mixture incubates, prepare a cell suspension in growth medium at 4x10⁵ cells/ml (trypsinise first if necessary). Then add 500 μ l of suspended cells to each well.

Step 2. Transfect: Transfer tubes from ice to rack at room temperature. Add 450 μ l pre-warmed growth medium to each tube prepared in step 1 so each tube totals 500 μ l volume, and mix <u>thoroughly</u>. Add drop- by-drop to cells in the well, gently swirling the plate to mix until the volume totals 1ml. Incubate plates as usual for 24-72 hours.

Alternative volumes for other plates' formats

Plate	Confluence	Well surface area	Media (Vol/Well)	Transfection mix volume	Fresh media volume	Plasmid transfection	
						pDNA	Nanocin PLASMID
24-well	30-60%*	$2 \ cm^2$	500 µl	19 µl	481 µl	0.38 µg	0.94 µl
	60-80%	$2 \ cm^2$	500 µl	25 µl	475 μl	0.5 µg	1.25 µl
12-well	30-60%*	4 cm ²	1 ml	38 µl	962 µl	0.75 µg	1.88 µl
	60-80%	4 cm ²	1 ml	50 µl	950 µl	1 μg	2.5 μl
6-well	30-60%*	10 cm ²	2.5 ml	94 µl	2406 µl	1.88 µg	4.69 µl
	60-80%	10 cm ²	2.5 ml	125 µl	2375 µl	2.5 μg	6.25 μl
60-mm	30-60%*	20 cm ²	5 ml	188 µl	4812 µl	3.8 µg	9.4 μl
	60-80%	20 cm ²	5 ml	250 μl	4750 μl	5 µg	12.5 µl

Notes:

• growth medium may be with or without FCS and antibiotics

use transfection mixture within 60 minutes after preparation

• mix thoroughly at all mixing steps by pipetting the full volume up and down 5-10 times

