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NANOPARTICLE-based *In Vivo* Transfection Reagent (cat#5030, 5031, 5032) *In Vivo* Transfection Protocol

Cat. No. 5030	Size: 0.5 ml
Cat. No. 5031	Size: 1.5 ml
Cat. No. 5032	Size: 8.0 ml

Contents and Shipping:

Nanoparticle-based *in vivo* transfection reagent is supplied in liquid form at a concentration of 2.4 mg/ml. Transfection Enhancer is supplied in liquid form at a concentration of 1 mg/ml. Shipped at ambient temperature.

Description:

Nanoparticle-based *in vivo* transfection reagent is a proprietary, animal-origin-free formulation of chemically engineered nanoparticles optimized for siRNA, shRNA, miRNA, and plasmid DNA *in vivo* delivery.

Storage:

Store reagent at 4°C upon receipt. If stored properly, reagent is stable for 6 months.

Product Qualification:

Reagents are tested for functional activity, absence of nuclease contamination and microbial contamination.

Reagent Properties:

Nanoparticle conjugated complexes are stable in serum for at least 16 hours.
Efficient delivery to the lung, liver, pancreas, kidney, and certain tumor types via systemic administration.
Efficient siRNA and plasmid DNA delivery via direct subcutaneous tumor injection (various tumor types).
Minimal toxicity.

Recommended Modes of Administration:

Tail vein systemic intravenous (i.v.) injection.
Direct intratumoral (i.t.) injection.

Intended Use:

For research use only. Not intended for any diagnostic or therapeutic use.

Safety Data Sheet (SDS):

SDS documents are available online at <https://altogen.com>

General Guidelines:

Handle animals and conduct experiments according to national regulations and approvals by the local IACUC and ethics committee. Please note that mouse tail vein injection may require skills, experience, and associated equipment (e.g. restraining device). All persons handling animals should be properly trained.

Guidelines for Small RNA (siRNA, shRNA, microRNA) Administration:

Prepare siRNA stock solution in DNase- and RNase-free water.

Recommended dose of siRNA (per injection):

75-100 µg siRNA with chemical modification (resistant to nuclease degradation *in vivo*)

250-400 µg standard, non-modified siRNA

Recommended Protocol for Systemic Tail Vein (i.v.) Administration (for 1 injection):

1. Dilute 60 µg of plasmid DNA and/or 100 µg of siRNA in 100 µl RNase-/DNase-free water. Vortex gently.
2. Add 100 µl of diluted DNA (and/or siRNA) to the sterile tube containing 50 µl Transfection Reagent.
3. Incubate for 15-20 minutes at room temperature.
4. Add 10 µl of Transfection Enhancer Reagent. Vortex gently.
5. Incubate for 5 minutes at room temperature.
6. Add required amount of sterile solution of 5% glucose (w/v):

Animal body weight (g)	Final injection volume (ml)
10-14	0.2
15-19	0.3
20-24	0.4
25-29	0.5
30-35	0.6

7. Inject animals. Please note that delivery efficiency can be significantly increased by performing secondary injection (at least 12 hours after first injection).
8. Assay for target gene expression. Maximum mRNA expression effect is typically observed 12-36 hours after injection, while maximum effect on protein level is achieved 24-48 hours post-injection.

Recommended Protocol for Direct Intratumoral (i.t.) Administration (for 1 injection):

1. Dilute 60 µg of plasmid DNA and/or 100 µg of siRNA in 50 µl RNase-/DNase-free water. Vortex gently.
2. Add 50 µl of diluted DNA (and/or siRNA) to the sterile tube containing 50 µl Transfection Reagent.
3. Incubate for 15-20 minutes at room temperature.
4. Add 10 µl of Transfection Enhancer Reagent. Vortex gently.
5. Incubate for 5 minutes at room temperature.
6. Inject animals.
7. Assay for target gene expression. Maximum mRNA expression effect is typically observed 12-36 hours after injection, while maximum effect on protein level is achieved 24-48 hours post-injection.

Dosing:

Animal body weight (g)	Transfection Reagent (µl) / per Injection
10 – 35 g (mouse)	50 µl
35 – 200 g (small rodents)	100 µl
200 – 450 g (rat)	250 µl

Limited Use Label License:

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