

EVO Transfer, DNA

Product Number: TR1812

Shipping and Storage

This product is transported at room temperature and stored for a long time at 4°C, with a validity period of 12 months.

This product is safe to use and no biological or chemical toxicity has been found. If accidentally contaminated, rinse with clean water.

Description

EVO Transfer, DNA is a nano polymer transfection reagent developed and synthesized by our company. This reagent is synthesized using nanotechnology and is a new generation of non viral transfection reagents. Due to the application of nanotechnology, EVO Transfer, DNA exhibits excellent low toxicity and high efficiency performance during cell transfection. This product is used for in vivo transfection of DNA.

Features

The method is simple and fast, with no obvious inflammatory response to animals, and is safe for operators.

Application

1. In vivo studies of gene therapy in animals
2. In vivo studies on RNA interference in animals
3. In vivo study of protein function in animals

Note

1. The preparation of transfection complexes is carried out at room temperature. Before operation, please restore the nucleic acid, transfection reagents, diluents, etc. to room temperature. Low temperature operation, long-term storage, minimal or no use of diluents, and mixing nucleic acids and transfection reagents in large reaction systems may cause the aggregation of transfection complexes and result in turbidity. In general, slight turbidity does not affect the transfection effect. If turbidity needs to be avoided, it is recommended to operate at room temperature, ready to use, using diluents, and avoiding large amounts of preparation at once.
2. Nucleic acid dosage. The initial dose for intravenous administration of nucleic acid is 0.625mg/kg. The higher the dosage, the better the effect, with the standard of not causing animal inflammation and death. The following table shows some common dosages and volumes of administration.

Table 1. Dosage and volume of administration

Administration route	Nucleic acid dosage	Maximum administration volume	Administration route
Adult mice	Caudal vein	10-50µg	200µl-600µl
	Ventricle	1-2.5µg	5µl
	Peritoneum	100µg	0.6ml-1ml
	Testis	3-5µg	10µl
Nude mice	Subcutaneous tumor	10-50µg	100µl
Adult rats	Ventricle	2-5µg	20µl
	Caudal vein	500µg-2.25mg	1-2ml
Adult rabbit	Pulmonary and tracheal perfusion	300-700µg	300-700µl

3. Requirements for nucleic acid testing. It is recommended to dissolve with pure water and ensure high purity and endotoxin free. The recommended concentration of nucleic acid is 0.5-1µg/µl. The purity of nucleic acid will greatly affect the transfection efficiency, so it is necessary to use high-purity and high-quality nucleic acid.

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- The amount of EVO Transfer, DNA used. In general, nucleic acid (μg) and EVO Transfer, DNA (μl) are used in a ratio of 1:2.
- Tail vein injection: To maintain the working concentration of the transfection complex and blood isotopes during injection, it is necessary to dilute with solutions such as glucose (preferred) and physiological saline.

The following table shows the composition of each component (nucleic acid concentration $1\mu\text{g}/\mu\text{l}$) when a tail vein injection of 20g body weight mice was administered at a dose of $100\mu\text{l}$ at a dose of $0.625\text{mg}/\text{kg}$.

Table 2. Composition of transfection complexes after tail vein injection of $100\mu\text{l}$

100 μl of transfection complexes	Nucleic acid diluent	12.5 μg (12.5 μl)nucleic acid
		12.5 μl water
		25 μl 10% glucose solution
	Transfection reagent diluent	25 μl transfection reagent
		25 μl 10% glucose solution

- Local injection: First, determine the maximum volume that needs to be finally injected. After determining the maximum volume, the dosage is determined based on the ratio of nucleic acid (μg): transfection reagent (μl)=1:2. For example, when the local injection volume is $30\mu\text{l}$, the composition of each component is determined, with a nucleic acid concentration of $1\mu\text{g}/\mu\text{l}$ (which can be dissolved in physiological saline or pure water).

Note: When not using or using less diluent, mixing nucleic acid and transfection reagents can easily produce turbidity. Slight turbidity does not affect the experiment. If turbidity needs to be avoided, it is recommended to use diluent and avoid preparing in large quantities at once.

- Table 3. Composition of Local Injection $30\mu\text{l}$ Transfection Complex

30 μl of transfection complexes	Nucleic acid solution	8 μg (8 μl)nucleic acid
	Nucleic acid diluent	6 μl
	Transfection reagent	16 μl transfection reagent

Protocol

Taking 12.5 μg of nucleic acid and 25 μl of transfection reagent, with a total injection volume of $100\mu\text{l}$, 20g of mice were injected into the tail vein as an example.

- Dilution of nucleic acid.** Dilute 12.5 μg of nucleic acid with an appropriate amount of endotoxin free pure water to $1\mu\text{g}/\mu\text{l}$ (if the concentration of the nucleic acid solution is small, the injection volume will increase), add 12.5 μl of water, and then add 25 μl of 10% glucose solution (w/v) to the final volume of $50\mu\text{l}$. Mix well.
- Dilution of transfection reagents.** Dilute 25 μl of EVO Transfer, DNA reagent with 25 μl of 10% glucose solution to a final volume of $50\mu\text{l}$, and mix thoroughly.
- Transfection complex formation.** Add the diluted transfection reagent to the diluted nucleic acid solution at room temperature, shake thoroughly and mix well.
- Let it stand at room temperature for 15 minutes.** The prepared transfection complex should be prepared and used immediately, and should not be stored for a long time.
- Animal injection.**

Explanation: 1) In some cases, due to the amount of nucleic acid and transfection reagents used, it may not be possible to maintain a glucose concentration of 5% (or 0.9% of physiological saline) in the injection solution. In this case, it is only necessary to ensure that nucleic acid (μg): transfection reagent (μl)=1:2, and the amount and final concentration of glucose solution increase or decrease moderately, without affecting transfection efficiency.

2) When injecting into the tail vein, please master the injection technique. Generally, the distal one-third of the vein should be used for injection. If strong resistance is felt, please stop the injection and search for the vein again for operation. Do not forcefully push the injection, otherwise it is easy to inject the medication into the tail, causing the tail to ulcerate. After the injection is completed, remove the needle and press the needle hole for more than 10 seconds to prevent the medication from flowing out.

3) A dosage of $0.625\text{mg}/\text{kg}$ is the initial dosage, and if the animal can tolerate it, the dosage can be increased

proportionally; If the animal cannot tolerate it, the dose can be appropriately and proportionally reduced.

4) Local injection, injecting as much medication as possible, is beneficial for improving the transfection effect.

6. **Gene expression detection.** Generally speaking, based on the differences in injection methods and target organs, the gene expression effect is better after 12-48 hours.

Explanation: 1) It is recommended to first use qRT PCR to detect the transfection effect at the molecular level;

2) If protein level detection is required, Western blot method is generally used. At the tissue and organ level, immunohistochemistry method can be used;

3) If using live animal imaging methods, it is recommended to use radioactive isotope methods. Fluorescence excitation methods such as GFP may not be able to observe due to sensitivity issues.

7. **Long term administration.** The optimal time for a drug administration test is 12-48 hours after injection. If long-term effects need to be maintained, multiple injections can be used, with an injection frequency generally every 2-3 days, or it can be appropriately extended to every 7 days according to experimental conditions.

Common problems and solutions

Problem	Reason	Solution
The transfection complex shows precipitation	Dissolve nucleic acid with TE or other substances	Dissolve nucleic acid in pure water
	Mixing nucleic acid and transfection reagents at low temperatures	Nucleic acid and transfection reagents should be mixed at room temperature
	The final concentration of nucleic acid during injection is too high	The final concentration of nucleic acid during injection is $\leq 0.5\mu\text{g}/\mu\text{l}$
	Transfection complexes left for too long	Transfection complexes are best prepared and used immediately
	The reaction system of transfection complex is too large	Suggest starting from a small volume to prepare transfection complexes
Low transfection level	Low dosing	Increase the dosage of medication without causing animal death
	Injection operation error	Inject the medication correctly into the corresponding area
Animals with inflammatory reactions or death	Excessive nucleic acid dosage	Reduce the amount of nucleic acid used, maintain the ratio of nucleic acid to transfection reagents, and simultaneously reduce the amount of transfection reagents used.
	Injection contains endotoxins	Before using endotoxin free nucleic acid injection, use a $0.22\mu\text{m}$ filter to filter and sterilize

Related products

TR1811: Used for in vivo transfection of RNA in animals.

TR2000: Used to transfect long fragments of nucleic acid into cells.

TR1000: Used to transfect short nucleic acids into cells.