



COSMO BIO CO., LTD.  
Inspiration for Life Science

# Antibody Transfection Reagent Ab-Carrier™

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Users Manual  
ver. 1.1

Cat# PTN-P-101-100

Cat# PTN-P-101-25

**Expiry** 6 months after manufacture (4°C)

## **Specifications**

- Endotoxin level < 0.05 EU /  $\mu$ L
  - Antibody transfection activity (relative value)\* 80 ~ 150 %
- \*  $\beta$ -galactosidase-conjugated goat IgG was transfected to HeLa cells and incubated for 4 hrs. Antibody transfection activity was estimated by  $\beta$ -galactosidase activity in lysates of the cells. The values represent relative activity (%) to that of Lot N12J15 as standard.

## **Features**

- Suitable for use with 10 % serum-containing media.
- Ready -to-use. Easy and quick procedure.
- Very low cytotoxicity.
- Suitable for various antibodies (polyclonal, monoclonal).
- Suitable for transfection of fluorescent-labeled or enzyme-conjugated antibodies.

## **Contents**

- Ab-Carrier 12.5  $\mu$ L x 1 (P-101-25), 12.5  $\mu$ L x 4 (P-101-100)
- Storage 4 °C (for long-period storage over 6 months : -20 °C)
- Solution PBS (including 25 mM glycine )

## **Number of reactions (ex. HeLa cell )**

Product Code No.	Standard number of reactions (24 well plate ; 1 $\mu$ g IgG / well)
PTN-P-101-25	25 reactions
PTN-P-101-100	100 reactions

## **Standard protocol (ex. HeLa cell )**

1. Prepare 24 hrs-cultured cells before transfection of antibody.

Culture size	Recommended number of cells
6-well plate	1.0 ~ 2.0 x 10 <sup>5</sup> cells / well
12-well plate	0.5 ~ 1.0 x 10 <sup>5</sup> cells / well
24-well plate	0.25 ~ 0.5 x 10 <sup>5</sup> cells / well
96-well plate	0.5 ~ 1.0 x 10 <sup>4</sup> cells / well

2. Dilute antibody with PBS to adjust 0.1 ~ 0.2 mg / mL.
3. Add Ab-Carrier 0.5  $\mu$ L per 1  $\mu$ g of antibody, mix and leave to stand for 20 min at room temp.
4. Add the reaction mixture to the culture, incubating for 2-4 hrs\* at 37 °C, 5% CO<sub>2</sub> .

Culture size	Recommended amount of Antibody	Recommended amount of Ab-Carrier
6-well plate	2 ~ 4 $\mu$ g IgG / well	2 $\mu$ L / well
12-well plate	1 ~ 2 $\mu$ g IgG / well	1 $\mu$ L / well
24-well plate	0.5 ~ 1 $\mu$ g IgG / well	0.5 $\mu$ L / well
96-well plate	0.1 ~ 0.2 $\mu$ g IgG / well	0.1 $\mu$ L / well

- \* Optimization of incubation time is needed for each antibody and cell line.

## Practical example

Seed HeLa cells in a 12-well plate.

( $1.0 \times 10^5$  cells / well; culture volume : MEM (+10% FBS) 1 mL / well)

↓ incubate at 37 °C for 24 hrs at 5% CO<sub>2</sub>

Add Ab-Carrier 1μL to FITC-labeled IgG (0.1 mg / mL) 20μL and mix.

↓ leave to stand 20 min at room temp.

Add the reaction mixture into 24-hrs culture of HeLa cell (21μL / well).

↓ incubate at 37 °C for 24 hrs at 5% CO<sub>2</sub>

Remove the medium and wash twice with PBS (1 mL / well) .

↓

Add 0.25% Trypsin-EDTA (100 μL / well) .

↓ incubate at 37 °C for 2 min at 5% CO<sub>2</sub>

Add 1 mL of MEM (+10% FBS) to terminate reaction.

↓

FACS

### Antibody transfection activity (estimated by FACS)

Upper: Percentage of transfected cells (%)

Lower: Amount of transfected antibody per cell (relative fluorescent intensity)

Goat polyclonal IgG	93.2%
	14.0
Rabbit polyclonal IgG	92.6%
	17.9
Mouse monoclonal IgG1	81.1%
	3.6
Mouse monoclonal IgG2a	78.1%
	4.8

## Attention

### • Preservatives such as NaN<sub>3</sub>

Antibody including preservatives such as NaN<sub>3</sub> may cause inhibition of antibody transfection activity because of their cytotoxicity. It is recommended to remove them by desalting or buffer exchanging.

### • After cryopreservation

After thawing, Ab-Carrier should be treated by ultrasonic for about 10 sec.

## Order Information

Product Name	Contents	Code No.
Ab-Carrier	12.5 $\mu$ L $\times$ 1 ( 25 reactions* )	PTN-P-101-25
	12.5 $\mu$ L $\times$ 4 ( 100 reactions* )	PTN-P-101-100

\* (24 well plate ; 1  $\mu$ g IgG / well)

## Related Products ( columns and resins for purification of antibody )

Product Name	Contents	Code No.
Ab-Capcher	2 mL	PTN- P-002-2
	10 mL	PTN- P-002-10
Ab-Rapid PuRe (prepacked column)	2 columns	PTN- P-012-2
Ab-Rapid SPiN (Spin column)	0.1 mL gel/column x 10 ( including 2 mL empty tube x 20)	PTN- P-013-10
Ab-Capcher ExTra	2 mL	PTN- P-003-2
	10 mL	PTN- P-003-10

*For research use only.*



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