

iN-fect™ *in vitro* Transfection Reagent

Cat. No. 15081 500µl

Description

iN-fect™ *in vitro* Transfection Reagent is a polymer-based transfection reagent. iN-fect™ *in vitro* Transfection Reagent is proprietary formulation for the transfection of DNA and RNA into eukaryotic cells. iN-fect™ *in vitro* Transfection Reagent is suitable for many cell types and providing the highly efficiency and low cellular-toxicity.

STORAGE

iN-fect™ *in vitro* Transfection Reagent is stable for 12 months at 4 °C.

Transient transfection of adherent cells

For optimal transfection conditions with iN-fect™ *in vitro* Transfection Reagent, the cells should be 90-95% confluent. Typically, for transfection in 24-well plates, 5×10^4 to 1×10^5 cells are seeded per well, 24 hours before transfection. For other culture formats, see Table 1.

Table 1. Number of cells to seed before the day of transfection and transfection mix preparation for different cell culture formats

Culture Vessel	Number of Cells to seed	Volume of Medium per vessel (ml)	Amount of Plasmid DNA (µg)	Volume of Transfection Reagent (µl)	Total vol. of DNA/reagent complex (µl)
1well/96well	$1 \times 10^4 - 1.7 \times 10^4$	0.1	0.05 - 0.2	0.15 - 0.4	10
1well/24well	$5 \times 10^4 - 1 \times 10^5$	0.5	0.2 - 1.0	0.6 - 2.0	30
1well/6well	$2 \times 10^5 - 4 \times 10^5$	2.0	1.0 - 3.0	3.0 - 9.0	120
35mm	$2 \times 10^5 - 4 \times 10^5$	2.0	1.0 - 3.0	3.0 - 9.0	120
60mm	$4 \times 10^5 - 8 \times 10^5$	5.0	3.0 - 5.0	6.0 - 15.0	300
10cm	$1 \times 10^6 - 6 \times 10^6$	10.0	5.0 - 10.0	15.0 - 30.0	600

Table 2. A guideline for seeding suspension cells prior to transfection in different culture formats recommended

Culture Vessel	Number of Cells to seed	Volume of Medium per vessel (ml)	Amount of Plasmid DNA (µg)	Volume of Transfection Reagent (µl)	Total vol. of DNA/reagent complex (µl)
1well/96well	$2 \times 10^4 - 5 \times 10^5$	0.05 - 0.2	0.15 - 0.4	0.05 - 0.2	10
1well/24well	$1 \times 10^5 - 2 \times 10^5$	0.2 - 1.0	0.6 - 2.0	0.2 - 1.0	30
1well/6well	$2 \times 10^5 - 5 \times 10^5$	1.0 - 3.0	3.0 - 9.0	1.0 - 3.0	120
35mm	$5 \times 10^5 - 2 \times 10^6$	1.0 - 3.0	3.0 - 9.0	1.0 - 3.0	120
60mm	$2 \times 10^6 - 5 \times 10^6$	3.0 - 5.0	6.0 - 15.0	3.0 - 5.0	300
10cm	$5 \times 10^6 - 1 \times 10^7$	5.0 - 10.0	15.0 - 30.0	5.0 - 10.0	600

Protocol for transfection

The following protocol is given for transfection in 24-well plates. Use 1 µl of iN-fect™ *in vitro* Transfection Reagent and 1 µg of DNA per well as follows. See table 1 or 2 for other culture vessel formats.

- Preparation of iN-fect™ working transfection reagent : Dilute 1 µl iN-fect™ *in vitro* Transfection Reagent with 30 µl serum-free medium (without antibiotics), and then mix by gentle pipetting or vortexing for one second. Incubate the working transfection reagent for 5 minutes at room temperature.
Note : avoiding the iN-fect™ *in vitro* Transfection Reagent to contract the plastic tube surface.
- Add the 1 µg plasmid DNA into the iN-fect™ working transfection reagent and mix by gentle pipetting.
- Incubate the mixture of DNA and iN-fect™ *in vitro* Transfection Reagent solution at room temperature for 15 minutes. For some cell line the incubation may be up to 30 minutes.
- Add DNA/ iN-fect™ *in vitro* Transfection Reagent complex to cells in each well and mix by gentle shaking plate, incubate cells at 37 °C in CO₂ incubator for 18-48 hrs.



■ Transfection with siRNA

The following protocol is given for transfection in 24-well plates.

1. Preparation of iN-fect™ working transfection reagent :
Dilute 1 ul iN-fect™ *in vitro* Transfection Reagent with 30 ul serum-free medium (without antibiotics), and then mix by gentle pipetting or vortexing for one second. Incubate the working transfection reagent for 5 minutes at room temperature.
2. Add 100 pmol siRNA into the iN-fect™ working transfection reagent and mix by gentle pipetting.
3. Incubate the mixture of siRNA and iN-fect™ *in vitro* Transfection Reagent solution at room temperature for 15 minutes.
4. Add siRNA/ iN-fect™ *in vitro* Transfection Reagent complex to cells in each well and mix by gentle shaking plate, incubate cells at 37 °C in CO₂ incubator for 27-72 hrs.

Table 3. Cell lines transfected with iN-fect™ *in vitro* Transfection Reagent

293T	C-6	H-4-II-E	HepG2	Jurket	MRC-5	RIE1	SW480
A549	Calu-3	H9C2	HL-60	K562	NCI-H1299	RKO	T274
AGS	Chang	HaCAT	hMSC	L929	NCI-H596	ROS	TCCSUP
B16-F0	CHO	HCT 166	HOS	LoVo	NIH-3T3	SaO52	U251
Bend 3	Cos-1	HCT-8	HT-1080	MC-3T3_E1	P388D1	SH-SY5Y	U2OS
BHK	Cos-7	HEC-1A	HT-29	MCF-10A _{ras}	PC-12	SiHa	U373
BV-2	CV-1	HEK-293	Huh-7	MCF-7	PC-3	SK-BR3	U87-MG
C2C12	DG44	HeLa	HUVEC	MDCK	PT-697(48hl)	SK-OV-3	UMUC3
C3A	EJ	Hep3B	IMCD	MeWo	Raw264.7	SNJ16	Vero
C3H/10T1/2	ES	HePa1c1c	J82	MG-63	RBL-2H3	SNJ-620	W38VA13

TECHNICAL DATA

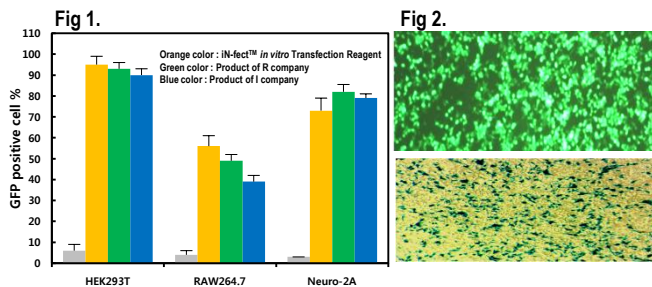


Fig 1. Comparison iN-fect™ *in vitro* Transfection Reagent with other brands.

Fig 2. Example shows exceptional efficiency of iN-fect™ *in vitro* Transfection Reagent on 293T cells.

RELATED PRODUCTS

Additional materials listed are available from iNTRON. More information is available at <http://www.intronbio.com>

Product Name	Cat. No.
DNA-spin™ Plasmid DNA Purification Kit	17096/17097/17098
DNA-midi™ SV Plasmid DNA Purification Kit	17252
DNA-maxi™ SV Plasmid DNA Purification Kit	17253
DNA-midi™ GT Plasmid DNA Purification Kit	17254
β-Gal Stainer™	21032

