

Code No. 293-77101 (0.2 mL)
299-77103 (1 mL)

For Genetic Research

ScreenFect™A plus

ScreenFect™A plus is a transfection reagent consisting of a new cationic liposome screened^{※1} by click chemistry. It can be used with various eukaryote-derived cells and can be added directly to mediums containing antibiotics or serum. DNA and siRNA can be transfected into general experimental cell strains (HeLa, HepG2, MDCK, COS-7, etc.), stem cells (mouse ES cells, etc.), blood cells (macrophages, THP-1, RAW264.7, etc.), microglia, and primary (initial subculture) cells.

In addition, medium replacement after transfection is not required due to low cytotoxicity. And the constituent reagents do not contain any poisonous or deleterious substance.

The optimization range (DNA : reagent ratio) of lipoplex was more flexible than ScreenFect™A, thus ScreenFect™A plus is improved in point of transfection efficiency compared with ScreenFect™A.

※1 *Biomaterials*. 2012 Nov; 33(32):8160-6. 2012

[Kit contents]

Code No.	Size	ScreenFect™A plus Transfection Reagent	Dilution Buffer for ScreenFect™A plus
293-77101	0.2mL	0.2mL	10mL
299-77103	1mL	1mL	50mL

[Storage]

Keep at 2-10 °C (do not freeze).

[Important Guidelines]

• We recommend easy “One-Step” reverse cell transfection method. The one-step method saves a lot of time. However, there are cases where certain cell types or certain applications may require transfection of adherent cell monolayers. In that cases, please choose “Two-Step” transfection protocol and try medium replacement before adding transfection complexes to the adherent cells.

• Transfection performance strongly depends on ScreenFect™A plus reagent : DNA ratio, amount of DNA and siRNA, types of cell lines, etc. Thus, we highly recommend optimal transfection. For additional information and protocols on transfection, please visit Wako’s web site.
(<http://screenfect.jp>)



[Transfection Amounts]

DNA Transfection

DNA transfection (/well)					
Plate size	Surface area	Medium volume	Total volume SF-DNA complex	DNA /Dilution Buffer	ScreenFect™A plus /Dilution Buffer
96 wells	0.3cm ²	80 μ L	20 μ L	~100ng / 10 μ L	~0.4 μ L / 10 μ L
24 wells	2cm ²	350 μ L	80 μ L	~500ng / 40 μ L	~2.0 μ L / 40 μ L
12 wells	4cm ²	700 μ L	140 μ L	~1000ng / 70 μ L	~4.0 μ L / 70 μ L
6 wells	10cm ²	1250 μ L	240 μ L	~2500ng / 120 μ L	~10 μ L / 120 μ L

siRNA Transfection

siRNA transfection (/well)					
Plate size	Surface area	Medium volume	Total volume SF-DNA complex	siRNA /Dilution Buffer	ScreenFect™A plus /Dilution Buffer
96 wells	0.3cm ²	80 μ L	20 μ L	2-3pmol / 10 μ L	0.1-0.3 μ L / 10 μ L
24 wells	2cm ²	350 μ L	80 μ L	10-20pmol / 40 μ L	0.5-1.5 μ L / 40 μ L
12 wells	4cm ²	700 μ L	140 μ L	20-40pmol / 70 μ L	1.0-3 μ L / 70 μ L
6 wells	10cm ²	1250 μ L	240 μ L	20-60pmol / 120 μ L	3.5-7.5 μ L / 120 μ L

Note: Although ScreenFect™A plus work well for siRNA transfection, for optimal results we highly recommend the use of our specialized reagent ScreenFect™siRNA. Visit our homepage to order a free sample. (<http://screenfect.jp>)

Note: For large scale transfections it helps to split sample between several tubes.

E. g. for 10 cm dish transfection, take 5 – 6 samples of the 6-well transfection volumes.

[Protocol]

The one-step protocol for ScreenFect™A plus transfection is described in the next page.

The optimal DNA-to-reagent-ratio for efficient cell transfection stays constant at around 1 : 3 to 1 : 4 (DNA 100ng : ScreenFect™A plus reagent 0.1 μ L = 1 : 1) and we recommend splitting cells when they reach 70%-90% confluence to avoid contact inhibition of cell proliferation.

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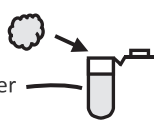
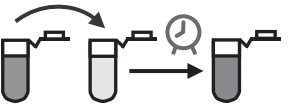
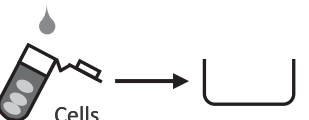
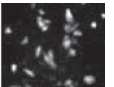
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ScreenFect™A plus One-Step Protocol

ScreenFect™A plus Transfection Reagents Protocol

Transfect cells according to the following table. Each reaction mix is given per well and accounts for pipetting variations.

Timeline		Steps
1	Day 0	 Cultured cells
2		 Suspended cells
3	Day 1	 Reagent Dilution Buffer Plasmid DNA Dilution Buffer
4		 Add Diluted DNA to each tube of diluted ScreenFect™A plus Reagent and incubate for 5 (~20) minutes
5		 Cells Add DNA-lipid complex to the cell suspension and transfer to wells
6	Day 2	 Visualize/analyze transfected cells

Procedure Details						
Component	96-well		24-well		6-well	
Adherent cells or suspension cells	1.0-4.0 × 10 ⁴		0.5-2.0 × 10 ⁵		0.25-1.0 × 10 ⁶	
Detach cells by trypsin or Accutase® for transfection.						
Dilution Buffer for ScreenFect™A plus	10 μ L		40 μ L		120 μ L	
DNA : Transfection Reagent ratio	1 : 3	1 : 4	1 : 3	1 : 4	1 : 3	1 : 4
ScreenFect™A plus Transfection Reagent	0.3 μ L	0.4 μ L	1.5 μ L	2 μ L	7.5 μ L	10 μ L
Dilution Buffer for ScreenFect™A plus	10 μ L		40 μ L		120 μ L	
DNA (0.1-2.5 μ g / μ L)	100 ng		500 ng		2500 ng	
Diluted DNA	10 μ L		40 μ L		120 μ L	
Diluted ScreenFect™A plus Transfection Reagent	10 μ L		40 μ L		120 μ L	
Component [per well]	96-well		24-well		6-well	
DNA-lipid complex	20 μ L		80 μ L		240 μ L	
DNA amount	100 ng		500 ng		2500 ng	
ScreenFect™A plus Transfection Reagent used	0.3 and 0.4 μ L		1.5 and 2.0 μ L		7.5 and 10 μ L	
Incubate cells for 1-4 days at 37°C. Then, analyze transfected cells.						