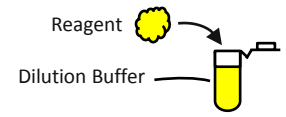
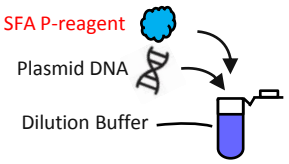
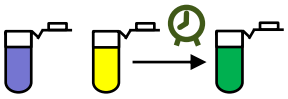
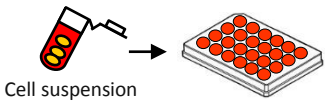
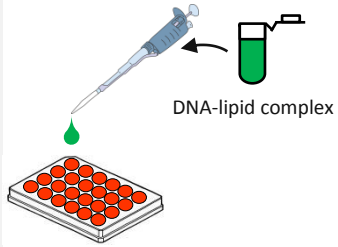
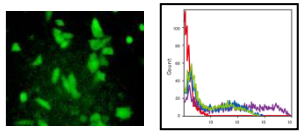
ScreenFect™A *plus* (+SFA P-reagent) Quick Protocol

ScreenFect™A *plus* Transfection Protocol

The optimal condition for successful transfection varies. Start any new transfection by testing the recommended two concentrations of ScreenFect™A *plus* Reagent to determine an optimum amount.

1-Step method (Reverse transfection method)

Timeline	Steps	Procedure Details
1		**Dilute ScreenFect™A *plus* Reagent※1 in Dilution Buffer, and then mix well** **※1 Vortex the reagent before use**
1		**Dilute DNA and SFA P-reagent in Dilution Buffer, and then mix well**
2		**Add diluted DNA (+SFA P-reagent) to diluted ScreenFect™A *plus* Reagent, and then incubate for 5 minutes ～ at room temperature※2** **※2 Incubation is available until the step 4 has been completed**
3		**Prepare required cells for transfection**
4		**Detach cells and prepare the cell suspension, and then transfer the required numbers of cell suspension to cell culture plate**
5		**Add DNA-lipid complex from step 2 to well of cell culture plate from step 4**
6		**Visualize/analyze transfected cells**

Day 0

Day 1～

Procedure Details

Component	96-well		24-well		12-well		6-well	
Dilution Buffer for ScreenFect™A *plus*	5 µL		25 µL		50 µL		125 µL	
DNA : Transfection Reagent ratio	1 : 3	1 : 4	1 : 3	1 : 4	1 : 3	1 : 4	1 : 3	1 : 4
ScreenFect™A *plus* Transfection Reagent	0.15 µL	0.2 µL	0.75 µL	1.0 µL	1.5 µL	2.0 µL	3.75 µL	5.0 µL
Dilution Buffer for ScreenFect™A *plus*	5 µL		25 µL		50 µL		125 µL	
DNA (0.1-2.5 µg / µL)	50 ng		250 ng		500 ng		1250 ng	
SFA P-reagent (2µL / µg DNA)	0.1 µL		0.5 µL		1.0 µL		2.5 µL	
Diluted DNA (+SFA P-reagent)	5 µL		25 µL		50 µL		125 µL	
Diluted ScreenFect™A *plus* Transfection Reagent	5 µL		25 µL		50 µL		125 µL	
Adherent cells or suspension cells	1.0-4.0 × 10^4		0.5-2.0 × 10^5		1.0-4.0 × 10^5		0.25-1.0 × 10^6	
Cell Detachment (Trypsin or *Accutase*®)								

Final composition〔per well〕	96-well	24-well	12-well	6-well
DNA-lipid complex	10 µL	50 µL	100 µL	250 µL
DNA amount	50 ng	250 ng	500 ng	1250 ng
SFA P-reagent used	0.1 µL	0.5 µL	1.0 µL	2.5 µL
ScreenFect™A *plus* Transfection Reagent used	0.15 or 0.2 µL	0.75 or 1.0 µL	1.5 or 2.0 µL	3.75 or 5.0 µL
Medium volume	100 µL	500 µL	1000 µL	2000 µL

Incubate cells for 1 day ～ at 37℃. Then, analyze transfected cells.

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ScreenFect™A *plus* Transfection Protocol

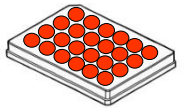
The optimal condition for successful transfection varies. Start any new transfection by testing the recommended two concentrations of ScreenFect™A *plus* Reagent to determine an optimum amount.

2-Step method (Forward transfection method)

Timeline

Day 0

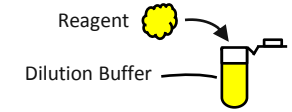
1

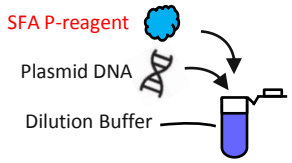


Pre-Cultured cells

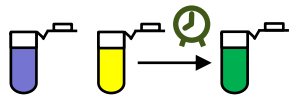
Day 1

2



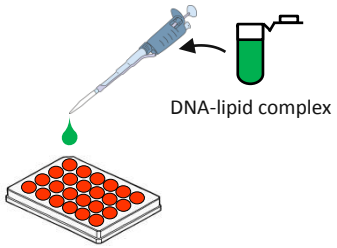


3



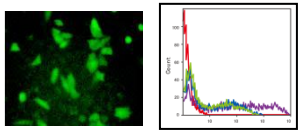
DNA-lipid complex

4



Day 2～

5



Steps

Seed cells to be 70-90% confluent at transfection

Dilute ScreenFect™A *plus* Reagent※1 in Dilution Buffer, and then mix well
※1 Vortex the reagent before use

Dilute DNA and SFA P-reagent in Dilution Buffer, and then mix well

Add diluted DNA (+SFA P-reagent) to diluted ScreenFect™A *plus* Reagent, and then incubate for 5 minutes ～ at room temperature※2
※2 Incubation is available until the step 4 has been completed

Add DNA-lipid complex from step 3 to well of cell culture plate from step 1

Visualize/analyze transfected cells

Procedure Details

Component	96-well	24-well	12-well	6-well
Adherent cells or suspension cells	1.0-4.0 × 10^4	0.5-2.0 × 10^5	1.0-4.0 × 10^5	0.25-1.0 × 10^6

Seed cells to be 70-90% confluent at transfection. The medium replacement may improve a transfection efficiency in a 2-Step method.

Dilution Buffer for ScreenFect™A *plus*	5 µL		25 µL		50 µL		125 µL	
DNA : Transfection Reagent ratio	1 : 3	1 : 4	1 : 3	1 : 4	1 : 3	1 : 4	1 : 3	1 : 4
ScreenFect™A *plus* Transfection Reagent	0.15 µL	0.2 µL	0.75 µL	1.0 µL	1.5 µL	2.0 µL	3.75 µL	5.0 µL
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Diluted DNA (+SFA P-reagent)	5 µL		25 µL		50 µL		125 µL	
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Final composition〔per well〕	96-well	24-well	12-well	6-well
DNA-lipid complex	10 µL	50 µL	100 µL	250 µL
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