Gxpress 293 Transfection & Medium Kit II

for 293 Cells “Gene Expression System”

Description

Gxpress 293 Transfection & Medium Kit II has been developed for the gene expression system of human embryonic kidney (HEK) 293 cells, such as Expi293F or 293T cells, and the high production of recombinant proteins or the transfection assay in serum-free culture. All contents of Gxpress 293 Transfection & Medium Kit II are a chemically defined composition that no contains serum, protein, animal origin, hydrolysates, and no unknown composition. (Storage; 2℃ to 8℃ / Protect from light)

Package Contents (Catalog Number GX293-MK-0010)

● Gxpress 293 Transfection Kit*	(Catalog Number GX293-RK-0010)
*Gxpress 293 TF Reagent	2.8 mL (1.4 mL x 2)
*293 Complexing Solution	100 mL (50 mL x 2)
*Gxpress 293 Enhancer	60 mL
● HE400 medium (1,000 mL)	(Catalog Number HE400-0010)

Important Points

- Subculture 293 cells a minimum of three times to allow them to recover from thawing before use.
- Plasmid DNA must be clean.
- Gently mix Transfection Reagent and DNA into 293 Complexing Solution.
- Use 293 Complexing Solution in transfection assay.

Culture Conditions

- Cell line: 293 cells
- Culture type: Suspension
- Culture vessels: Flask or culture bag, etc.
- Incubate atmosphere: Humidified atmosphere of 5–8% CO_2 in air
- Temperature range: 36℃ to 38℃
- Shaker culture: 120–130 rpm

Required Materials

- Plasmid DNA
- Erlenmeyer shaker flask, vent-cap
- Expi293F or 293T cells
- CO_2 Incubator and Shaker

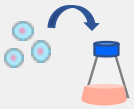
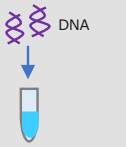
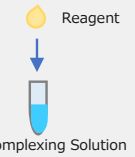
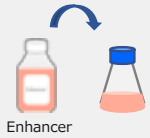
Gxpress 293 TF Reagent

Gxpress 293 TF Reagent is a cationic lipid-based reagent for transferring DNA into mammalian cells. Gently mix Reagent by pipetting it up and down before use. Use 293 Complexing Solution in transfection assay. Gently mix Reagent and DNA into 293 Complexing Solution.

HE400 medium

HE400 medium has been developed for the growth of 293 cells and the high production of recombinant proteins or the transfection assay in serum-free culture. Medium contains no L-alanyl-L-glutamine and requires supplementation with L-glutamine or L-alanyl-L-glutamine before use. Medium contains no antibiotics. Please supply to the medium as necessary.

Gxpress 293 TF Reagent Transfection Protocol (30 mL Scale)

Schedule		Steps	Procedures
Day -1 (Preculture)	1 	Seeding of 293 cells	Transfer 9 x 10^7 viable cells into 125-mL flask containing 30 mL of HE400 medium. (a seeding density of 3 × 10^6 cells/mL)
	2 	Shaker culture	Temperature : 37℃, Shaker speed : 125 rpm, Atmosphere : 5−8% CO_2 in air.
Day 0 (Transfection)	3 	Preparation of cells	Determine viable cell density. The density should be 3−5 x 10^6 cells/mL (> 95% viability). Add 7.5 x 10^7 cells into 125-mL flask containing 25 mL of HE400AZ medium. (a seeding density of 3 × 10^6 cells/mL)
	4 	Prepare DNA mixtures	Dilute 30 µg of plasmid DNA in 1.5 mL of 293 Complexing Solution. Mix gently by inversion one time.
	5 	Prepare Gxpress 293 TF Reagent mixtures	Dilute 84 µL of Gxpress 293 TF Reagent in 1.5 mL of 293 Complexing Solution. Mix gently by inversion one time. Incubate for 5 min at room temperature.
	6 	Prepare DNA/Reagent complexes	After 5 min incubation, add diluted Gxpress 293 TF Reagent to diluted DNA. **Mix gently by inversion one time.** Incubate for 5 min at room temperature. Note; Do not mix vigorously.
	7 	Add DNA/Reagent to 293 cells	After 5 min incubation, transfer gently the complexes to 293 cells. Note; Longer incubation times may result in decrease expression.
	8 	Shaker culture	Temperature : 37℃, Shaker speed : 125 rpm, Atmosphere : 5−8% CO_2 in air.
Day 1 (Enhancer)	9 	Add Gxpress 293 Enhancer to 293 cells	After 18 − 24 h transfection, add 1.5 mL of Gxpress 293 Enhancer to 293 cells.
	10 	Shaker culture	Temperature : 37℃, Shaker speed : 125 rpm, Atmosphere : 5−8% CO_2 in air.
Day 2 − 7 (Expression)	11 	Harvest cells or media	Evaluate recombinant protein expression. Culture time for optimal protein expression depends on the nature of recombinant protein.