

## Turbo293™ Transfection Reagent

SKU/Catalog#	Size
PXX1001	1 mL
PXX1002	20 mL
PXX1003	50 mL

### Description

Turbo293™ Transfection Reagent has a unique polycationic polymer formulation designed for transfection of HEK293 cells grown in suspension culture. It is ideal for mammalian protein production through transient transfection method. Turbo293™ Transfection Reagent is derived from non-animal sources and gives minimal cellular toxicity. It is provided as a sterile, ready-to-use solution. Benefits of this product include:

- Optimized for transient transfections of HEK293 suspension cultures
- Minimal cellular toxicity
- Derived from non-animal sources
- Protocol easily scales up for production
- Compatible with both serum-containing and serum-free media
- Eliminates the need for media changes

### General Considerations

- Use only high quality, endotoxin-free DNA. DNA should be at a concentration of 1-2 µg/µL. If DNA concentration is lower, concentrate the DNA to reach optimal concentration.
- Propagate HEK293 suspension cultures on an orbital shaker at 125 rpm at 37°C (8% CO2).
- Passage cells regularly (e.g., every 2-3 days). Avoid density higher than 4 X 10<sup>6</sup>/mL. Use only rapidly proliferating cells for transfection (e.g., doubles in 24 hours). To ensure reproducibility, keep cell growth conditions and density consistent.
- Make sure that the cells are healthy and greater than 90% viable before proceeding to transfection.
- Turbo293™ Transfection Reagent is compatible with both serum-containing and serum-free media.

Note: Do not include serum and antibiotics during the formation of the transfection reagent/DNA complex.

### Protocol

The following protocol is optimized for transfection of 25 mL HEK293 cells in suspension in a 125-ml

polycarbonate Erlenmeyer flask. However, the amounts of reagent and DNA can be varied if necessary. For other culture sizes, reagent and DNA amounts can be scaled up or down proportionally (a guideline is provided in Table 1 below).

1. The day before transfection, passage HEK293 suspension cells at approximately  $1.0 - 1.5 \times 10^6$  cells/ml. Incubate at  $37^{\circ}\text{C}$  (8% CO<sub>2</sub>) overnight with 125 rpm rotation. Cell density should be at approximately  $2.5 \times 10^6$  cells/ml at the time of transfection.
2. On the day of transfection, dilute the cells to  $2.5 \times 10^6$  cells/mL and place 25 ml of cells in a 125 mL shake flask.
3. Place 1.25 mL serum-free Opti-MEM or MEM into a sterile tube. Add 25  $\mu\text{g}$  DNA and mix well.  
**Note:** Do not use the serum-free culture medium in which HEK293 cells were grown.
4. Place 1.25 mL serum-free Opti-MEM or MEM into a separate sterile tube. Add 75  $\mu\text{L}$  of Turbo293™ Transfection Reagent and mix well.
5. Combine the diluted Turbo293™ Transfection Reagent from step 4 with the diluted DNA tube from step 3 and mix well.
6. Incubate transfection mixture at room temperature for 15 minutes to allow formation of Turbo293™ Transfection Reagent and DNA complex.
7. Add entire volume of transfection mixture drop wise to the prepared HEK293 cell suspension.
8. Incubate cultures for 3–5 days with 125 rpm rotation at  $37^{\circ}\text{C}$  (8% CO<sub>2</sub>).
9. Harvest cells for protein purification, characterization, or reporter assays

Table 1. Preparation of Transfection Mixture for Various Culture Sizes

	Culture Size			
Transfection Culture Volume (mL)	25	200	450	900
Number of cells ( $\times 10^6$ )	62.5	500	1125	2250
Volume of MEM or PBS in the transfection mixture (mL)	1.25	12.5	25	50
Volume of Turbo293™ Transfection Reagent ( $\mu\text{L}$ )	75	750	1500	3000
Amount of plasmid DNA ( $\mu\text{g}$ )	25	250	500	1000