



FuGENE HD

Quick Protocol

Preparing the FuGENE HD® Transfection Reagent

1. Before use, allow the vial of FuGENE® HD Transfection Reagent to reach room temperature
2. Mix by inverting or vortexing briefly. If a precipitate is visible, briefly warm at 37 degrees C then cool to room temperature

General Transfection Protocol

1. To a sterile tube or U- or V-bottom plate add room temperature medium to so that the final volume after adding FuGENE HD® & DNA in Step 2 & 3 is 100µl total volume.
2. Add 2µg of plasmid DNA (0.2–1µg/µl) to prewarmed media and vortex.
3. For a 3:1 FuGENE® HD Transfection Reagent:DNA ratio, add 6µl of FuGENE® HD Reagent directly to medium, and mix immediately. For other ratios, consult Table 1.

	Ratios of FuGENE® HD to DNA					
	6:1	4:1	3:1	2.5:1	2:1	1.5:1
Medium to Final Volume	100ul	100ul	100ul	100ul	100ul	100ul
DNA Amount	2ug	2ug	2ug	2ug	2ug	2ug
Volume of FuGENE® HD	12ul	8ul	6ul	5ul	4ul	3ul

4. Incubate the FuGENE® HD Transfection Reagent/DNA mixture for 5-30 minutes at room temperature. (Some cell lines show increased expression with the longer 30 min incubations)
5. Add 2–10µl of the FuGENE® HD Transfection Reagent/DNA mixture per well to a 96-well plate containing 100µl of cells in growth medium. Mix by pipetting or using a plate shaker. Return cells to the incubator for 24–48 hours.
6. Measure transfection efficiency using an assay appropriate for the reporter gene. For transient transfection, cells are typically assayed 24–48 hours after transfection.
7. See additional protocol information in Technical Manual available on www.fugene.com
8. For additional support please contact us at www.fugene.com