See the Unseen FuGENE®

FuGENE HD Quick Protocol

Preparing the FuGENE HD® Transfection Reagent

1. Seed cells to be 50-90% confluent at time of transfection
2. Before use, allow the vial of FuGENE® HD Transfection Reagent to reach room temperature
3. 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 (transfection mix enough to transfect one 35mm dish)

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.

Table 1: Volumes of FuGENE HD for Various FuGENE HD: DNA Ratios

	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	8ul	7ul	6ul	5ul	4ul	3ul

4. Incubate the FuGENE® HD Transfection Reagent/DNA mixture for 5-15 minutes at room temperature.
5. Add transfection Reagent/DNA mixture to 35mm dish containing 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