THE WORLD'S MOST AFFORDABLE TRANSFECTION REAGENT

Van Heron Labs has created a versatile transfection reagent to enable nucleic acid delivery at nominal cost.

Our novel methodology for preparation has resulted in an affordable reagent capable of tackling a range of challenges.

Though the product has been tested for plasmid DNA transfection into model mammalian cell lines, we hope that our pricing opens up opportunities for our partners to explore new use cases anywhere where cellular delivery is needed, such as for primary, stem cell lines and other hard-to-transfect systems, among even more exotic challenges and diverse nucleic acids or other cellular payloads.

This document outlines best practices and optimization guidelines for mammalian transfection based on plasmid DNA for suspension and adherent cultures, but we welcome all explorations for testing our transfection reagent in your unique system or cell line. Our reagent should generally be compatible with nearly any bio-system or protocol due to it's composition and extremely low cytotoxicity across cell types.

For best results for each experiment, significant optimization and replication should be deployed in order to determine optimal conditions.

INTRODUCTION: MAMMALIAN PLASMID TRANSFECTION

First, ensure you have high-purity endotoxin free DNA by confirming purity with nanodrop (or similar). If low quality, perform additional purification, as needed.

Importantly, FOR BEST RESULTS a range of reagent to DNA ratios should be assessed. We suggest a range of 0.5 - 12uL reagent per 1 ug DNA. If necessary, 3 - 4uL could be used to start.

PROCEDURE:

For 1 well of a 12-well plate:

- Plating complete media volume - 1mL
- Serum and antibiotic free media - 100uL
- Reagent - 0.5 - 12uL

Recipe/ratios scalable to T75, 96 well + others.

1) Cells should generally be at high density and actively proliferating for the transfection procedure (~ 60 - 85% confluence). For wells of a 12 well cell culture plate, plate the cells 18 - 36 hours beforehand in 1mL volume prior to the transfection protocol. For suspension cultures plate ~ 200 - 500K cells/mL and for adherent cultures plate ~ 50 - 300K cells/mL. To test a range of DNA:reagent ratios, multiple well-plates may be needed.

2) After incubation, prepare for the transfection. First warm the reagent and target DNA to room temperature.
3) Label several tubes for DNA:reagent mastermixes to test a range of DNA:reagent ratios (0.5 - 12uL reagent to 1 ug DNA)
4) Add 100 uL of serum and antibiotic free media to each of the tubes. Chemistry will need to be multiplied for any mastermixes created for replicate wells.
5) Pipet DNA stock to mix and to each tube add 1 ug (1 uL of a 1ug/uL stock) of the plasmid DNA
6) Add the reagent to each tube in the various concentrations (0.5uL - 12uL)
7) Pipet several times to mix completely. This step is key. Try not to introduce bubbles.
8) Incubate at 30 min at room temperature.
9) Pipet gently to mix once more before adding the mastermix to the culture.
10) Use a transfer pipet to add the mastermixes dropwise to the culture wells. It is important to add each drop to a different area of the well each time and ensure any replicate wells receive the same number of drops.
12) Gently rock the culture back-and-forth and side-to-side to distribute.
13) Incubate for 1 - 3 days. 24 - 48 hours is optimal.
14) Perform desired assays and experiments.
14) If creating stable transfections, transfer cells to selection media as needed.

The reagent may be used and stored up to 1 year. Keep refrigerated and away from light.