

**Lipofectamine 2000 Transfections:
MKN45 and AGS cells**

24 well plate of cells. Approximately 50-60% confluent on the day of transfection. One confluent T-25 split into 24 wells give about the right density. Alternatively, you can plate 100-200,000 cells/well.

DNA: 600-800ng total per well

Lipofectamine 2000: 1.5 μ l per well

Needed for 3 wells:

DNA: 1800-2400 ng in 150 μ l serum-free media (SFM)

Lipid: 4.5 μ l in 150 μ l SFM

1. Separately make up DNA and lipid mixtures; incubate 5 minutes at RT
2. Mix 150 μ l DNA + 150 μ l diluted lipid; Incubate 20 minutes at RT
3. Add 100 μ l DNA/lipid mixture per well
4. Incubate overnight.
5. Change media the next day.
6. Stimulate on Day 3.