Welcome to OriGene!

Thank you for choosing OriGene Synthetic RNA for your CRISPR experiment! You have purchased world-class guide RNA that offers an unbeatable combination of quality, speed, accuracy, scalability and price.

Step 1: Dissolve & Dilute Your sgRNA

Wear Gloves !

OriGene synthetic sgRNA oligos are lyophilized prior to shipping. They are stable in this format for several weeks at room temperature, but it is best practice to store any undissolved RNA oligos at -20 °C.

Be sure to work in an RNase-free environment.

Before you start: We recommend that users dissolve OriGene RNA to concentrations based upon the labelled amount. The value printed on the tube or plate represents the dry yield of full-length product present in the tube. OriGene combines data obtained via IP-HPLC-UV and IP-UPLC-ESI-MS to accurately quantitate this amount

1. Briefly centrifuge your tubes or plates containing single guide RNA (sgRNA) oligos to ensure that the dried RNA pellet is collected at the bottom.
2. Carefully dissolve RNA in the provided nuclease-free 1X TE Buffer (Tris-EDTA, pH8.0) to an appropriate stocl concentration.
   Recommended protocol: For sgRNA (1nmol), add 20µl of nuclease-free 1x TE buffer for a final concentration of 50µM (50pmol/µl).
3. Dissolved RNA should be stored at -20 °C. Under these conditions, RNA will be stable for at least one year.

Step 2: Dilute Your sgRNA

1. To make a working stock, add 10µl of 50µM sgRNA oligo to 40µl of the provided nuclease-free water to make a total of 50µl of 10µM (10pmol/µl) sgRNA. This will be your working stock.
2. Use diluted sgRNA immediately or store at -20 °C for up to three months.

Step 3: You are Now Ready to Use Your OriGene sgRNA!

OriGene recommends forming ribonucleoprotein (RNP) complexes for your genome editing experiments in order to maximize editing efficiency and reduce off-target effects. To form RNP complexes, proceed to Step 4.

Step 4: Form an RNP Complex with Cas9 (Optional)

If you are using lipofectamine-based transfection, electroporation or microinjection, RNP formation with Cas9 nuclease is recommended.

Be sure to use the appropriate Cas9 (e.g., Cas9 wild-type, Cas9-NLS etc.) for your cell type or application.

1. OriGene Cas9 (Cat#TP790148) has a concentration of 20µM (20pmol/µl) and requires no further dilution. Cas9 nuclease from other vendors should be diluted to 20µM (20pmol/µl) in a suitable buffer.
2. Form Cas9:sgRNA RNP Complexes: For each transfection/electroporation/microinjection, add 2µl (20pmol) of sgRNA and 1µl (20pmol) of Cas9 nuclease to either sterile growth media (lipofectamine), electroporation buffer or microinjection buffer to a final volume of 12.5µl. Note that you may need to experimentally determine the optimum Cas9:sgRNA ratio for your cell type or experiment.
3. Incubate at room temperature for 5-10 minutes to assemble the RNP Complexes.
4. Store RNP Complexes on ice until ready to use, or store at 4° C for up to one week.

Step 5: Transfect, Electroporate or Microinject Cells

For protocols, please visit origene.com/support/protocols

Any questions, please send us an email at techsupport@origene.com