Resuspension of duplexed oligonucleotides (<50 nmol yield*)

A protocol for resuspending dried, annealed oligos, including Dicer-Substrate siRNAs (DsiRNAs)

1. Centrifuge tubes before opening to ensure duplexed oligos are at the bottom of the tube.
2. Resuspend duplexed oligos in Nuclease-Free Water (catalog # 11-04-02-01) to make a stock solution (concentration ≥100 µM). For example:

Duplexed oligo amount	Nuclease-Free Water (100 µM final concentration)
2 nmol	20 µL
10 nmol	100 µL
25 nmol	250 µL
50 nmol	500 µL

3. Make further dilutions (<100 µM) using a buffer containing 100 mM Na^+ or K^+ (e.g., a Nuclease-Free Duplex Buffer [100 mM Potassium Acetate; 30 mM HEPES, pH 7.5, catalog # 11-05-01-03]). For example:

Final concentration	100 µM duplexed oligo (from Step 2)	Buffer (containing 100 mM Na^+ or K^+)
50 µM	20 µL	20 µL
20 µM	20 µL	80 µL
10 µM	10 µL	90 µL

To calculate other dilutions, use the online IDT Dilution Calculator at https://www.idtdna.com/Calc/Dilution/.

Visit www.idtdna.com/protocols to verify that you are using the most current version of this protocol.

* For instructions for resuspending duplexed oligos of ≥50 nmol yield, turn card over. Salts will be present following annealing and dry-down processes at IDT. To maintain suitable salt concentrations for the duplex structure of your product, we recommend these resuspension protocols.

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1. Centrifuge tubes before opening to ensure duplexed oligos are at the bottom of the tube.
2. Resuspend duplexed oligos in Nuclease-Free Water (catalog # 11-04-02-01) to make a stock solution (volume ≤500 µL). For example:

Final concentration	Duplexed oligo amount	Nuclease-Free Water
200 µM	100 nmol	500 µL
500 µM	250 nmol	500 µL

3. Make further dilutions (>500 µL) using a buffer containing 100 mM Na^+ or K^+ (e.g., a Nuclease-Free Duplex Buffer [100 mM Potassium Acetate; 30 mM HEPES, pH 7.5, catalog # 11-05-01-03]). For example:

Final concentration	200 µM duplexed oligo (from Step 2)	Buffer (containing 100 mM Na^+ or K^+)
50 µM	25 µL	75 µL
10 µM	5 µL	95 µL

Final concentration	500 µM duplexed oligo (from Step 2)	Buffer (containing 100 mM Na^+ or K^+)
50 µM	10 µL	90 µL
10 µM	2 µL	98 µL

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