

# TCEP Reaction for Thiol-Modified siRNA/ RNA Oligonucleotides

Thiol-modified siRNA and RNA oligonucleotides with the thiol group in a protected or oxidized form prevent the formation of dimers. Prior to use, the thiol groups on the thiol-modified oligonucleotide can be deprotected or reduced using the following protocol.

## *Materials Required*

- BOND-BREAKER™ TCEP [Tris (2-carboxyethyl) phosphine hydrochloride] solution, neutral pH, 0.5M (Pierce Cat.# 77720)
  - Make 1.0 mL of 3% TCEP solution in RNase-free water
- Molecular Grade Water, RNase-free (HyClone Cat# SH30538.01)
- 9.5 M Ammonium Acetate
- 200 proof, 99.5% Ethyl Alcohol

## *Protocol*

1. Add 400 µL of 3% TCEP solution to dry oligonucleotide
2. Vortex sample until it is in solution
3. Place sample on rocker for 1 hour
4. Add 150 µL of 9.5 M Ammonium Acetate
5. Add 1.5 mL of 200 proof Ethyl Alcohol
6. Place in -80°C freezer for at least 20 minutes
7. Remove sample from freezer and spin in a micro-centrifuge at 13,000 RPM for 15 minutes
8. Carefully pour off supernatant into a 2 mL tube and save
9. Allow sample to dry completely in SpeedVac®
10. Add desired amount of RNase-free water to reconstitute dry sample
11. Quantitate sample by obtaining an absorbance at 260 nm
  - If quantity of sample is acceptable, discard supernatant

## *Contact Information*

For technical questions regarding the use of siRNA reagents, please contact Dharmacon Products Technical Support at:

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