



Product Specifications

Omni-Pure RNA Purification, Electrophoresis Reagents, Oligo Conjugation
Custom Primers and Probes
Hybridization and Detection Reagents

TCEP 0.5M Solution

| | Catalog Number | Description | Size |
|--------------------------|----------------|---------------------------|--------|
| <input type="checkbox"/> | 40-5116-01 | TCEP 0.5M Solution;100 µl | 100 µl |

Storage:

Shipped on ice. Store at -20°C.

Product Description:

TCEP (*tris*(2-carboxyethyl)phosphine) is a potent reducing agent and due to its odorless characteristic is a substitute for DTT and beta- mercaptoethanol use.

Supplied at 0.5M that is 100X concentration. Recommended usage is at a final concentration of 5 mM or Use at 10µL/mL.

Caution: Use RNase free tubes and reagents for further dilution and use.

Applications and Recommended Product Use:

TCEP (*tris*(2-carboxyethyl)phosphine) is a component in Gene Link Omni-Pure™ RNA purification kits (40-4090-XX and 40-4080-XX). It is used a reducing agent to break disulfide bonds in RNase A and thus ensure high yield of RNA extraction by inhibiting RNase A activity.

TCEP is also recommended for reducing thiol labeled siRNA and other oligonucleotides.

Properties

TCEP selectively and quantitatively reduces even the most water soluble resistive alkyl disulfides over a wide pH range. Reductions typically require less than 5 minutes and are conducted at room temperature. TCEP is odorless and unlike other reducing agents stable to air oxidation. Compared to dithiothreitol (DTT) TCEP is more stable, more effective in reducing disulfides.



TCEP does not contain thiols and does not have to be removed from solutions before performing reactions involving maleimide labeling or other cross linking reagents. TCEP concentrations of <10-20 mM are compatible with most maleimide reaction chemistry

Reduction of Thiol Modified oligos

Gene Link synthesized thiol modified oligos (all thiol modified DNA oligos, RNA, siRNA and molecular probes) are shipped lyophilized and as a disulfide protected* form to prevent the formation of dimers. These can be reduced by the end user after reconstitution and prior to use.

The preferred method is to reduce using TCEP [Tris (2-carboxyethyl) phosphine hydrochloride] The oligos can also be reduced by the classical DTT method. Both methods are given below.

TCEP Reduction Protocol

1. Prepare 1.0 mL of ~3% TCEP solution by adding 250 μ L of 0.5M TCEP to 750 μ L of sterile RNase free water. Prepare more as required. Preparing fresh 3% TCEP is recommended.
2. Add 400 μ L of 3% TCEP directly to lyophilized thiolated siRNA. Leave at room temperature (RT) for 1 hr to reduce the thiol groups. Vortex intermittently.
3. Add 50 μ L of 3M Sodium Acetate pH 5.2 and vortex.
4. Add 1.5 mL of absolute ethanol, vortex and store at -80°C for 20 minutes.
5. Centrifuge at 12K rpm for 10 minutes. Decant ethanol and air dry pellet.
6. Dissolve in 200 μ L of sterile RNase free water or choice of buffer volume as required.
7. Determine sample concentration by obtaining an absorbance at 260 nm

DTT Reduction Protocol

1. Prepare a 100mM DTT solution in sodium phosphate buffer, (pH 8.3 – 8.5). DTT is available as a solid from Sigma-Aldrich (Product No. D9779). For 5 mL of the 100 mM solution, add 77.13 mg of the DTT powder
2. Add 400 μ L of 100 mM directly to lyophilized thiolated siRNA. Leave at room temperature (RT) for 1 hr to reduce the thiol groups. Vortex intermittently.
3. Add 50 μ L of 3M Sodium Acetate pH 5.2 and vortex.
4. Add 1.5 mL of absolute ethanol, vortex and store at -80°C for 20 minutes.
5. Centrifuge at 12K rpm for 10 minutes. Decant ethanol and air dry pellet.
6. Dissolve in 200 μ L of sterile RNase free water or choice of buffer volume as required.
7. Determine sample concentration by obtaining an absorbance at 260 nm

*Prior to July 20, 2011 all Gene Link supplied thiolated oligos were supplied in a reduced state.

Handling & Storage of RNA Oligos

Follow established stringent RNase free handling conditions. The lyophilized RNA or siRNA duplex should be stored immediately at -20° C. The lyophilized siRNA is stable for ~6 months at -20° C.

References

1. K. C., Dixon, J.E., *Anal. Biochem*, 161, 524-528 (1987).
2. Gray, W. R., *Protein Science*, 2, 1732-1748, (1993).
3. Fisher, W.H., et al, *Mass Spectrometry*, 7, 225-228, (1993).
4. Oda, Y. et al, *Nature Biotech*, 19, 379-382 (2001).



Ordering Information

| Product | Catalog No. | Size |
|--|-------------|-----------|
| TCEP 0.5M solution ; 100 µL | 40-5116-01 | 100 µL |
| RNase A solution, DNase Free. 2 mg/ml;; 1 mL | 40-5101-10 | 2 mg |
| RNase A solution, DNase Free. 10 mg/ml; 1 mL | 40-5101-01 | 10 mg |
| DNase I, RNase-free; 2u/µl | 40-5111-05 | 500 units |
| Proteinase K; 10 mg/ml | 40-5203-02 | 200 µL |
| Lytic Enzyme; 4000 units/ml | 40-5205-02 | 200 µL |
| Glycogen Solution; 10 mg/ml | 40-5112-02 | 200 µL |

Related Products Ordering Information

| Omni-Pure™ RNA Purification Systems | | |
|--|-------------|-------|
| Product | Catalog No. | Size* |
| Omni-Pure™ Blood & Bodily fluids RNA Spin Column Purification System | 40-4080-05 | 50 |
| Omni-Pure™ Blood & Bodily fluids RNA Spin Column Purification System | 40-4080-10 | 100 |
| Omni-Pure™ Blood & Bodily fluids RNA Spin Column Purification System | 40-4080-50 | 500 |
| Omni-Pure™ Universal RNA Spin Column Purification System | 40-4090-05 | 50 |
| Omni-Pure™ Universal RNA Spin Column Purification System | 40-4090-10 | 100 |
| Omni-Pure™ Universal RNA Spin Column Purification System | 40-4090-50 | 500 |
| Omni-Pure™ Blood & Bodily fluids RNA Purification System | 40-4081-05 | 50 |
| Omni-Pure™ Blood & Bodily fluids RNA Purification System | 40-4081-10 | 100 |
| Omni-Pure™ Blood & Bodily fluids RNA Purification System | 40-4081-50 | 500 |
| Omni-Pure™ Universal RNA Purification System | 40-4091-05 | 50 |
| Omni-Pure™ Universal RNA Purification System | 40-4091-10 | 100 |
| Omni-Pure™ Universal RNA Purification System | 40-4091-50 | 500 |
| Omni-Pure™ Viral RNA Spin Column Purification System | 40-3650-01 | 100 |
| Omni-Pure™ Viral RNA Spin Column Purification System | 40-3650-05 | 500 |

**Unit of size is purification performed. Sample volume for each purification system varies. Each purification yields sufficient quantity for desired applications.

All Gene Link products are for research use only.

