

Certificate of Analysis & Product Specifications

Omni-Pure RNA Purification, Electrophoresis Reagents, Oligo Conjugation

Custom Primers and Probes

Hybridization and Detection Reagents

TCEP 0.5M Solution

Product Label Information

		Catalog	Number	Description		Size
		40-5116	5-01	TCEP 0.5M Solution; 100 μL	•	100 μL
REF		40-5116	-10 TCEP 0.5M Solution; 1 mL			1 mL
		40-5116	5-C0010	TCEP 0.5M Solution; 10 mL	10 mL	
—.		40-5116	5- C0100	TCEP 0.5M Solution; 100 m	L	100 mL
		40-5116	5- C0500	TCEP 0.5M Solution; 500 m	L	500 mL
RUO Research Use Only				-20°C	LOT	
Research Use Only			S	Storage Store at -20°C to -10°C	Lot Number Stated on product tube and packing slip	
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Expiry			Instructions QR Code			
As marked on product			Cor	Consult product description Visit Gene Link website for product description		

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All component reagents have been manufactured using molecular biology grade water and certified to be DNase and RNase Free.

Appropriate nuclease free handling, dispensing and storage conditions required.

Manufacturing lot numbers are stated on the label of each product and accompanying packing slip.

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 $10\mu L/mL$.

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 stale to air oxidation. Compared to dithiothreitol (DTT), TCEP is more stable and 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.

RNA Purification Protocols & RNase Inactivation

TCEP is used as a reducing agent to break disulfide bonds in RNase A and thus ensure high yield of RNA extraction by inhibiting RNase A activity. It is supplied as 100X concentrated for use at 10 μ L/mL to reduce RNase A disulfide bonds. Add 10 μ L/mL to RNA purification reagent. Add TCEP prior to use of reagent.



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 0.4 mL of 0.1M ($^{\sim}$ 3%) TCEP solution by adding 80 μ L of 0.5M TCEP to 320 μ L of sterile RNase free water. Prepare more as required. Preparing fresh 0.1M TCEP is recommended.
- 2. Add 400 μ L of 0.1M TCEP directly to lyophilized thiolated oligos. Vortex to completely dissolve. Leave at room temperature (RT) for 1 hr to reduce the thiol groups. Vortex intermittently.
- 3. Add 50 µL of 3M Sodium Acetate pH 5.2 and vortex.
- 4. Add 1.5 mL of absolute ethanol, vortex and store at -20°C for 20 minutes.
- 5. Centrifuge at 12K rpm for 10 minutes. Decant ethanol and air dry pellet.
- 6. Dissolve in 200 μL of sterile RNase free water or choice of buffer volume as required.
- 7. Determine sample concentration by obtaining an absorbance at 260 nm

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 $^{\sim}6$ 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
TCEP 0.5M solution ; 1mL	40-5116-10	1 mL
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						
Catalog No.	Size*					
40-4080-05	50					
40-4080-10	100					
40-4080-50	500					
40-4090-05	50					
40-4090-10	100					
40-4090-50	500					
40-4081-05	50					
40-4081-10	100					
40-4081-50	500					
40-4091-05	50					
40-4091-10	100					
40-4091-50	500					
40-3650-01	100					
40-3650-05	500					
	Catalog No. 40-4080-05 40-4080-10 40-4080-50 40-4090-05 40-4090-10 40-4090-50 40-4081-05 40-4081-10 40-4091-05 40-4091-10 40-4091-50 40-3650-01					

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



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