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## Catalog # 15-6375

Tris(3-hydroxypropyl)phosphine, min. 84% THPP

# Catalog # 15-7400

Tris(2-carboxyethyl)phosphine, hydrochloride, 99% TCEP

### Properties of Tris(3-hydroxypropyl)phosphine and Tris(2-carboxyethyl)phosphine, hydrochloride

Tris(3-hydroxypropyl)phosphine (TPP or THPP) and Tris(2-carboxyethyl)phosphine, hydrochloride are, water soluble and used as neutral sulfhydryl reducing agents. They have a greater reducing capacity than dithiothreitol (DTT). In addition, THPP and TCEP are suitable for use in immobilized metal affinity chromatography because they do not reduce the metals involved.

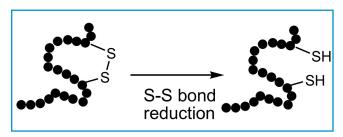
### **Advantages of THPP and TCEP as Reducing Agents**

In biological systems, to maintain biological activity, it is important that the thiol groups in peptides and proteins remain in their reduced state. Thiols, such as 2-mercaptoethanol and DTT, are commonly used as disulfide reductants. However, DTT can interfere in the desired reactions of SH groups. It can also be oxidized by metal-affinity columns, and thus must be re moved prior to peptide modification or purification.

Trialkylphosphines, on the other hand, exhibit high selectivity for disulfide bonds, but are generally not water soluble and have an unpleasant odor. The high water solubility of THPP and TCEP, therefore, make these two phosphines ideal alternatives to DTT. Unlike DTT, they generally do not need to be removed prior to thiol modification. THPP has the added advantage of being effective over a wide pH range<sup>[1]</sup>.

### **Applications of THPP and TCEP as Reducing Agents**

THPP and TCEP are used to break disulfide bonds within and between proteins in a wide range of biological applications, such as protein cleavage and/or precipitation for molecular diagnostics, and gene chips and microarrays. In fact, THPP and TCEP are both found in numerous enzyme kits for biological processing, such as protein extraction<sup>[2]</sup>, specific amino acid sequence cleavage<sup>[3]</sup>, and protein in-gel tryptic digestion<sup>[4]</sup>.



HO C OH + R S R 
$$H_2O$$
 HO C OH + 2R—SH Reduction of organic disulfide bonds with TCEP.

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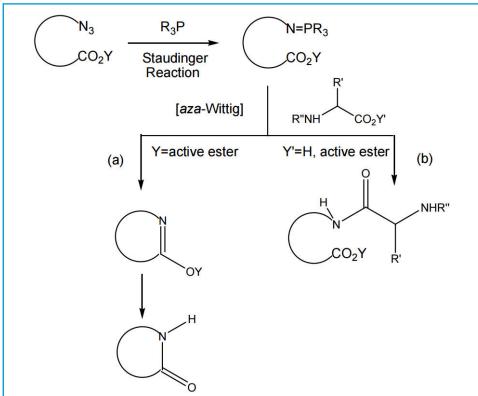
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### Some example literature citations that demonstrate the range of uses for THPP and TCEP include:

- "The 3'-Flap Pocket of Human Flap Endonuclease 1 Is Critical for Substrate Binding and Catalysis." L.D. Finger, et al., *J. Biol. Chem*, **2013**, *288*, 34239-34248.
- "Flavin-linked Erv-family sulfhydryl oxidases release superoxide anion during catalytic turnover." V.D. Daithankar, et al., *Biochem.*, **2012**, *51*(1), 265–272.
- "Site-specific chemical modification of recombinant proteins produced in mammalian cells by using the genetically encoded aldehyde tag." P. Wu, et al., *Nat Protoc.*, **2012**, *7*(6), 1052–1067.
- "A tris(2-carboxyethyl)phosphine (TCEP) related cleavage on cysteine-containing proteins." P. Liu, et al. *J. Am. Soc. Mass Spec.* **2010**, *21*(*5*), 837-844.
- "Reducing agent-mediated precipitation of high-abundance plasma proteins." S.E. Warder, et al., *Anal Biochem.* **2009**, *387(2)*, 184-93.
- "Novel reductant for the determination of total plasma homocysteine." B.M. Gilfix, et al., Clin. Chem., 1997, 43(4), 687-688.

### Other Applications of THPP and TCEP

THPP is also useful for the removal of protecting groups from S-protected cysteins, the deoxygenation of sulfoxides, N-oxides, and other sulfur and nitrogen compounds<sup>[5]</sup>. It is also a reagent for peptide synthesis, particularly in Mitsunobu and Staudinger reactions<sup>[6]</sup>.



Peptide-bond formation, using Staudinger-Wittig reaction

TCEP, meanwhile, does not impede maleimide attachment, and is suitable as a reagent for the quantitative analysis of sulfides, disulfides, hypochlorite ions, iodine, iodate ions, sulfoxides, N-oxides, azides<sup>[7]</sup>, and the selective reduction of disulfides for organic synthesis<sup>[8]</sup>.

#### References:

- 1. US Patent Application 60/469, 821.
- 2. Novagen Protocol TB245 Rev. E 0304, BugBuster® Protein Extraction Reagent; Novagen User Protocol TB316 Rev. B 0804, YeastBuster™ Protein Extraction Reagent.
- 3. Takara HRV 3C Protease Product Manual v201304Da.
- 4. Agilent 5188-2749Protein In-Gel Tryptic Digestion Kit Instructions.
- 5. G.A. Olah, B.G.B. Gupta, S.. Narang, SYnthesis. 137 (1978)
- 6. (a.) I. Bosch, F. Urpf, J. Vilarrasa, J. Chem. Soc., Chem. Commun. 91(1995); (b) H.Fuwa e.a., Tetrahedron Lett. 45 2323 (2004); J. S. Davies, J. Peptide Sci. 9 471-501 (2003).
- 7. A.-M.Faucher, C. Grand-Maitre, Synth. Commun. 33 3503 (2003).
- 8. J.A. Burns, J.C. Butler, J.Moran, G.M. Whitesides, J. Org. Chem. 1991, 56, 2648-2650.

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