

Protein Purification

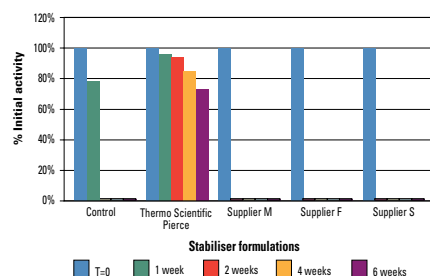
Purification reagents - Stabilising/reducing agents

Protein stabilising cocktail, Thermo Scientific Pierce

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Luciferase activity stabilised by the Thermo Scientific Pierce protein stabilising cocktail.

Commercially available luciferase was stored at 50µg/mL in our protein stabilising cocktail in a 30°C accelerated stability study. Fluorescence was measured at time 0 (dark bars) and after 1, 2, 4 and

6 weeks of storage. Luciferase stored in protein stabilising cocktail maintained 85% of its original activity after four weeks compared to a Tris-buffered salt formulation (control) and other suppliers stabilising agents, which were completely inactive after two weeks.

Extends shelf life of precious proteins. The Thermo Scientific Pierce Protein stabilising cocktail is a versatile stabilising solution that increases the shelf life of purified or partially purified proteins during routine storage.

- Does not destabilise biomolecules in downstream assays
- All components can be removed by dialysis
- Easy to pipette (vs. 50% glycerol)
- Restriction enzyme
- Cytokine
- Phosphatase
- Luciferase
- Antibody
- Peroxidase
- Kinase

Protein classes tested:

Provided as an easily pipettable, buffered 4X concentrate. After dilution in protein stabilising cocktail, the protein may be stored in the manner typical for the specific protein (4°C or -20°C). Although the degree of stabilisation is protein-specific, the cocktail significantly stabilises most proteins compared with conventional buffer alone.

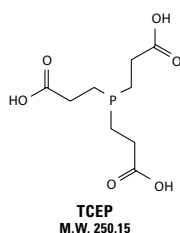
Catalogue No	Description	Quantity
PN89806	Protein stabilising cocktail, 4X concentrated solution Sufficient reagent to make 40mL of storage solution	10mL

Disulfide reducing agents, Thermo Scientific Pierce

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Reduce disulfide bonds to produce sulfhydryl groups for immobilisation.

Free sulfhydryls are required for immobilisation onto sulfhydryl-reactive affinity supports. Cysteines in proteins and peptides usually exist as cystines (disulfide bridges) and must be reduced to expose sulfhydryls for coupling. Reduction can be accomplished with free or immobilised reducing agents. Free reducing agents are efficient in reducing all disulfides in proteins, including those buried in the tertiary structure, but they must be removed from the reduced sample with a desalting column before coupling to the support. Immobilised reducing agents enable reduction of disulfides and simple removal of the reduced sample from the reducing agent.

Catalogue No	Description	Quantity
PN20408	2-Mercaptoethylamine • HCl	6 x 6mg
PN20290	DTT, Cleland's reagent (Dithiothreitol)	5g
PN20291	Dithiothreitol (DTT) in No-Weigh format 7.7mg DTT/tube. Makes 100µL of 0.5M DTT	48 tubes
PN20490	TCEP•HCl, 1g (Tris[2-carboxyethyl]phosphine hydrochloride)	1g
PN20491	TCEP•HCl	10g
PN77720	Bond-Breaker TCEP solution, neutral pH, 5mL Stable, 0.5M solution	5mL
PN77712	Immobilised TCEP Disulfide reducing gel	5mL

Dithioerythritol (DTE, Erythro-1,4-dimercapto-2,3-butanediol), Electrophoresis grade, white crystals or powder

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Fisher BioReagents®

Dithioerythritol is used to reduce disulfide bonds in proteins and peptides and is added in low concentrations as an enzyme stabiliser during protein purification and enzymatic reactions.

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Catalogue No	Quantity
BPE173-5	5g
<chem>HSCH2CH(OH)CH(OH)CH2SH</chem>	
M.W. 154.25	
Product specification	
Assay	≥99%
Melting point	83° ±2°C
Corrected absorbance of disulfide (S-S) in 0.02M solution at 285nm	≤0.13
Electrophoresis	Passes test
IR	Conforms to standard

Dithiothreitol white crystals or powder, (DTT, Cleland's Reagent), Molecular Biology grade

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Fisher BioReagents®

Dithiothreitol is used to reduce disulfide bonds in proteins and peptides and is added in low concentrations as an enzyme stabiliser during protein purification and enzymatic reactions.

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Catalogue No	Quantity
BPE172-5	5g
BPE172-25	25g
<chem>HSCH2CH(OH)CH(OH)CH2SH</chem>	
M.W. 154.25	
Product specification	
Assay	≥99%
IR	Conforms to standard
Melting point	41°C ±2°C
Corrected absorbance of disulfide (S-S) in 0.02M solution at 280nm	≤0.04
Electrophoresis	Passes test
DNase	Not detected
RNase	Not detected
Protease	Not detected

Glutathione reduced white crystalline powder, (γ-L-Glutamyl-cysteinylglycine)

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Glutathione is a useful tripeptide involved in many aspects of metabolism, including transport of γ-glutamyl amino acids and reductive cleavage of disulfide bonds. It is also used in purification of GST Tag proteins.

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Catalogue No	Quantity
BPE2521-1	1g
BPE2521-5	5g
BPE2521-10	10g
BPE2521-50	50g
<chem>C6H12N2O6S</chem>	
M.W. 307.3	
Product specification	
Assay	≥98%
Solubility 3.5% aqueous solution	Clear, colourless solution
pH	Acidic
FTIR	Conforms to standard
Optical rotation (c=2, H ₂ O)	-16°C ±5°C

2-Mercaptoethanol (2-Hydroxy-1-ethanethiol, 2-Hydroxyethylmercaptan, Thioglycol), Molecular Biology Grade

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Reduces disulfide linkages in proteins and peptides and is used in many enzymatic reactions as a protein stabiliser.

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Catalogue No	Quantity
BPE176-100	100g
<chem>HSCH2CH2OH; C2H6OS</chem>	
M.W. 78.13	
Product specification	
Assay	≥98%
Total disulfide (S-S) absorbance of a 0.02M solution at 285nm	≤0.05
Electrophoresis	To pass test
DNase	Not detected
RNase	Not detected