

Optimize protein integrity and yields during workflows

Broad-spectrum protease and phosphatase inhibitor tablets.

Inside a cell, proteins are commonly separated from proteolytic enzymes through differential localization. Disruption of cellular and tissue architecture during protein extraction distorts the *in vivo* state by making all proteins potentially accessible for degradation or modification by endogenous proteases and phosphatases. Unregulated, these activities can reduce protein yield and function in a sample. To minimize these losses, protease and phosphatase inhibitors can be added during cell lysis to prevent degradation of extracted proteins and preserve protein structure, function, and yields during sample preparation.

Protease and phosphatase inhibitors

Protease and phosphatase inhibitor cocktails and tablets are ideal for protecting proteins during extraction procedures or lysate preparation using primary cells, cultured mammalian cells, animal tissues, plant tissues, yeast cells, or bacterial cells. Formulations are packaged in multiple sizes, and EDTA-free versions are available for divalent cation-sensitive assays. Thermo Scientific™ Pierce™ inhibitor mini tablets have been reformulated to dissolve quickly into a clear solution and are fully compatible with all Pierce protein assays. Figures 1 and 2 provide representative examples of the effectiveness of Pierce Protease and Phosphatase Inhibitor Mini Tablets.

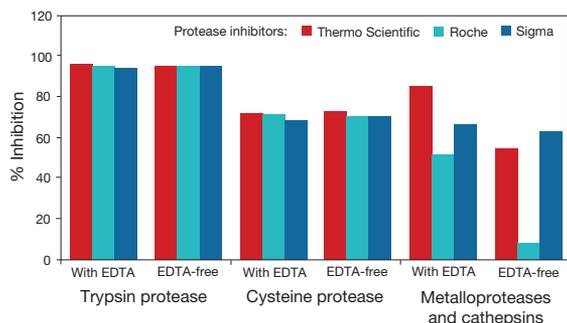


Figure 1. Performance comparison between three commercially available protease inhibitor tablets. Pancreatic extract (100 μ L, 0.5 μ g/ μ L) was incubated with cleavable fluorogenic substrates for trypsin and cysteine proteases, metalloproteases, and cathepsins in the presence of Thermo Scientific™ Pierce™ Protease Inhibitor Mini Tablets (Cat. No. A32953), Roche cOplete™ Protease Inhibitor Tablets, and Sigma-Aldrich SIGMAFAST™ Protease Inhibitor Cocktail Tablets, with and without EDTA. Reactions were incubated for 1 hr at 37°C, and fluorescence was determined at the appropriate detection emission wavelengths on a Thermo Scientific™ Varioskan™ Flash microplate reader. Percent protease inhibition is shown for each protease inhibitor formulation.

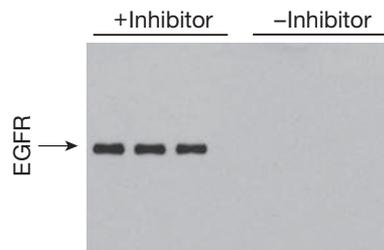


Figure 2. Inhibition of phosphatase activity in human colorectal carcinoma cell lysate. HCT116 cells were serum-starved and treated with epidermal growth factor (EGF) for 15 min or left untreated as controls (data not shown). Cell lysates were prepared in Thermo Scientific™ Pierce™ IP Lysis Buffer (Cat. No. 87788) with or without Thermo Scientific™ Pierce™ Protease and Phosphatase Inhibitor Mini Tablets, EDTA-Free (Cat. No. A32961) and incubated with anti-phosphotyrosine antibody overnight at 4°C. The immunoprecipitated complex was incubated with Thermo Scientific™ Pierce™ Protein A/G Magnetic Beads (Cat. No. 88803) for 1 hr at room temperature. After washing the beads, a low-pH elution was performed, and this eluate was run on a gel, transferred to a membrane, and probed with anti-EGF receptor (EGFR) antibody prior to chemiluminescence western blot detection.

Download the free Protein Preparation Handbook

The Protein Preparation Handbook provides technical information on our broad portfolio of reagents and tools for protein extraction, cleanup, immunoprecipitation, and purification, including the inhibitor tablets discussed here. Practical information, selection guides, and relevant data are included to help improve protein yield, facilitate downstream analysis, and optimize workflows. Download this free brochure at thermofisher.com/proteinprephandbook. We have also produced a series of self-paced animated eLearning modules—including a free course on protein sample preparation—which can be accessed at thermofisher.com/elearningcourses. ■

Product	Quantity	Cat. No.
Pierce™ IP Lysis Buffer	100 mL	87787
	250 mL	87788
Pierce™ Protease Inhibitor Mini Tablets	30 tablets	A32953
Pierce™ Protease Inhibitor Tablets	20 tablets	A32963
Pierce™ Protease Inhibitor Mini Tablets, EDTA-free	30 tablets	A32955
Pierce™ Protease Inhibitor Tablets, EDTA-free	20 tablets	A32965
Pierce™ Phosphatase Inhibitor Mini Tablets	20 tablets	A32957
Pierce™ Protease and Phosphatase Inhibitor Mini Tablets	20 tablets	A32959
Pierce™ Protease and Phosphatase Inhibitor Mini Tablets, EDTA-free	20 tablets	A32961