

Ubiquitin Linkage Screening Kit I (Cat. # J3200)

*The supplied UBE1 is sufficient for setting up 50 x 20 μ l reactions. Each Ub mutant is sufficient for setting up 10 X 20 μ l reactions.

Description

Ubiquitin Linkage Screening Kit I is designed for two purposes: 1) to identify the ubiquitin lysine residue that is used to link the polyubiquitin chain conjugated on a protein substrate; 2) to determine whether a protein substrate is polyubiquitinated or conjugated with multiple monoubiquitin.

A typical 20 μ l reaction contains 100 nM UBE1, 2 μ M E2 (not supplied), 50 μ M Ub or Ub mutants, 4 mM ATP, 2 μ M E3 (not supplied) and 2 μ M substrate (not supplied) in the ubiquitination buffer.

Components

• 20X UBE1 (2 μ M)	50 μ l
• 10X Ub (500 μ M)	20 μ l
• 10X Ub(K0) (500 μ M)	20 μ l
• 10X Ub(K6R) (500 μ M)	20 μ l
• 10X Ub(K11R) (500 μ M)	20 μ l
• 10X Ub(K27R) (500 μ M)	20 μ l
• 10X Ub(K29R) (500 μ M)	20 μ l
• 10X Ub(K33R) (500 μ M)	20 μ l
• 10X Ub(K48R) (500 μ M)	20 μ l
• 10X Ub(K63R) (500 μ M)	20 μ l
• 10X ATP (40 mM)	125 μ l
• 10X Ubiquitination Buffer	1 ml

Note

1. 10X Ubiquitination Buffer: 400 mM Tris, pH 7.1 at 37°C, 400 mM NaCl, 10 mM β ME, 50 mM $MgCl_2$

