Lot # XXXXX

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MATERIAL DATA SHEET

MuRF1/TRIM63 Auto-Ubiquitination Kit
Cat. # K-100

This kit is designed to perform E3 ligase auto-ubiquitination reactions *in vitro*, which requires the activities of the ubiquitin E1 activating enzyme (**E-305**), the E2 conjugating enzyme UbcH5c (**E2-627**) and E3 ligase MurF-1 (**E3-100**). The E1 enzyme charges the ubiquitin by forming an ATP-dependent high energy thiolester bond. The activated ubiquitin is subsequently transferred to UbcH5c then to the MurF-1. The MurF-1-S-Ub complex has the ability to both auto-ubiquitinate itself and/or transfer the Ub to protein substrates (eg. troponin). Alternatively labeled Ub proteins may be substituted for biotin-ubiquitin for detection such as fluorescein-ubiquitin (**U-590**) and rhodamine-ubiquitin (**U-600**).

NOTE: Kit contains reagents sufficient for 10 x 20 µl reactions.

Concentration of components vary with Lot #.

Product Information

		Concentration	Volume
Supplied:	**1. 10X E1 Enzyme**	X mg/ml (X µM)	20 µl
	2. 10X UbcH5c	X mg/ml (X µM)	20 µl
	3. 5X His_6-MuRF-1	X mg/ml (X µM)	40 µl
	4. 10X Biotin-Ubiquitin	X mg/ml (X mM)	20 µl
	5. 10X Reaction Buffer	X mM	20 µl

Storage: Store at -80°C. Avoid multiple freeze/thaw cycles.

Background

MurF1 (**Mu**scle-specific **R**ING-**f**inger protein **1**) is a RING-finger E3 ligase found in striated muscle (heart and skeletal) and iris tissues. MurF1 shares 62% and 77% sequence homology with MurF2 and MurF3 respectively. The protein contains a RING-finger/B-box/coiled-coil tripartite fold known as TRIM, and it can form homo- and hetero-oligomers vial the coiled-coil dimerization motifs. The N-terminal E3 ligase activity regulates the proteasomal degradation of cardiac troponin and probably other sarcomeric-associated proteins. The exact cellular roles and substrates for MurF1 are not yet known but it interacts with various proteins including titin, isopeptidase3, SUMO-3, UbcH9 and GMEB-1. MurF1 may have a role in muscle adaptation, atrophy and hypertrophy mediated by ubiquitin-proteasome pathways (UPP). Muscle atrophy is associated with many diseases including cancer, diabetes and AIDS.

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