## Ubiquitin activating enzyme E1 (human), (recombinant) (Histag)

Ediquitintativation continues to the first spensible for the first step in ubiquitin-protein isopeptide bond formation and is a critical component for the initiation of *in vitro* conjugation reactions. E1 activates ubiquitin by first adenylating with ATP the C-terminal glycine carboxyl group of ubiquitin and, thereafter, linking this residue to the sulphydryl side chain moiety of a cysteine residue in E1 by forming a high energy thiol ester bond and liberating free AMP. There are two active sites within the E1 molecule allowing it to accommodate two ubiquitin molecules at one time, with a new ubiquitin forming an adenylate intermediate as the previous one is transferred to the thiol site. The activated ubiquitin is then transferred to the lysine of target proteins via the E2/E3 conjugation cascade.

Results demonstrate the formation of ubiquitin thioester linked E2 conjugates of the expected size in all TE +ve control reactions. The absence of such conjugates in TE –ve control reactions demonstrates that their formation is ATP dependent (required for E1 activation) and hence derived from the ubiquitin cascade.

Citations: 29

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**Ordering Information** 

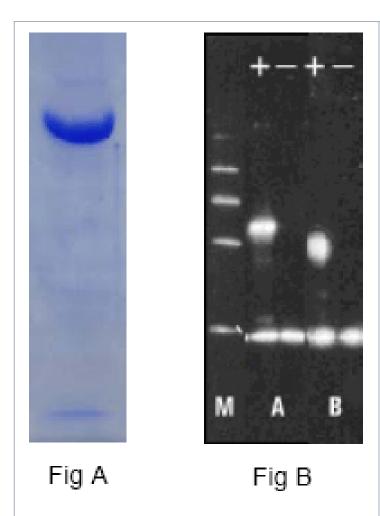
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BML-UW9410-0050

50µg

Manuals, SDS & CofA

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**Figure A:** Product purity determined by Coomassie staining following SDS-PAGE

**Figure B:** Western Blot of thioester assays (TE +ve/-ve controls) for E2 conjugating enzymes. Biotinylated-ubiquitin-enzyme conjugates were detected by western blotting on thioester assays containing: A: UbcH2 (Prod. No. BML-KW9025), B: UbcH8 (KW9135), respectively, using Streptavidin-HRP detection system. M: Biotinylated SDS molecular weight markers (Sigma, SDS-6B) from bottom: 20.1, 29.0, 39.8, 58.1kDa.

## **Handling & Storage**

**Handling** Avoid freeze/thaw cycles.

Long Term Storage -80°C

Shipping Dry Ice

Regulatory Status RUO - Research Use Only

**Product Details** 

**Formulation** Liquid. In 50mM TRIS-HCI, pH 7.5, containing 1.0mM DTT.

**MW** 118 kDa

Purity ≥90% (SDS-PAGE)

**Purity Detail** Purified by multi-step chromatography.

**Source** Produced in *E. coli* BL21 (λDE3) expression system and containing a C-terminal His6-

tag. Full length human ubiquitin-activating enzyme E1.

UniProt ID P22314



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