Differential Maturation of SUMO Precursors by SUMO-specific Protease, SENP1

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Small ubiquitin-related modifier (SUMO) is a member of the ubiquitin-like protein family that regulates cellular function of a variety of target proteins. SUMO proteins are expressed as their precursor forms. Cleavage of the residues after the "G-G" region of these precursors by SUMO specific proteases in maturation is a prerequisite for subsequent sumoylation. To further understand this proteolytic processing, we expressed and purified SENP1, one of the SUMO specific proteases, using an *E. coli* expression system. We show that SENP1 is able to process all SUMO-1, -2 and -3 *in vitro*, however the proteolytic efficiency of SUMO-1 is the highest followed by SUMO-2 and SUMO-3. We further demonstrate the catalytic domain of SENP1 (SENP1C) alone can determine the substrate specificity towards SUMO-1, -2 and -3. Using mutagenesis analysis, two residues immediately after the "G-G" region are mapped to be essential for the differential maturation. At present, crystals of inactive SENP1C and SUMO-1 have been obtained. Future structural analysis will provide insight into the molecular basis of the differential maturation process.

Keywords: small ubiquitin-related modifier (SUMO), sentrin-specific protease 1 (SENP1) proteases, sumoylation