Linear ubiquitin binding via A20 zinc finger7 is involved in the canonical NF-κB regulation

Fuminori Tokunaga¹, Hiroshi Nishimasu², Ryuichiro Ishitani², and Osamu Nureki²

¹Lab. of Mol. Cell Biol., Inst. for Mol. and Cell. Reg., Gunma Univ., Maebashi 371-8512, Japan, and ²Dept. of Biophys. and Biochem., Grad. Sch. of Sci., The Univ. of Tokyo, Bunkyo-ku, Tokyo 113-0032, Japan

Abstract

LUBAC (linear ubiquitin chain assembly complex) composed of HOIL-1L, HOIP, and SHARPIN, regulates the canonical NF-κB pathway through linear polyubiquitination of NEMO and RIP1. However, deubiquitinase (DUB), which down-regulates LUBAC-mediated NF-κB activation, remains elusive. We identified that A20 (TNFAIP3) suppresses LUBAC-mediated NF-κB activation by the specific binding to linear polyubiquitin via the C-terminal seventh zinc finger (ZF7), whereas CYLD suppresses it through DUB activity. We determined the crystal structures of A20 ZF7 in complex with linear diubiquitin at 1.70–1.98 Å resolutions. The crystal structures revealed that A20 ZF7 simultaneously recognizes the Met1-linked proximal and distal ubiquitins with a novel manner. Importantly, genetic mutations associated with B cell lymphomas, such as N772K and E781D, map to the ubiquitin-binding sites, and our functional analysis indicated that the binding of A20 ZF7 to linear polyubiquitin contributes to the recruitment of A20 into a TNF receptor (TNFR) signalling complex containing LUBAC and IκB kinase (IKK), which results in NF-κB suppression. These findings provide new insight into the regulation of immune and inflammatory responses, and the linear ubiquitin-binding through A20 ZF7 will provide a novel therapeutic target for B cell lymphomas.