USP11 (human; full length), pAb

Alternate Names: Ubiquitin carboxyl-terminal hydrolase 11, Deubiquitinating enzyme 11, Ubiquitin thioesterase 11, Ubiquitin-specific-processing protease 11, UHX1

Cat. No.	**68-0028-100**	Quantity:	100 µg
Lot. No.	**30265**	Storage:	-20˚C

FOR RESEARCH USE ONLY

NOT FOR USE IN HUMANS

CERTIFICATE OF ANALYSIS

This antibody was developed and validated by the Medical Research Council Protein Phosphorylation and Ubiquitylation Unit (University of Dundee, Dundee, UK).

Background

Deconjugating enzymes (DCEs) are proteases that process ubiquitin or ubiquitin-like gene products, reverse the modification of proteins by a single ubiquitin or ubiquitin-like protein (UBL) and remodel polyubiquitin (or poly-UBL) chains on target proteins (Reyes-Turcu *et al.*, 2009). The deubiquitylating – or deubiquitinating – enzymes (DUBs) represent the largest family of DCEs and regulate ubiquitin-dependent signalling pathways. The activities of the DUBs include the generation of free ubiquitin from precursor molecules, the recycling of ubiquitin following substrate degradation to maintain cellular ubiquitin homeostasis and the removal of ubiquitin or ubiquitin-like proteins (UBL) modifications through chain editing to rescue proteins from proteasomal degradation or to influence cell signalling events (Komander *et al.*, 2009). There are two main classes of DUB, cysteine proteases and metalloproteases. Ubiquitin specific processing peptidase 11 (USP11) is a member of the cysteine protease enzyme family and cloning of the gene in humans was first described by Swanson *et al.* (1996). USP11 interacts with RanBPM, a Ran-binding protein, which is required for correct microtubule nucleation. USP11 specifically associates with RanBPM and inhibits its ubiquitylation and degradation. Ran has been well char-

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Physical Characteristics

Quantity: 100 µg

Concentration: to be provided on shipping

Source: sheep polyclonal antibody

Immunogen: human USP11 (residues 1-963)

Purification: affinity-purified using immobilized immunogen

Formulation: phosphate-buffered saline

Specificity: detects USP11 at ~110 kDa

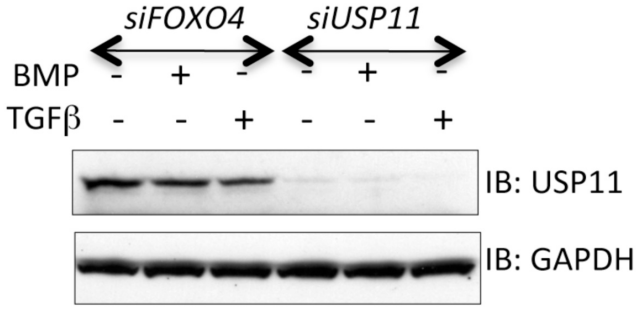
Reactivity: human; other species not tested.

Stability/Storage: 12 months at -20˚C; aliquot as required

Research Applications and Quality Assurance

Western Immunoblotting:
Use 0.1 µg/ml

Immunoprecipitation:
Use 2 µg/mg cell extract



Western Blotting Analysis:
HaCaT cells stimulated with either TGFβ or bone morphogenic protein (BMP) were transfected with siRNA against USP11 or siRNA targeting FOXO4 as a control. The cells were then lysed and the lysates denatured in SDS and subjected to SDS-PAGE on 8% gels. Western Blotting was carried out with 0.1 µg/ml anti-USP11 antibody (Cat. No 68-0028-100) on 20 µg total cell extract.

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Lot-specific COA version tracker: v1.0.0

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Background

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acterised as a molecule required for nuclear protein import and export, and is also involved in spindle formation of mitotic and meiotic cells (Ideguchi *et al.*, 2002). USP11 has been shown to bind to HPV-16E7 (a major transforming protein, which has been implicated in the development of cervical cancer) and trims ubiquitin from HPV-16E7 which avoids the targeting of HPV-16E7 for proteasome degradation and functionally extends the half-life of HPV-16E7. Additionally, USP11 augments the HPV-16E7 function in modulating downstream target genes, such as tumour suppressor retinoblastoma protein (pRb), Bcl-2, and Cdc-2, suggesting that this interaction may contribute to cell transformation by HPV-16E7 (Lin *et al.*, 2008). USP11 also plays a critical role in the downregulation of NF-κB activation. Ubiquitylation-mediated IκBα degradation immediately precedes and is required for NF-κB nuclear translocation and activation. USP11 is an IκBα associated deubiquitylase, is constitutively associated with IκBα and attenuates IκBα degradation to negatively regulate TNFα-induced NF-κB activation (Sun *et al.*, 2010).

Antibody Production:
Anti-USP11 (human) polyclonal antibody was raised in sheep against USP11 (residues 1-963 of human USP11). The antibodies were purified by the Medical Research Council Protein Phosphorylation and Ubiquitylation Unit (MRC-PPU, University of Dundee, Dundee, U.K.) by affinity purification of the anti-USP11 pAbs from the sheep serum using a GST-tagged antigen-agarose column. Anti-USP11 (human) pAb was sourced by Ubiquigent directly from the MRC-PPU.

General References:

Ideguchi H, Ueda A, Tanaka M, Yang J, Tsuji T, Ohno S, Hagiwara E, Aoki A, Ishigatsubo Y (2002) Structural and functional characterization of the USP11 deubiquitinating enzyme, which interacts with the RanGTP-associated protein RanBPM. *Biochem J* **367**, 87-95.

Lin C, Chang HS, Winston Y (2008) USP11 Stabilizes HPV-16E7 and Further Modulates the E7 Biological Activity. *J Biol Chem* **283**, 15681–15688.

Maertens G, Messaoudi-Aubert S, Elderkin S, Hiom K, Peters G (2010) Ubiquitin-specific proteases 7 and 11 modulate Polycomb regulation of the INK4a tumour Suppressor. *EMBO J* **29**, 2553–2565.

Schoenfield AR, Apgar S, Dolios G, Wang R, Aaronson SA (2004) BRCA2 is ubiquitinated *in vivo* and interacts with USP11, a deubiquitinating enzyme that exhibits prosurvival function in the cellular response to DNA damage. *Mol Cell Biol* **24**, 7444-55.

Sun W, Tan X, Shi Y, Xu G, Mao R, Gu X, *et al.* (2010) USP11 negatively regulates TNFalpha-induced NF-kappaB activation by targeting on IkappaBalpha *Cellular Signalling* **22**, 386-94.

Swanson DA, Freund CL, Ploder L, McInnes RR, Valle, D (1996) A ubiquitin C-terminal hydrolase gene on the proximal short arm of the X chromosome: implications for X-linked retinal disorders. *Hum Molec Genet* **5**, 533-538.

Application Reference:

Al-Salihli MA, Herhaus L, Macartney T and Sapkota GP (2012) USP11 augments TGFbeta signalling by deubiquitylating ALK5. *Open Biology* **2**, 120063.

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Lot-specific COA version tracker: v1.0.0