

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

## Background

The enzymes of the NEDDylation pathway play a pivotal role in the activation of the largest class of ubiquitin E3 ligases called Cullin-RING-Ligases (CRLs). Akin to ubiquitylation three classes of enzymes are involved in the process of mammalian NEDDylation; E1 activating enzyme (APP-BP1/ UBA3 heterodimer), E2 conjugating enzymes (UBE2M or UBE2F) and E3 ligases the defective in Cul NEDDvlation 1 domain-containing proteins (DCUN1D1-5) (Meyer-Schaller et al., 2009; Huang et al., 2011). There are 5 human DCUN1D1-5 proteins are also named defective in Cul NEDDylation 1 like proteins (DCNL1-5) (Meyer-Schaller et al., 2009). Cloning of DCNL5 was first described by Lamesch et al. (2007). The DCNLs have distinct aminoterminal domains, but share a conserved Cterminal potentiating NEDDylation (PONY) domain (Kurz et al., 2008). It has been determined that the interaction between the DCNLs and Cul1 occurs through the PONY domain and the Winged Helix DNA binding domain (WHB) respectively (Kurz et al., 2008; Scott et al., 2011). Pairwise analysis of 30 combinations of the five DCNL PONY domains and six cullin WHB subdomains by isothermal titration calorimetry have all shown interaction albeit with differing affinities (Monda et al., 2013).

# DCNL5 (mouse; full length), pAb

Alternate Names: DCUN1D5, FLJ32431, FLJ37425, MGC2714

	Quantity: Storage:	100 µg -20°С
--	-----------------------	-----------------

FOR RESEARCH USE ONLY

**CERTIFICATE OF ANALYSIS** 

NOT FOR USE IN HUMANS

Page 1 of 2

# **Physical Characteristics**

Quantity: 100 µg

Concentration: to be provided on shipping

Source: sheep polyclonal antibody

Immunogen: mouse DCNL5 (residues 1 – 237) [GST-tagged]

Purification: affinity-purified using immobilized immunogen

Formulation: phosphate-buffered saline

Specificity: detects DCNL5 at ~28 kDa

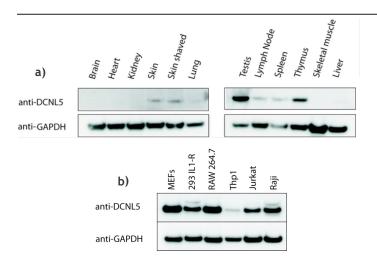
Reactivity: mouse; other species not tested

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

# **Research Applications and Quality Assurance**

Western Immunoblotting: Use 1.0 µg/ml

Immunoprecipitation: Use 5.0 µg/mg of cell extract



#### Western Blotting Analysis:

By western blotting the specific recognition of DCNL5 could be observed in several a) mouse tissues and b) cell lines when probed with 1.0 µg/ml of anti-mouse DCNL5 antibody (Cat# 68-0009-100).

Continued on page 2



**ORDERS / SALES SUPPORT** International: +1-617-245-0020 US Toll-Free: 1-888-4E1E2E3 (1-888-431-3233) Email: sales.support@ubiquigent.com

#### **UK HQ and TECHNICAL SUPPORT**

International: +44 (0) 1382 381147 (9AM-5PM UTC) US/Canada: +1-617-245-0020 (9AM-5PM UTC) Email: tech.support@ubiquigent.com

Email services@ubiquigent.com for enquiries regarding compound profiling and/or custom assay development services.

© Ubiquigent 2014. Unless otherwise noted, Ubiquigent, Ubiquigent logo and all other trademarks are the property of Ubiquigent, Ltd.

Limited Terms of Use: For research use only. Not for use in humans or for diagnostics. Not for distribution or resale in any form, modification or derivative OR for use in providing services to a third party (e.g. screening or profiling) without the written permission of Ubiquigent, Ltd.

Lot-specific COA version tracker: v1.0.0



### Background

Continued from page 1

#### **Antibody Production:**

Anti-DCNL5 (mouse) polyclonal antibody was raised in sheep against DCNL5 (residues 1-237 of mouse DCNL5). 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-DCNL5 pAbs from the sheep serum using an antigenagarose column followed by depletion of any anti-GST pAbs using a GST-agarose column. Anti-DCNL5 (mouse) pAb was sourced by Ubiquigent directly from the MRC-PPU.

#### General References:

Huang G, Kaufman AJ, Ramanathan Y, Singh B (2011) SCCRO (DCUN1D1) promotes nuclear translocation and assembly of the neddylation E3 complex. *J Biol Chem* **286**, 10297-10304.

Kurz T, Chou YC, Willems AR, Meyer-Schaller N, Hecht ML, Tyers M, Peter M, Sicheri F (2008) Dcn1 functions as a scaffold-type E3 ligase for cullin neddylation, *Mol Cell* **29**, 23-35.

Kurz T, Ozlü N, Rudolf F, O'Rourke SM, Luke B, Hofmann K, Hyman AA, Bowerman B, Peter M (2005) The conserved protein DCN-1/Dcn1p is required for cullin neddylation in *C. elegans and S. cerevisiae*, *Nature* 435, 1257-1261.

Lamesch P, Li N, Milstein S, Fan C, Hao T, Szabo G, Hu Z, Venkatesan K, Bethel G, Martin P, Rogers J, Lawlor S, McLaren S, Dricot A, Borick H, Cusick ME, Vandenhaute J, Dunham I, Hill DE,Vidal M (2007) hOR-Feome v3.1: a resource of human open reading frames representing over 10,000 human genes. *Genomics* **89**, 307-315.

Meyer-Schaller N, Chou YC, Sumara I, Martin DD, Kurz T, Katheder N, Hofmann K, Berthiaume LG, Sicheri F, Peter M (2009) The human Dcn1like protein DCNL3 promotes Cul3 neddylation at membranes. *PNAS* **106**, 12365-12370.

Monda J.K,Scott DC, Miller DJ, Lydeard J, King D, Harper JW, Bennett EJ, Schulman BA (2013) Structural Conservation of Distinctive N-terminal Acetylation-Dependent Interactions across a Family of Mammalian NEDD8 Ligation Enzymes. *Structure* **21**, 42-53.

Scott D.C, Monda JK, Bennett EJ, Harper JW, Schulman B.A (2011) Nterminal acetylation acts as an avidity enhancer within an interconnected multiprotein complex, *Science* **334**, 674-678.

# DCNL5 (mouse; full length), pAb

Alternate Names: DCUN1D5, FLJ32431, FLJ37425, MGC2714

Cat. No. Lot. No.	68-0009-100 30246	Quantity: Storage:	100 µg -20°С	
FOR RESEARCH	I USE ONLY	NOT FOR USE IN	NOT FOR USE IN HUMANS	
CERTIFICATE OF ANALYSIS		Page 2 of 2		

# WWW.Ubiquigent.com Dundee, Scotland, UK

# **ORDERS / SALES SUPPORT**

 International:
 +1-617-245-0020

 US Toll-Free:
 1-888-4E1E2E3 (1-888-431-3233)

 Email:
 sales.support@ubiquigent.com

#### **UK HQ and TECHNICAL SUPPORT**

 International:
 +44 (0) 1382 381147
 (9AM-5PM UTC)

 US/Canada:
 +1-617-245-0020
 (9AM-5PM UTC)

 Email:
 tech.support@ubiquigent.com

Email services@ubiquigent.com for enquiries regarding compound profiling and/or custom assay development services.

© Ubiquigent 2014. Unless otherwise noted, Ubiquigent, Ubiquigent logo and all other trademarks are the property of Ubiquigent, Ltd.

Limited Terms of Use: For research use only. Not for use in humans or for diagnostics. Not for distribution or resale in any form, modification or derivative QR for use in providing services to a third party (e.g. screening or profiling) without the written permission of Ubiquigent, Ltd.

Lot-specific COA version tracker: v1.0.0