

**Datasheet for 200-301-912****HEF1 Antibody****Overview**

|                      |                                                                 |
|----------------------|-----------------------------------------------------------------|
| <b>Description:</b>  | Anti-HEF1 (aa 82-398) (MOUSE) Monoclonal Antibody - 200-301-912 |
| <b>Item No.:</b>     | 200-301-912                                                     |
| <b>Size:</b>         | 100 µg                                                          |
| <b>Applications:</b> | IF, IP, WB                                                      |
| <b>Reactivity:</b>   | Human                                                           |
| <b>Host Species:</b> | Mouse                                                           |

**Product Details****Background:**

HEF1, also known as Enhancer of filamentation 1, CRK-associated substrate-related protein, CAS-L, CasL, p105 and Neural precursor cell expressed developmentally down-regulated 9 is the product of the NEDD9 (CASGL) gene. HEF1 functions as a docking protein that plays a central coordinating role for tyrosine-kinase-based signaling related to cell adhesion. HEF1 may also function in transmitting growth control signals between focal adhesions at the cell periphery and the mitotic spindle in response to adhesion or growth factor signals initiating cell proliferation. HEF1 may also play an important role in integrin beta-1 or B cell antigen receptor (BCR) mediated signaling in B- and T-cells. Integrin beta-1 stimulation leads to recruitment of various proteins including CRK, NCK and SHPTP2 to the tyrosine phosphorylated form. HEF1 forms a homodimer and can heterodimerize with HLH proteins ID2, E12, E47 and also with p130cas. HEF1 also forms complexes in vivo with related adhesion focal tyrosine kinase (RAFTK), adapter protein CRKL and LYN kinase and also interacts with MICAL and TXNL4/DIM1. This protein localizes to both the cell nucleus and the cell periphery and is differently localized in fibroblasts and epithelial cells. In fibroblasts is predominantly nuclear and in some cells is present in the Golgi apparatus. In epithelial cells localized predominantly in the cell periphery with particular concentration in lamellipodia but is also found in the nucleus. HEF1 is widely expressed although higher levels are detected in kidney, lung, and placental tissue. HEF1 is also detected in T-cells, B-cells and diverse cell lines. HEF1 is activated upon induction of cell growth. Cell cycle-regulated processing produces four isoforms: p115, p105, p65, and p55. Isoform p115 arises from p105 phosphorylation and appears later in the cell cycle. Isoform p55 arises from p105 as a result of cleavage at a caspase cleavage-related site and it appears specifically at mitosis. The p65 isoform is poorly detected. Isoforms p105 and p115 are predominantly cytoplasmic and associate with focal adhesions while p55 associate with the mitosis spindle.

**Synonyms:** mouse anti-HEF1 antibody, mouse anti-NEDD-9 antibody, mouse anti-CASL antibody, Cas like docking antibody, CASL antibody, Crk associated substrate related protein antibody, dJ49G10.2 antibody, dJ761I2.1 antibody, Enhancer of filamentation 1 antibody

**Host Species:** Mouse

**Clonality:** Monoclonal

**Clone ID:** 14A11

**Format:** IgG2b

## Target Details

**Gene Name:** NEDD9

**Reactivity:** Human

**Immunogen Type:** Conjugated Peptide

**Immunogen:** Anti-HEF1 monoclonal antibody was produced by repeated immunizations with a synthetic peptide corresponding to amino acid residues 82-398 of human HEF1 protein.

**Purity/Specificity:** This Protein A purified antibody is directed against human HEF1 protein. The product was purified from tissue culture supernatant by chromatography. This antibody has only been tested on human cells. Reactivity against multiple isoforms is expected. Reactivity against homologues from other sources is not known. Specificity was determined by partial epitope mapping.

**Relevant Links:**

- [UniProtKB - Q14511](#)
- [NCBI - 5453680](#)
- [GeneID - 4739](#)

## Application Details

**Tested Applications:** IF, IP, WB

**Application Note:** This monoclonal antibody has been tested for use in western blotting, immunoprecipitation and immunofluorescence. This clone recognizes HEF1 under non-denaturing conditions. Specific conditions for reactivity should be optimized by the end user. Expect bands approximately 115 and 105 in size corresponding to isoforms of HEF1 protein by western blotting in the appropriate cell lysate or extract. This antibody does not recognize p130Cas. Sin1 has not been tested. IF was performed using 4% PFA fixed cells. This monoclonal mostly detects HEF1 localized at the focal adhesion sites.

**Assay Dilutions:** All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.

|               |                    |
|---------------|--------------------|
| <b>ELISA:</b> | 1:5,000 - 1:20,000 |
| <b>IF:</b>    | 1:100              |
| <b>IP:</b>    | 1:100              |
| <b>WB:</b>    | 1:500              |

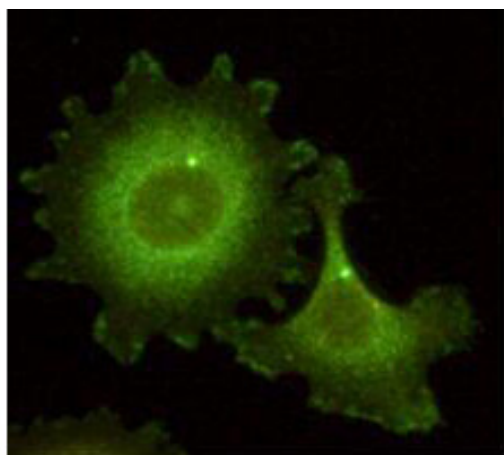
## Formulation

|                        |                                                            |
|------------------------|------------------------------------------------------------|
| <b>Physical State:</b> | Liquid (sterile filtered)                                  |
| <b>Concentration:</b>  | 1.0 mg/mL by UV absorbance at 280 nm                       |
| <b>Buffer:</b>         | 0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2 |
| <b>Preservative:</b>   | 0.01% (w/v) Sodium Azide                                   |
| <b>Stabilizer:</b>     | None                                                       |

## Shipping & Handling

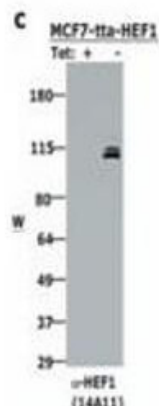
|                            |                                                                                                                                                                                                                                                                                                                                               |
|----------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <b>Shipping Condition:</b> | Dry Ice                                                                                                                                                                                                                                                                                                                                       |
| <b>Storage Condition:</b>  | Store vial at -20° C prior to opening. Aliquot contents and freeze at -20° C or below for extended storage. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4° C as an undiluted liquid. Dilute only prior to immediate use. |
| <b>Expiration:</b>         | Expiration date is one (1) year from date of receipt.                                                                                                                                                                                                                                                                                         |

## Images



### Immunofluorescence Microscopy

Immunofluorescence microscopy using Rockland's Monoclonal anti-HEF1 antibody (clone 14A11) shows detection of HEF1 localized at the centrosome (bright dots) and focal adhesion sites. The antibody was used at a 1:100 dilution with a 1-min exposure time. Personal Communication. Elena Pugacheva, Fox Chase Cancer Center, Philadelphia, PA.



### Western Blot

Western blotting using Rockland's Monoclonal anti-HEF1 antibody (clone 14A11) shows detection of HEF1 present in MCF-7 cells induced to express HEF1 by tetracycline removal (right lane). See Pugacheva et al for details.

## Disclaimer

This product is for research use only and is not intended for therapeutic or diagnostic applications. Please contact a technical service representative for more information. All products of animal origin manufactured by Rockland Immunochemicals are derived from starting materials of North American origin. Collection was performed in United States Department of Agriculture (USDA) inspected facilities and all materials have been inspected and certified to be free of disease and suitable for exportation. All properties listed are typical characteristics and are not specifications. All suggestions and data are offered in good faith but without guarantee as conditions and methods of use of our products are beyond our control. All claims must be made within 30 days following the date of delivery. The prospective user must determine the suitability of our materials before adopting them on a commercial scale. Suggested uses of our products are not recommendations to use our products in violation of any patent or as a license under any patent of Rockland Immunochemicals, Inc. If you require a commercial license to use this material and do not have one, then return this material, unopened to: Rockland Inc., P.O. BOX 5199, Limerick, Pennsylvania, USA.