U2AF65 (D-8): sc-374333



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BACKGROUND

SF3 β is an U2 snRNP-associated protein complex essential for spliceosome assembly. SF3 β contains the spliceosomal proteins SAPs 49, 130, 145 and 155. SAPs 130, 145 and 155 associate with one another to form a complex that is present in HeLa nuclear extracts. SAPs 49 and 145 are known to interact directly with each other. Unexpectedly, the SAP 49-SAP 145 protein-protein interaction requires the amino-terminus of SAP 49, which contains two RNA-recognition motifs. SAP 49 and SAP 145 interact directly with both U2 snRNP and the pre-mRNA, which suggests that this protein complex plays a role in tethering U2 snRNP to the branch site. U2AF recruits SAP 49 to the branch point sequence during the initial steps of spliceosome assembly. U2AF exists as a heterodimer consisting of U2AF65 and U2AF35 and is required for splicing *in vivo*.

REFERENCES

- Zamore, P.D. and Green, M.R. 1989. Identification, purification and biochemical characterization of U2 small nuclear ribonucleoprotein auxiliary factor. Proc. Natl. Acad. Sci. USA 86: 9243-9247.
- 2. Kanaar, R., et al. 1993. The conserved pre-mRNA splicing factor U2AF from *Drosophila:* requirement for viability. Science 262: 569-573.
- 3. Potashkin, J., et al. 1993. U2AF homolog required for splicing *in vivo*. Science 262: 573-576.
- 4. Champion-Arnaud, P. and Reed, R. 1994. The prespliceosome components SAP 49 and SAP 145 interact in a complex implicated in tethering U2 snRNP to the branch site. Genes Dev. 8: 1974-1983.
- Wells, S.E., et al. 1996. CUS1, a suppressor of cold-sensitive U2 snRNA mutations, is a novel yeast splicing factor homologous to human SAP 145. Genes Dev. 10: 220-232.
- 6. lgel, H., et al. 1998. Conservation of structure and subunit interactions in yeast homologues of splicing factor 3β (SF3 β) subunits. RNA 4: 1-10.
- Das, B.K., et al. 1999. Characterization of a protein complex containing spliceosomal proteins SAPs 49, 130, 145, and 155. Mol. Cell. Biol. 19: 6796-6802.

CHROMOSOMAL LOCATION

Genetic locus: U2AF2 (human) mapping to 19q13.42; U2af2 (mouse) mapping to 7 A1.

SOURCE

U2AF65 (D-8) is a mouse monoclonal antibody specific for an epitope mapping between amino acids 2-25 at the N-terminus of U2AF65 of human origin.

PRODUCT

Each vial contains 200 μg lgM kappa light chain in 1.0 ml of PBS with <0.1% sodium azide and 0.1% gelatin.

Blocking peptide available for competition studies, sc-374333 P, (100 μg peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% stabilizer protein).

APPLICATIONS

U2AF65 (D-8) is recommended for detection of U2AF65 of mouse, rat and human origin by Western Blotting (starting dilution 1:100, dilution range 1:100-1:1000), immunoprecipitation [1-2 μ g per 100-500 μ g of total protein (1 ml of cell lysate)], immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

U2AF65 (D-8) is also recommended for detection of U2AF65 in additional species, including bovine.

Suitable for use as control antibody for U2AF65 siRNA (h): sc-37667, U2AF65 siRNA (m): sc-37668, U2AF65 shRNA Plasmid (h): sc-37667-SH, U2AF65 shRNA Plasmid (m): sc-37668-SH, U2AF65 shRNA (h) Lentiviral Particles: sc-37667-V and U2AF65 shRNA (m) Lentiviral Particles: sc-37668-V.

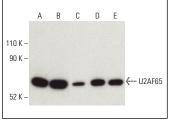
Molecular Weight of U2AF65: 65 kDa.

Positive Controls: SK-N-SH cell lysate: sc-2410, Jurkat whole cell lysate: sc-2204 or Hep G2 cell lysate: sc-2227.

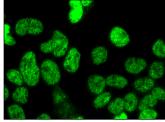
RECOMMENDED SUPPORT REAGENTS

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-lgG κ BP-HRP: sc-516102 or m-lgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunoprecipitation: use Protein L-Agarose: sc-2336 (0.5 ml agarose/2.0 ml). 3) Immunofluorescence: use m-lgG κ BP-FITC: sc-516140 or m-lgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

DATA



U2AF65 (D-8): sc-374333. Western blot analysis of U2AF65 expression in IMR-32 (A), Jurkat (B), Hep G2 (C), HL-60 (D) and SK-N-SH (E) whole cell lysates. Detection reagent used: m-lqGk BP-HRP: sc-516102.



U2AF65 (D-8): sc-374333. Immunofluorescence staining of formalin-fixed Hep G2 cells showing nuclear localization.

STORAGE

Store at 4° C, **DO NOT FREEZE**. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

RESEARCH USE

For research use only, not for use in diagnostic procedures.