Technical Data Sheet

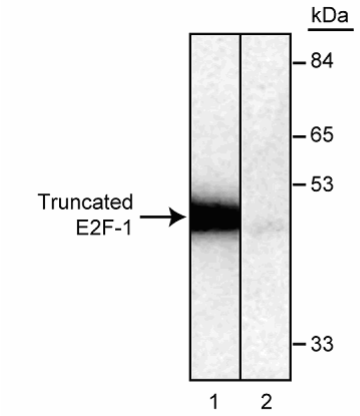
Purified Mouse Anti-Human E2F-1

Product Information

Material Number:	**554213**
Size:	0.1 mg
Concentration:	0.5 mg/ml
Clone:	KH95/E2F
Isotype:	Mouse IgG2a
Reactivity:	QC Testing: Human Tested in Development: Mouse, Rat
Target MW:	60 kDa
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The transcription factor E2F was originally characterized as a sequence-specific DNA-binding factor bound to the adenovirus E2A promoter. E2F-binding sites have now been identified in cellular promoters for genes involved in growth regulation, including c-myc, N-myc, Cdc2 and cyclin A. They have also been identified in promoters for genes whose products are required for DNA synthesis and replication, such as DNA polymerase a, thymidine kinase, thymidylate synthase, and dihydrofolate reductase. When bound to DNA, E2F has been found in complexes with key regulators of cell proliferation including the retinoblastoma protein (Rb) and the Rb related proteins, p107 and p130. Like a variety of other transcription factors, E2F binds to DNA as a heterodimer. The protein E2F-1 has E2F-like properties, it mediates E2F-dependent trans-activation and binds to underphosphorylated Rb. E2F dimerizes with DP-1, a protein which also has E2F-like properties. This dimerization leads to increased DNA binding activity and enhanced transcriptional activity. It is thought that E2F-1/DP association forms an active "E2F" transcription factor. E2F-1 migrates at a reduced molecular weight of ~60 kDa. Clone KH95/E2F (also referred to as KH95) recognizes an epitope between amino acids 342 and 386 of human E2F-1. It cross reacts with mouse and rat E2F-1. Full length, bacterially expressed, recombinant histidine-tagged human E2F-1 (amino acids 1-437) was purified and used as immunogen. Positive tissue culture supernatants were identified by immunoprecipitation of [35S]methionine-labeled, in vitro synthesized E2F-1 protein.



Detection of in vitro translated, [35S] methionine-labeled, truncated E2F-1 protein by immunoprecipitation using KH95/E2F (Cat. No. 554213). *Proteins were separated by SDS-PAGE and visualized by autoradiography. Lane 1, in vitro translated E2F-1. Lane 2, a negative control not containing E2F-1.*

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.
Store undiluted at 4°C.

Application Notes

Application

Western blot	Routinely Tested
Immunoprecipitation	Reported

Recommended Assay Procedure:

Applications include western blot analysis (dilute antibody 1-2 µg/ml). MOLT-4 leukemia cells (ATCC CRL-1582), Daudi lymphoma cells (ATCC CCL-213), and RS4; 11 leukemia cells (ATCC CRL-1873) are suggested as positive controls. Other applications include immunoprecipitation (1-2 µg/1x10^6 cells). For western blot, detailed protocol is available at:

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Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
3. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.

References

Dyson N, Dembski M, Fattaey A, Ngwu C, Ewen M, Helin K. Analysis of p107-associated proteins: p107 associates with a form of E2F that differs from pRB-associated E2F-1. *J Virol.* 1993; 67(12):7641-7647. (Clone-specific: Western blot)

Helin K, Wu CL, Fattaey AR, et al. Heterodimerization of the transcription factors E2F-1 and DP-1 leads to cooperative trans-activation. *Genes Dev.* 1993; 7(10):1850-1861. (Clone-specific: Immunoprecipitation, Western blot)

Huber HE, Edwards G, Goodhart PJ, et al. Transcription factor E2F binds DNA as a heterodimer. *Proc Natl Acad Sci U S A.* 1993; 90(8):3525-3529. (Biology)

Nevins JR. E2F: a link between the Rb tumor suppressor protein and viral oncoproteins. *Science.* 1992; 258(5081):424-429. (Biology)

Saito M, Helin K, Valentine MB, et al. Amplification of the E2F1 transcription factor gene in the HEL erythroleukemia cell line. *Genomics.* 1995; 25(1):130-138. (Clone-specific: Immunoprecipitation, Western blot)