Anti-Human CEBPE, monoclonal

Alternate Names: CCAAT/enhancer-binding protein epsilon

Cat. No. 14-104

FOR RESEARCH USE ONLY NOT FOR USE IN HUMANS

PRODUCT DESCRIPTION

UniProt Summary

UniProt
Primary Accession: Q15744

C/EBP are DNA-binding proteins that recognize two different motifs: the CCAAT homology common to many promoters and the enhanced core homology common to many enhancers. Binds DNA as a dimer and can form stable heterodimers with C/EBP delta. Nucleus. Strongest expression occurs in promyelocyte and late-myeloblast-like cell lines. Phosphorylated. Belongs to the bZIP family. C/EBP subfamily. Contains 1 bZIP (basic-leucine zipper) domain. Sequence=AAC51130.1; Type=Frameshift; Positions=4; Name=CEBPEbase; Note=CEBPE mutation db; URL="http://bioinf.uta.fi/CEBPEbase/";

Physical Characteristics

Quantity: 1 ml (culture supernatant) or 100ug at 1mg/ml (purified)

Format: culture supernatant or purified material

Host/Isotype: mouse IgG2a

Clonality: monoclonal; ID R536.2.1B2

Formulation: culture supernatant contains 0.02% NaN3. Purified material contains 30% glycerol, PBS and 0.02% NaN3

Specificity: monospecific for human CEBPE ; see “Microarray Analysis” below

Reactivity: human; not tested for cross reactivity in other species

Stability/Storage: 12 months long term: -20°C; short term: 4°C; avoid freeze-thaw cycles; aliquot as required

Handling Notes: small volumes of antibody may occasionally become entrapped in the seal of the product vial during shipment and storage; briefly centrifuge the vial on a tabletop centrifuge to dislodge any liquid in the container cap

Tested Research Applications

- **Western Immunoblotting - Not recommended**
- **Immunoprecipitation - PASS by SOP**

Antibody tested as purified IgG. Optimal dilution to be determined by user.

Quality Assurance

Notes:

1. Please refer to the SOP manual (click here) to for detailed explanation of the experimental setup and thresholds used to evaluate the results shown below.
2. Predicted MW of the target protein expressed in the western immunoblotting (WB) and immprecipitation (IP) experiment is **31.02kDa** and the three fusion tags (venus, 3xFLAG, V5) adds another **38.7kDa** to this protein. Therefore the protein is expected to migrate ~**69.72kDa** in a denatured SDS-PAGE gel.
3. See results below for any applicable cautionary notes.

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Lot-specific COA version tracker: v1.0.0

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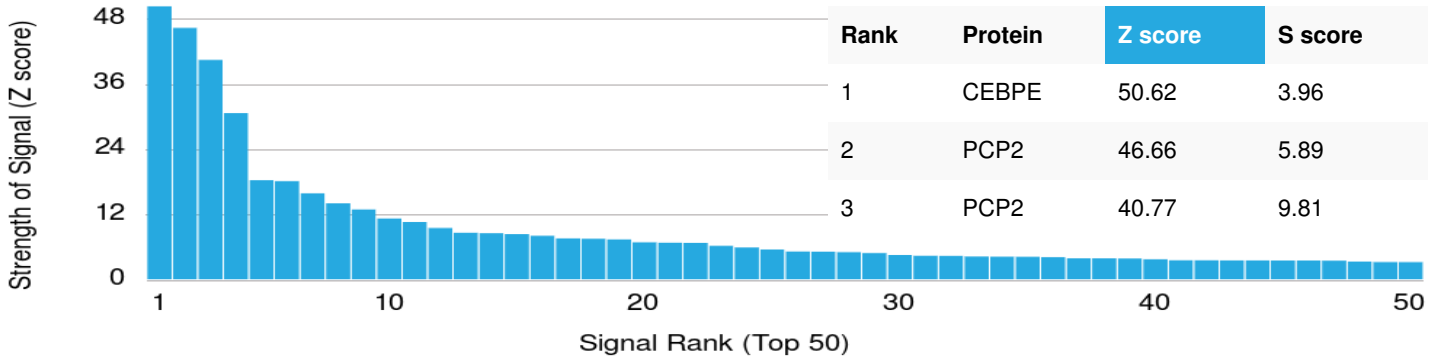
Quality Assurance (HuProt™ Array)



Rank	Protein	Z score	S score
1	CEBPE	50.62	3.96
2	PCP2	46.66	5.89
3	PCP2	40.77	9.81

About Z and S Scores: The z-score represents the strength of a signal that a mAb (in combination with a fluorescently-tagged anti-IgG secondary antibody) produces when binding to a particular protein on the HuProt™ array. Z-scores are described in units of standard deviations (SDs) above the mean value of all signals generated on that array. To be considered a definite binding event between a mAb and a protein target, the z-score must have a value of at least 2.5 (SDs above the mean). The s-score represents the relative target specificity of a mAb, and is the difference (also in units of SDs) between the z-score a mAb generates upon binding to one protein and the next highest z-score that the mAb generates against another protein. For example, if a mAb binds to protein X with an z-score of 43 and to protein Y with an z-score of 14, then the s-score for the binding of that mAb to protein X is equal to 29.

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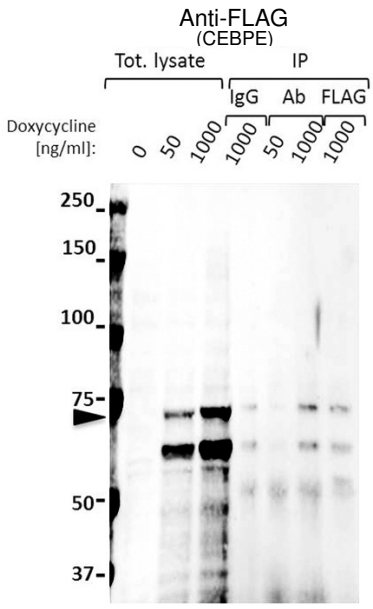
PRODUCT DESCRIPTION

Quality Assurance (continued)

IMMUNOPRECIPITATION - Predicted MW 69.72kDa



Tet-ON HeLa cells were transfected with construct encoding CEBPE (NM_001805.2) with an N-terminal fusion of FLAG, YFP (Venus) and V5 tags under a tet-inducible promoter. These cells were stimulated with 0, 50 or 1000 ng/ml doxycycline. Immunoprecipitation (IP) was carried out using 5µg of either IgG, CDI mAb Anti-CEBPE (cloneID# R536.2.1B2) or 1 µg of FLAG-M2 (Sigma). Immunoblotting was performed using rabbit Anti-FLAG (1:1000, Cell Signaling #2368). We observe that such fusion proteins are generally expressed as a doublet, with the upper band corresponding to the expected size of the CEBPE with an N-terminal fusion of FLAG, YFP (Venus) and V5 tags.

Cautionary notes for IP and WB experiments:

1. Significant signal in IgG lane
2. No or low signal seen in Flag IP lane
3. As per SOP conditions, this antibody performs poorly in IP

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