Human Interleukin-17A (hIL-17A)

Source: Recombinant human IL-17A (hIL-17A) Ile20-Ala155 (Accession #NP_002181) was expressed in human 293 cells at Cell Signaling Technology.

Molecular Characterization: Recombinant hIL-17A contains no "tags" and the nonglycosylated protein has a calculated MW of 15,535. DTT-reduced protein migrates as a 16-24 kDa polypeptide. Heterogeneity in SDS PAGE is due to glycosylation. The non-reduced cystine-linked homodimer migrates as a 28-37 kDa protein. The expected amino-terminal IVKAG of recombinant hIL-17A was verified by amino acid sequencing. The purity of recombinant hIL-17A was determined by SDS-PAGE of 6 μg reduced (+) and non-reduced (-) recombinant hIL-17A. All lots are greater than 97% pure.

Endotoxin: Less than 0.01 ng endotoxin/1 μg hIL-17A.

Purity: >97% as determined by SDS-PAGE of 6 μg reduced (+) and non-reduced (-) recombinant hIL-17A. All lots are greater than 97% pure.

Bioactivity: The bioactivity of recombinant hIL-17A was determined by its ability to induce IL-6 production by primary human fibroblasts. The ED\textsubscript{50} of each lot is between 1.5 - 3.5 ng/ml.

Western blot analysis of extracts from human foreskin fibroblasts untreated or treated with hIL-17A for 15 minutes, using Phospho-p38 MAPK (Thr180/Tyr182) (3D7) Rabbit mAb #9215 (upper) and p38 MAPK Antibody #9212 (lower).

The purity of recombinant hIL-17A was determined by SDS-PAGE of 6 μg reduced (+) and non-reduced (-) recombinant hIL-17A and staining overnight with Coomassie Blue.

Bioactivity: The bioactivity of recombinant hIL-17A was determined by its ability to induce IL-6 production by primary human fibroblasts. The ED\textsubscript{50} of each lot is between 1.5 - 3.5 ng/ml.

Formulation: With carrier: Lyophilized from a 0.22 μm filtered solution of PBS, pH 7.2 containing 20 μg BSA per 1 μg hIL-17A. Carrier free: Lyophilized from a 0.22 μm filtered solution of PBS, pH 7.2.

Reconstitution: With carrier: Add sterile PBS or PBS containing 1% bovine or human serum albumin or 5-10% FBS to a final hIL-17A concentration of greater than 50 μg/ml. Solubilize for 30 minutes at room temperature with occasional gentle vortexing.

Carrier free: Add sterile PBS or PBS containing protein to minimize absorption of hIL-17A to surfaces. Solubilize for 30 minutes at room temperature with occasional gentle vortexing. Stock hIL-17A should be greater than 50 μg/ml.

Storage: Stable in lyophilized state at -20°C for 1 year after receipt. Sterile stock solutions reconstituted with carrier protein are stable at 4°C for 2 months and at -20°C for 6 months. Avoid repeated freeze-thaw cycles.

Maintain sterility. Storage at -20°C should be in a manual defrost freezer.

Applications: Optimal concentration for the desired application should be determined by the user.

Background: IL-17A is a cystine-linked homodimeric pro-inflammatory cytokine produced by TH\textsubscript{17} cells, a distinct CD4+ T cell lineage (1,2). IL-17A stimulates the production of the pro-inflammatory cytokines IL-1β, TNF\textalpha, and IL-6. IL-17A also induces production of the neutrophil chemoattractants IL-8, CXCL1, and CXCL6 thereby bridging adaptive and innate immunity (1,2). IL-17A is intimately involved in mucosal immunity against bacterial infections (1,3) and has a putative role in some autoimmune disorders (1,4). IL-17A effects appear to be exerted primarily through binding to the IL-17RA (5). IL-17A binding induces production of cytokines, chemokines and other proteins through activation of the ERK1/2 MAP kinase, PI3k/Akt, p38, and NFkB pathways (3,4,6). Phosphorylation of some Jaks and Stats has been observed.

Background References: