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HUMANZYME
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HumanKine® IL-17

Effective Tool for Autoimmune Disease Research

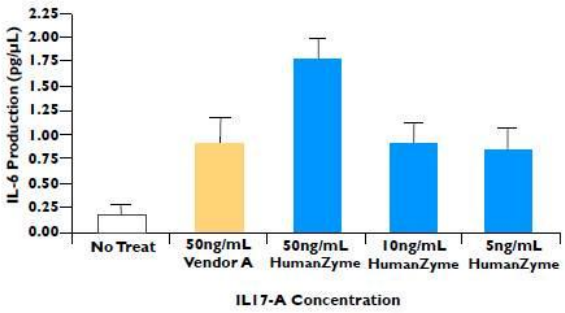
INTRODUCTION

The IL-17 family consists of six members (IL-17A, IL-17B, IL-17C, IL-17D, IL-17E, and IL-17F). Human IL-17A (IL-17) is the original member of the IL-17 family. It was identified in 1995 as a 155 amino acid protein that is a disulfide linked, homodimeric, secreted glycoprotein with a molecular mass of 35 kDa. Each subunit of the homodimer is approximately 15-20 kDa and has been shown to be expressed both glycosylated and non-glycosylated. IL-17A exhibits 72% amino acid identity with HVS13, an open reading frame from a T-lymphotropic Herpesvirus saimiri, and 63% with mouse CTLA8. The Human IL-17A gene and the human IL-17F gene are both located on chromosome 6 and have been found to work independently and synergistically.

Human IL-17A is produced by cells of both adaptive and innate immune systems, including Th17 cells (adaptive), γδ T cells (adaptive/innate), NK cells (innate), NKT cells (innate), neutrophils (innate), and eosinophils (innate). Th17 cells clear extracellular pathogens during infection. They also promote inflammation and have been implicated in the pathogenesis of numerous autoimmune diseases and inflammatory conditions. Activated Th17 cells produce IL-17A, IL-17F, IL-21, and IL-22. IL-17A feeds back and signals the Th17 cell to produce more IL-17A, IL-17F, IL-21, IL-22, GM-CSF, and potentially TNF and IL-6. IL-17 induces the expression of numerous pro-inflammatory cytokines (TNF, IL-1β, and IL-6), hematopoietic growth factors (G-CSF and GM-CSF), chemokines (CXCL1, CXCL8, and CXCL10), and metalloproteinases from many cell types (fibroblasts, endothelial cells, epithelial cells, keratinocytes and macrophages) due to the broad distribution of IL-17 receptors. The IL-17 family has been linked to many immune/autoimmune related diseases including rheumatoid arthritis, asthma, lupus, allograft rejection and anti-tumor immunity. Further research will be required to elucidate the role IL-17A plays in many disease states.

IL-17

Currently, commercially available IL-17(A) proteins are produced from E. coli as a non-glycosylated protein. HumanZyme has produced HumanKine IL-17A which is expressed in engineered human 293 cells as a glycosylated and disulfide-linked homodimer. The bioactivity of IL-17 was determined by the dose-dependent induction of IL-6 secretion from rheumatoid synoviocytes. Briefly, 500,000 cells/well were used in 6 well plates. Cells were starved for 12h with DMEM media without FCS. Cells were then stimulated for 24h in 2 ml of DMEM without FCS. The IL-6 production was analyzed by ELISA on the supernatants. The data indicate that HumanKine IL-17A is significantly more effective with 10-fold higher potency than the E. coli expressed cytokine.



HumanZyme has developed and continues to develop a growing range of tag-free cytokines, including difficult-to-express protein members of the TGFβ superfamily. HumanZyme cytokines are produced to be Xeno-free to address concerns caused by the presence of trace animal components or mammalian pathogens. All HumanKine proteins

are recombinant, animal component-free, and solely from human origin. There are no trace elements introduced as is commonly the case when exotic expression in E. coli, yeast and CHO is employed. Additionally, the internal machinery in human expression systems mean HumanKine cytokines will have bona-fide post-transcriptional modifications, such as phosphorylation and glycosylation, among others. HumanKine cytokines can be used as highly preferred reagents in a wide range of applications for cancer, inflammation, stem cell research, and antibody development.