Atopic Dermatitis Skin Biopsies Have High Numbers of Activated Mast Cells that Are Inhibited by Antolimab (AK002) After Stimulation Ex Vivo
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**BACKGROUND**
- Loss of epithelial barrier integrity is a critical step in the development of atopic dermatitis (AD) whereby the alarmin cytokines IL-33 and TSLP activate inflammatory cells such as mast cells (MCs) (Figure 1).
- While MCs have been shown to be elevated AD, there is need for further characterization of their pathogenic role.

**METHODOLOGY**
- Single-cell suspensions were prepared by enzymatic & mechanical digestion of fresh biopsies from patients clinically diagnosed with AD (n=6) or disease control subjects (n=10).
- Multi-color flow cytometery was performed to quantify immune cells and evaluate the activation state of eosinophils & mast cells as shown in Figure 4.
- Mast cells were FACS-sorted from AD biopsies or non-diseased skin tissues followed by overnight incubaition with or without PMA/albumin.
- Cell-free supernatants were collected the following day and cytokines were quantified using meso scale discovery (MSD) system.

**RESULTS**
- The following cytokines were analyzed: IL-4, IL-5, IL-6, IL-10, IL-13, IL-18, GM-CSF, INFγ, TNFα, CCL2, CCL3, CCL4, and VEGF.

**DISCUSSION**
- • Siglec-8 expression remains high on human skin mast cells independent of disease state
- • Resting mast cells in AD skin tissue display high levels of activation as evidenced by decreased surface markers of activation and cytokine production
- • Mast cells in AD skin biopsies are activated by IL-33/TSLP suggesting they are important target cells for alarmin cytokines released by epithelial cells
- • Treatment with AK002 significantly reduces IL-33/TSLP mast cell activation as evidenced by decreased surface markers of activation and cytokine production

**CONCLUSIONS**
- • Human skin mast cells express the inhibitory receptor Siglec-8, and activation of mast cells via FcεRI is inhibited with antolimab
- • Mast cells are elevated in number and are basally activated in AD biopsies with high levels of surface-bound IgE
- Antolimab inhibits IL-33/TSLP-mediated mast cell activation in AD skin biopsies, suggesting antolimab can broadly inhibit multiple modes of mast cell stimulation including, IgE, IL-33, and TSLP
- Mast cells appear to be important in AD, and targeting mast cells via Siglec-8 with antolimab may represent a novel therapeutic approach to the treatment of AD and other allergic diseases

**REFERENCES**
- Presented at the American Academy of Allergy, Asthma & Immunology (AAAAI), Philadelphia, PA, March 13th