



Supplementary Figure 5. Blockade of GABA_A receptor signaling prevented mucus overproduction. (A) PAS staining of lung sections from OVA-exposed mice with and without picotoxin treatment between P14-P20 during OVA exposure. Picotoxin was given intranasally once per day between P14-P20 (0.25µg/g body weight). Control mice received saline intra-nasally. Mice were analyzed at P21. Arrows point to mucin⁺ cells. Inserts show enlarged images of stained epithelium. Scale bar, 100µm. (B) qPCR assays for *Muc5ac* gene expression in OVA-exposed mice with and without picotoxin treatment. (C) Picotoxin had no worsening effect on airway reactivity. Lung slices were prepared from PBS- and OVA-exposed mice at P21 with and without the treatment of picotoxin. Airway reactivity was measured by the percentage of reduction in the airway lumen size in response to increasing doses of Mch. Mid-sized airways with a baseline luminal area between 14,000µm² and 20,000µm² were selected for imaging using an inverted microscope (DMI6000B; Leica Microsystems, Buffalo Grove, IL, USA). From the acquired images, the airway luminal area was measured using NIH Image J and normalized to the baseline value. For all experiments, N=5 mice for each condition from two independent experiments. n.s., not significant. *P<0.05.