

The PDE4 Inhibitor Tanimilast Restrains the Tissue-Damaging Properties of Human Neutrophils

Supplementary

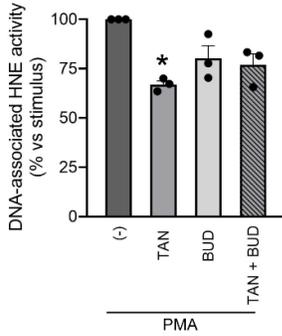


Figure S1. Cotreatment with budesonide does not potentiate tanimilast inhibition of NET production. Isolated neutrophils were left untreated (-) or exposed to PMA (100 ng/ml) in the presence of tanimilast (TAN) at 10^{-7} M or budesonide (BUD) at 10^{-7} M or both (TAN+BUD) at 10^{-7} M. Casted NETs were quantified in cell supernatants by analysing DNA-associated elastase (HNE) activity after a limited DNase I digestion. Results are expressed as fluorimetric readout (FU, Fluorimetric Unit) of elastase activity in the cell supernatants. Dots represent the result of individual donors (N=3). * $p < 0.05$ % One-sample t-test.

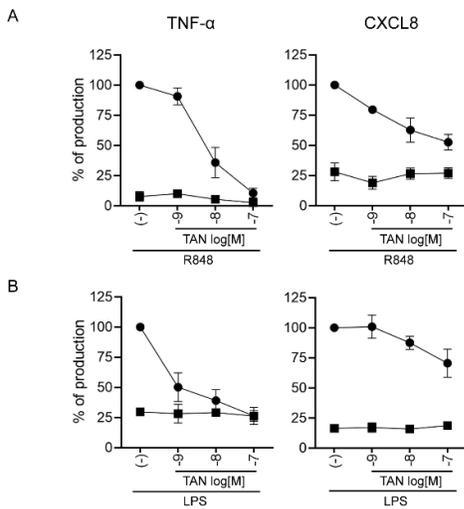


Figure S2. Cotreatment with tanimilast does not increase the already nearly-maximal inhibitory effect of budesonide against cytokine production. Purified neutrophils were incubated with vehicle (-) or tanimilast (TAN) at the indicated concentrations, in the absence (closed circles) or in the presence (closed squares) of budesonide at 10^{-7} M and stimulated for 20 hours with R848 (5 μ M) (A) or LPS (100 ng/ml) (B). Graphs depict the calculated inhibition of the secretion of TNF- α (left panels) and of CXCL8 (right panels). Results are expressed as the mean \pm SEM from three independent donors.