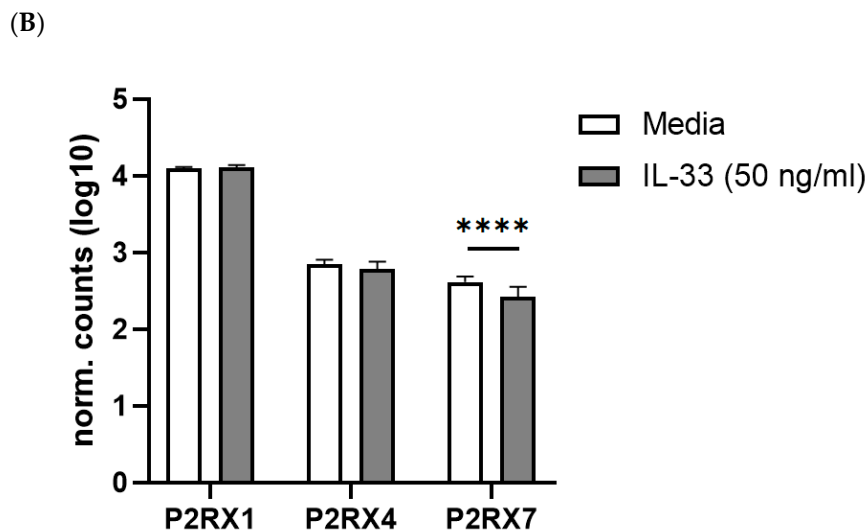
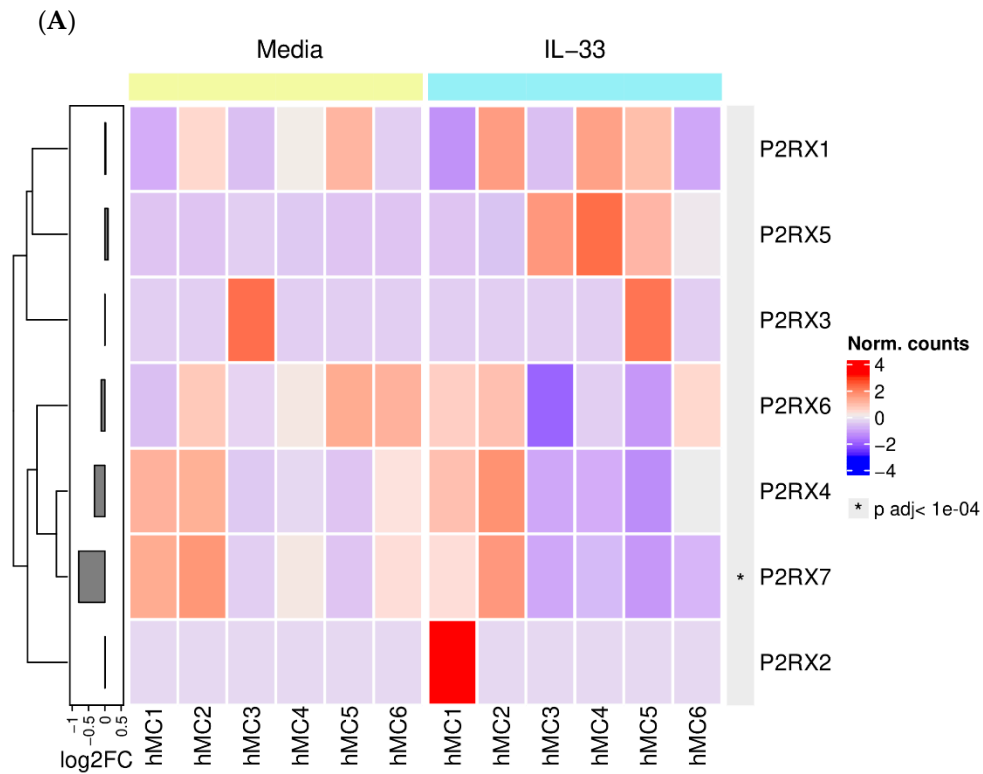


**Figure S1. Example of the gating strategy used when assessing degranulation of blood-derived mature human mast cells.**

(A) The MC population was selected according to size and granularity using forwards and side scatter. Dead cells were excluded using Live/dead discrimination, with single cells selected according to area and width parameters.

(B) Gating strategy showing the example of MCs degranulation based on CD63 externalisation upon IL-33 treatment and ATP or IgE/anti-IgE stimulation.

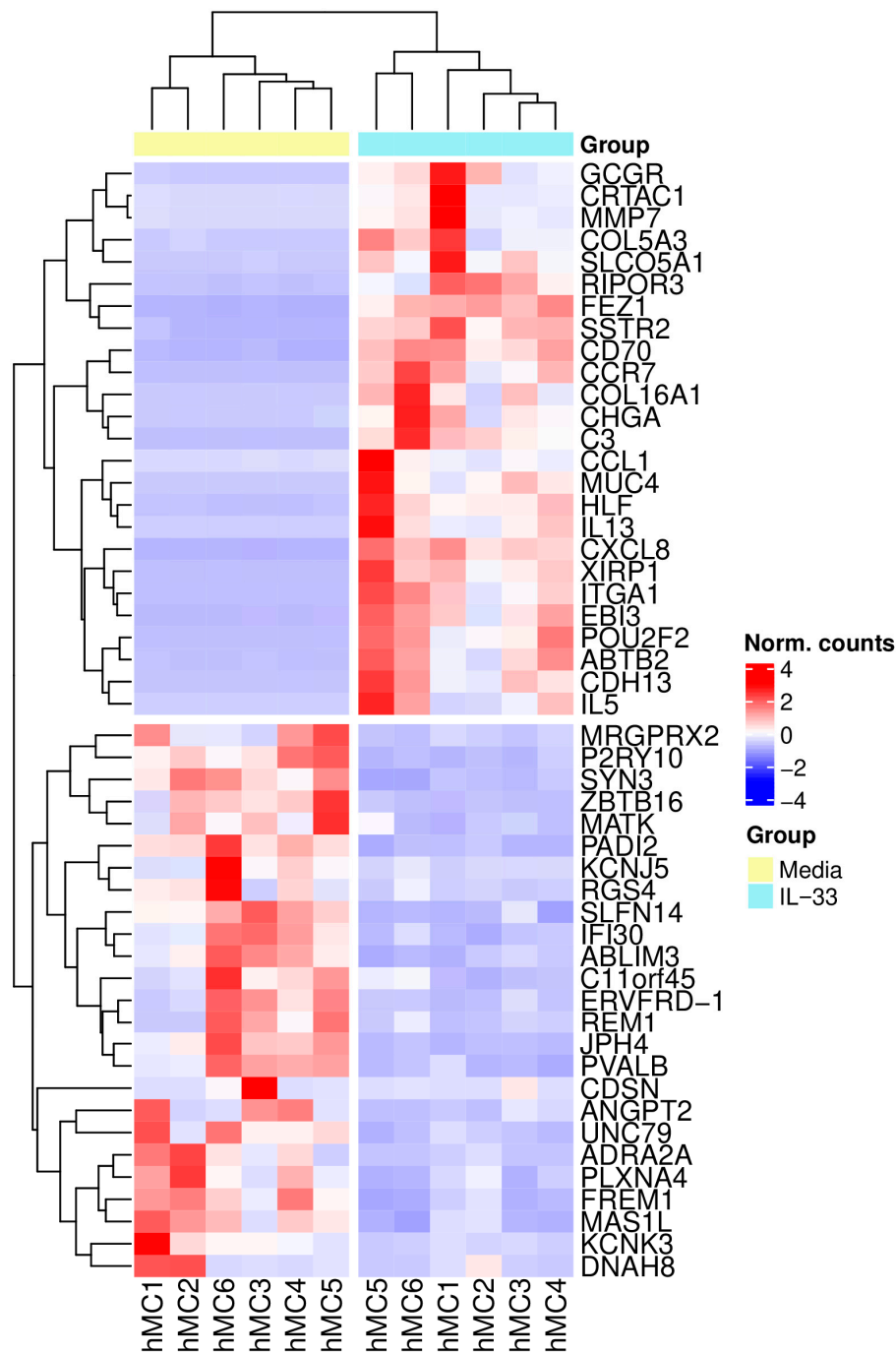


**Figure S2. Changes in P2X receptor gene expression upon IL-33 priming in mast cells.**

(A) The results for n=6 MC donors were visualised as log2fold change compared to media (left) and z-scores of normalized counts of selected P2XR genes were sorted by treatment group and Euclidean distance and plotted using the R ComplexHeatmap package.

(B) Normalized counts of P2X1, P2X4 and P2X7 were calculated using the R DESeq2 package.

Data are mean  $\pm$  SEM of n=3 separate experiments from individual MC cultures. Statistical differences are indicated, \*\*\*\* p<0.0001 (Wald test).



**Figure S3. Mast cell gene expression changes in response to 24-hour IL-33 priming.**

Top high and lowly expressed 50 protein-coding genes ( $\log_2$  fold change of  $\geq 2$  or  $\leq -2$ , adjusted p-value  $< 1e-04$ ) visualised as a z-score values. The results for  $n=6$  MC donors were visualised as a heatmap generated using the R pheatmap package and sorted by Euclidean distance.