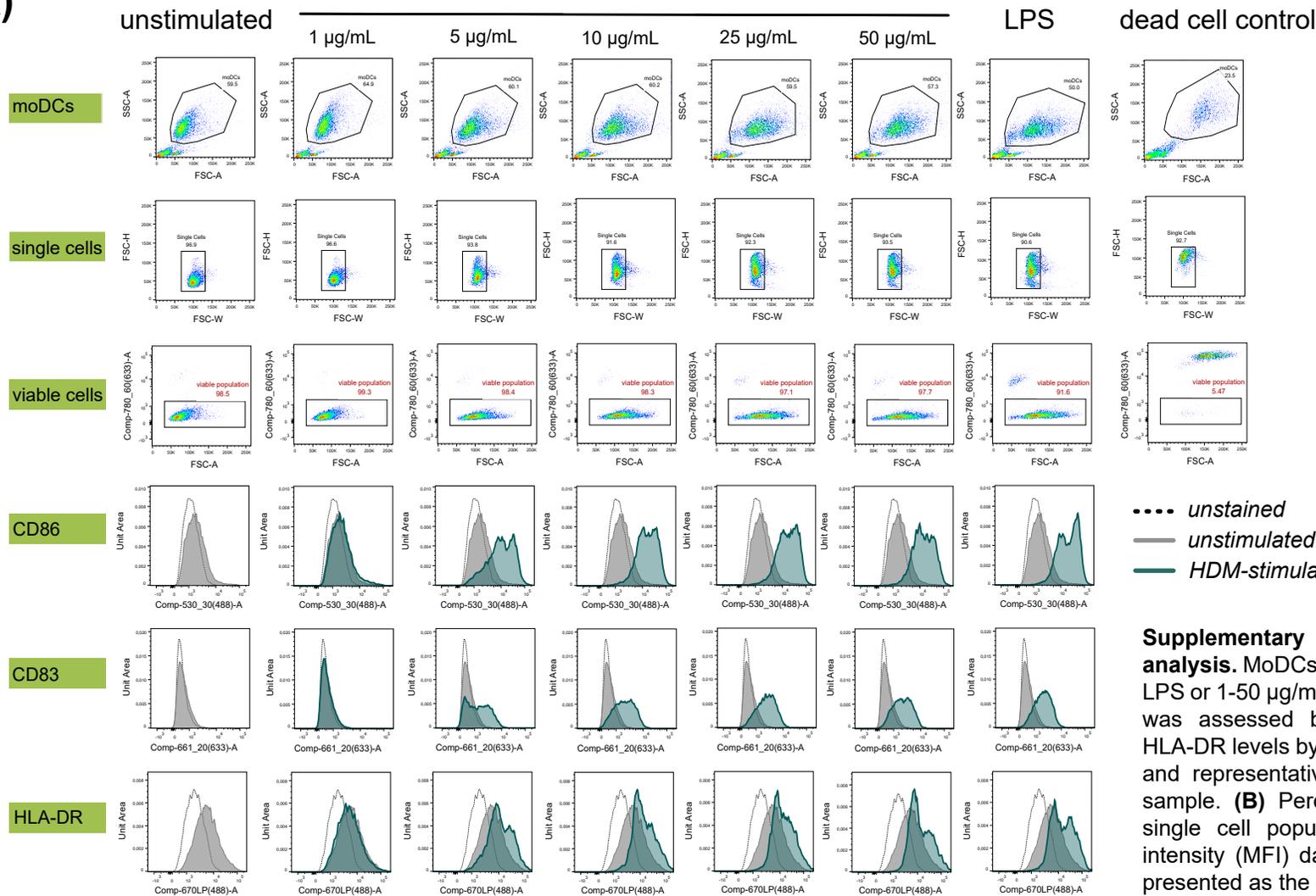


Supplementary Material

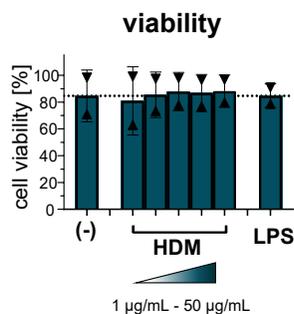
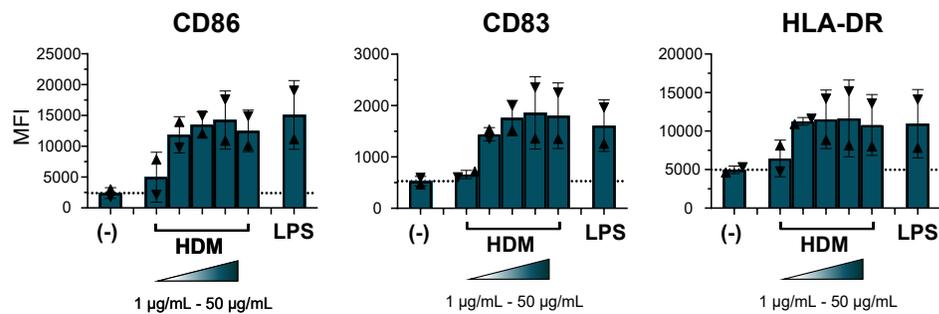
Toll-like receptor 4 and Syk kinase shape dendritic cell-induced immune activation to major house dust mite allergens.

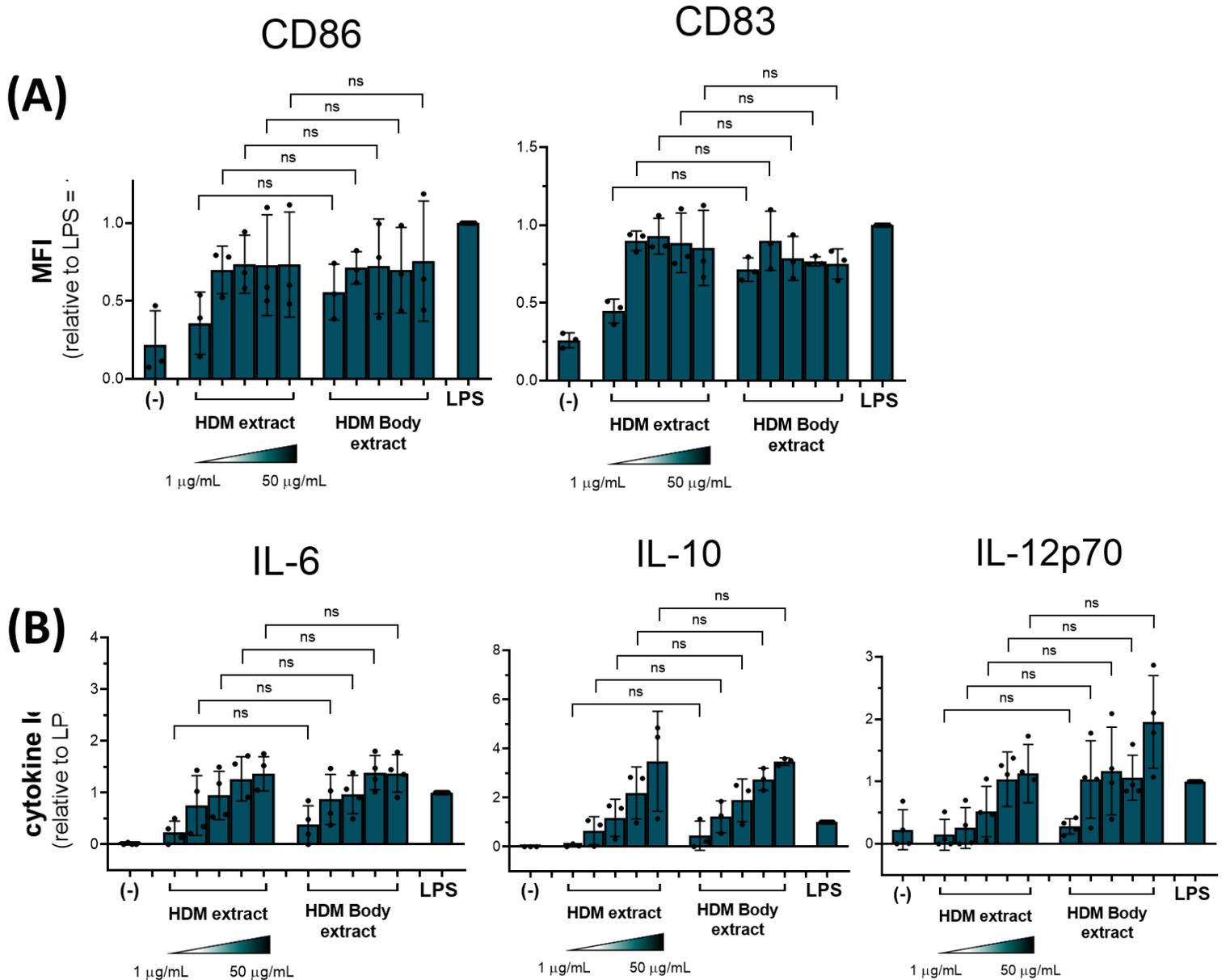
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(A)**HDM extract**

Supplementary Figure 1. MoDC maturation analysis. MoDCs were treated for 24 h with 10 ng/mL LPS or 1-50 $\mu\text{g/mL}$ of HDM extract. MoDC maturation was assessed by measuring CD86, CD83, and HLA-DR levels by flow cytometry. **(A)** Gating strategy and representative histograms of a representative sample. **(B)** Percentage of viable cells within the single cell population and **(C)** mean fluorescent intensity (MFI) data of the samples. The data are presented as the mean \pm SD ($n=2$ donors).

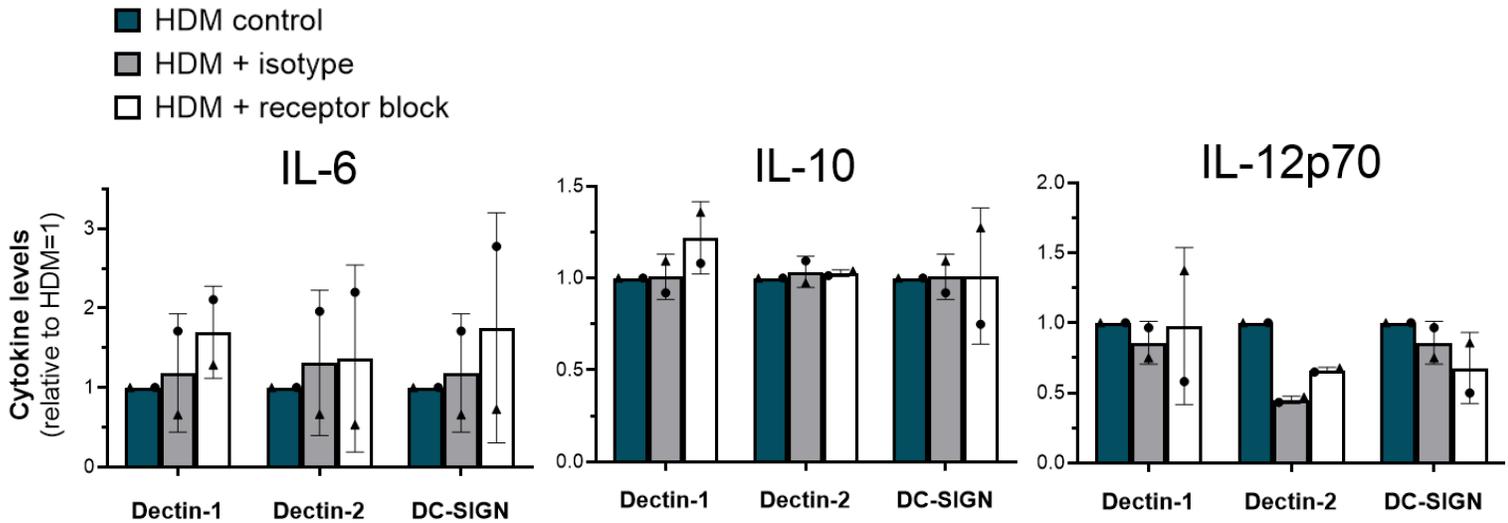
(B)**(C)**



Supplementary Figure 2. HDM whole culture extract and HDM body extract both potently activate moDCs. MoDCs were treated for 24 h with 10 ng/mL LPS or 1-50 μ g/mL of HDM extract derived either from whole mite culture or mite bodies. **(A)** MoDC maturation was assessed by flow cytometry. Mean fluorescent intensity (MFI) data of the samples are shown relative to levels obtained from cells stimulated with 10 ng/mL LPS. **(B)** Culture supernatants were collected and screened for the cytokines IL-6, IL-10, and IL-12p70 by ELISA. The measured data are shown relative to levels obtained from cells stimulated with 10 ng/mL LPS.

The data are presented as the mean \pm SD (n=3-4 donors).

n.s. $p > 0.05$, * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$, **** $p < 0.0001$ relative to unstimulated cells (RM-ANOVA with Tukey's multiple comparison test).



Supplementary Figure 3. The role of CLRs in HDM-induced moDC activation. MoDCs were incubated for 1 h at 37 °C with either Dectin-1-, Dectin-2-, or DC-SIGN-blocking antibody or the respective isotype controls and subsequently exposed for 24 h to 10 ng/mL LPS or 50 µg/mL HDM extract. Culture supernatants were collected and screened for the cytokines IL-6, IL-10, and IL-12p70 by ELISA. The measured data are shown relative to levels obtained from untreated (control group) cells stimulated with HDM extract. The data are presented as the mean ± SD (n=2 donors).