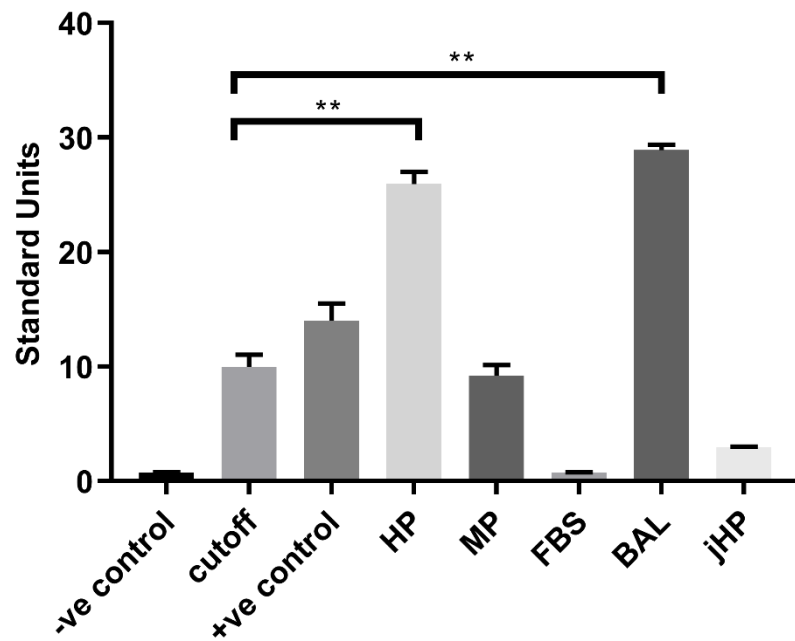


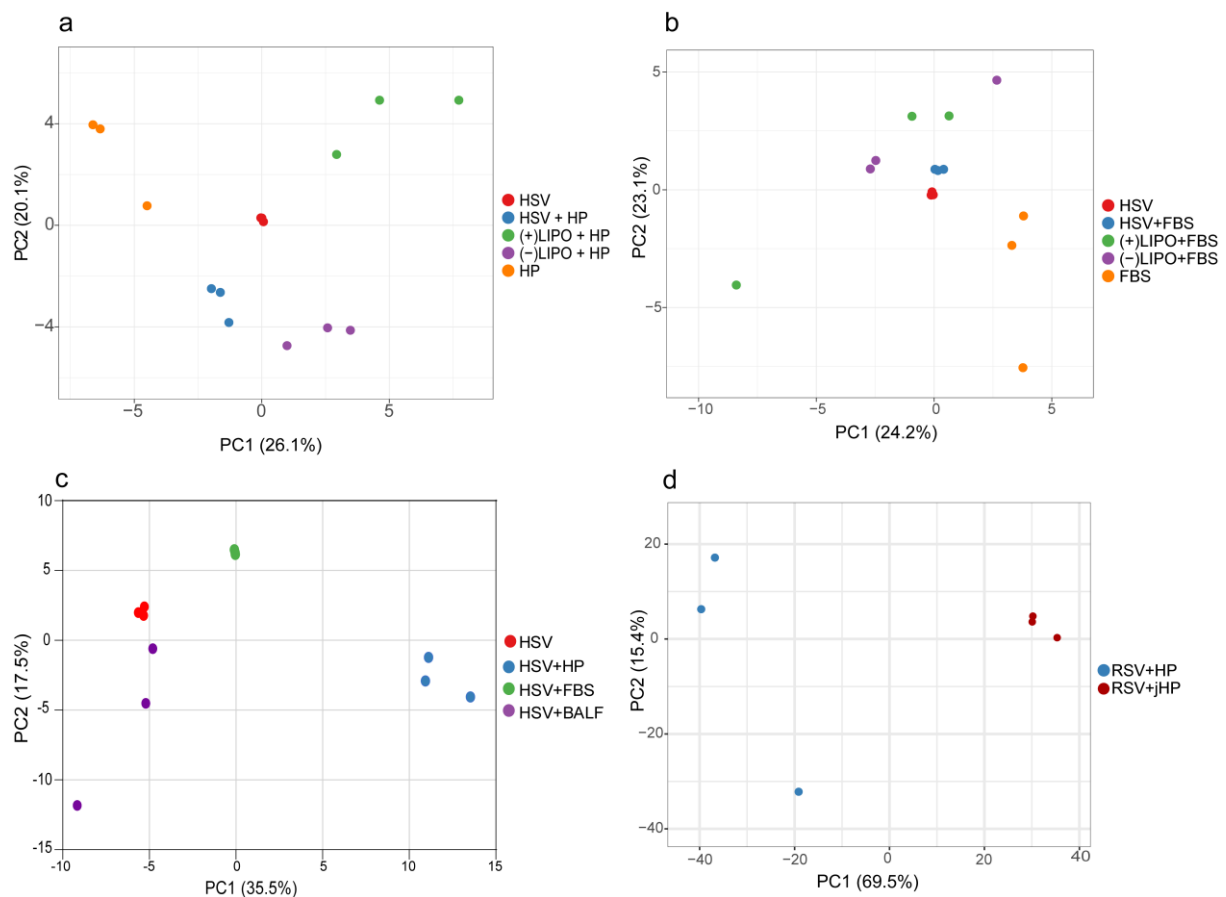
Supplementary Information

The Viral Protein Corona Directs Viral Pathogenesis and Amyloid Aggregation

Ezzat K. et al.

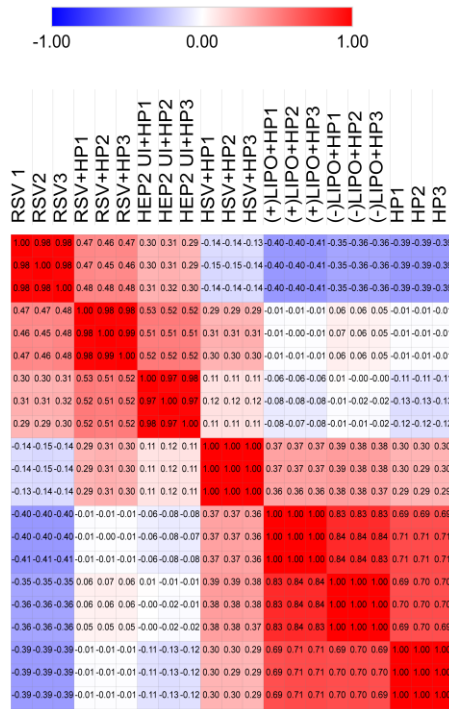


Supplementary Figure 1. ELISA analysis of anti-RSV IgG antibodies in different biological fluids. Different biological fluids were diluted to a protein concentration of 0.3 mg/ml, and then ELISA was used to quantify RSV specific IgG antibodies. Significant differences were assessed by Mann-Whitney test and are indicated by $^{**}P < .01$. Data are shown as means $\pm$ SEM of six replicates from two separate experiments.

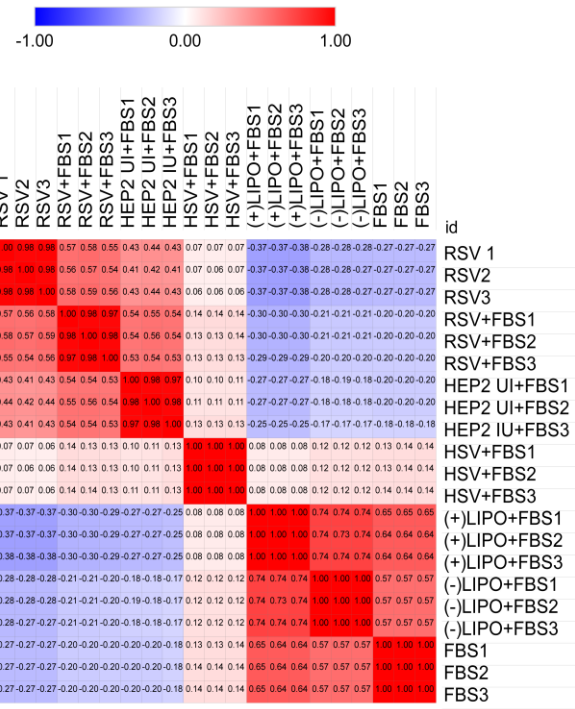


Supplementary Figure 2. Principal component analyses (PCA) of the corona proteomic profiles. Triplicate samples were incubated with 10% solutions of different biological fluids for 1h at 37 °C, then re-harvested, washed and finally analyzed by MS. (-)Lipo = negatively charged lipid vesicles, 200 nm, (+)Lipo = positively charged lipid vesicles, 200 nm. Only proteins significantly detected (FDR 1%) in all three replicates in each condition were used. **(a)** PCA comparing proteomic profiles of HSV-1 and controls in human plasma (HP) **(b)** PCA comparing proteomic profiles of HSV-1 and controls in fetal bovine serum (FBS). **(c)** PCA comparing proteomic profiles of HSV-1 in different biological fluids. **(d)** PCA comparing corona proteomic profiles of RSV in adult human plasma (HP) vs. juvenile human plasma (jHP).

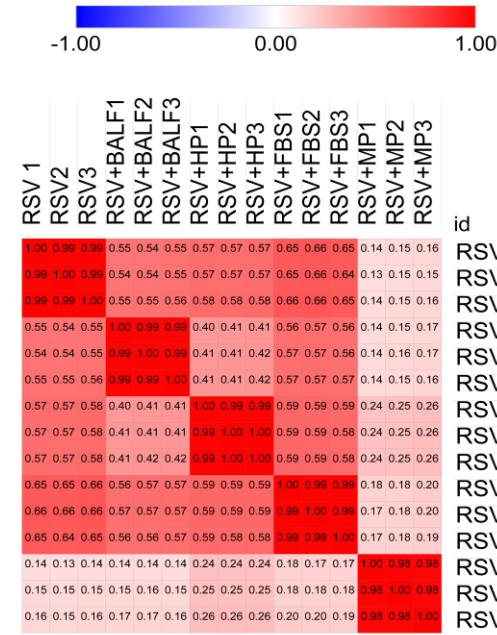
a



b



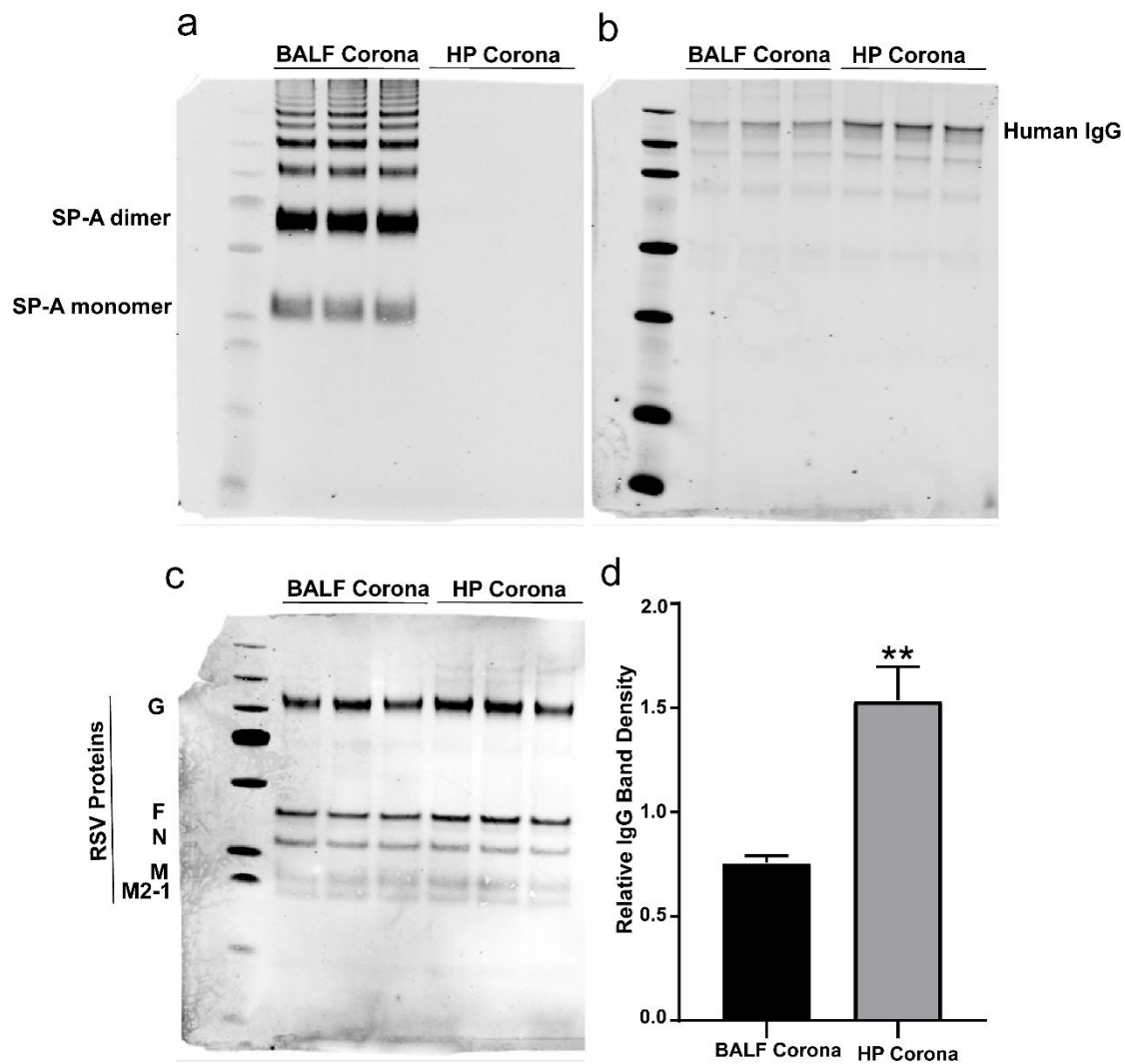
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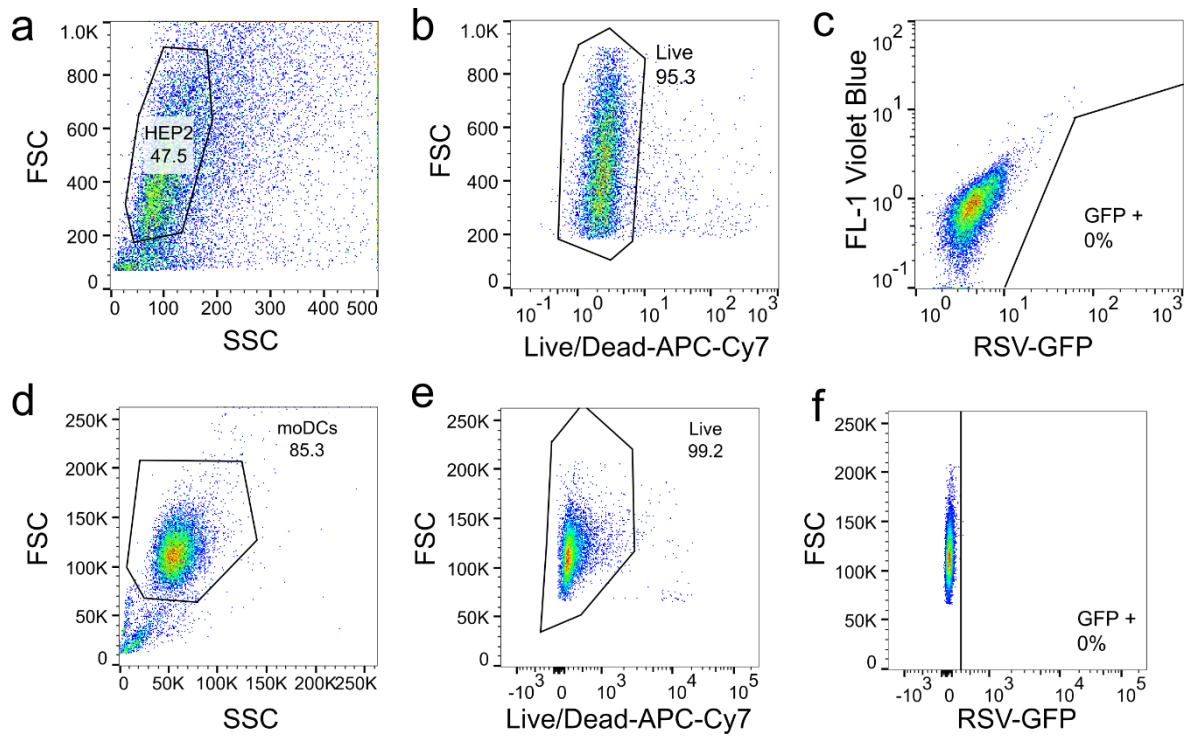
d

MATRIX	CV%
FBS	29
HP	19
(-)Lipo + FBS	42
(-)Lipo + HP	36
(+)Lipo + FBS	41
(+)Lipo + HP	28
HSV+FBS	16
HSV+HP	23
RSV+BALF	33
RSV+MP	31
NI+FBS	31
NI+HP	22
RSV+FBS	25
RSV+HP	26
RSV	33
HSV	19
RSV+jHP	14

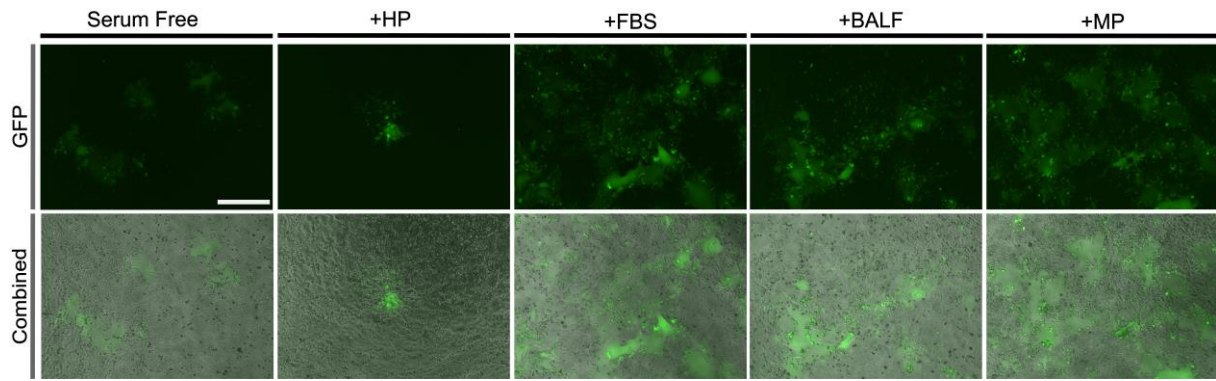
Supplementary Figure 3. Correlation matrices for each of the PCAs presented in Figure 1. (a-c) Spearman correlation coefficients between each of the samples in the PCA based on precursor area of proteins significantly detected (FDR 1%) in all three replicates, **(a)** for samples included in Fig. 1A, **(b)** For samples included in Fig. 1B and **(c)** for samples included in Fig 1C. **(d)** Average coefficients of variation (CV) calculated for replicates in each corona condition based on protein precursor area.



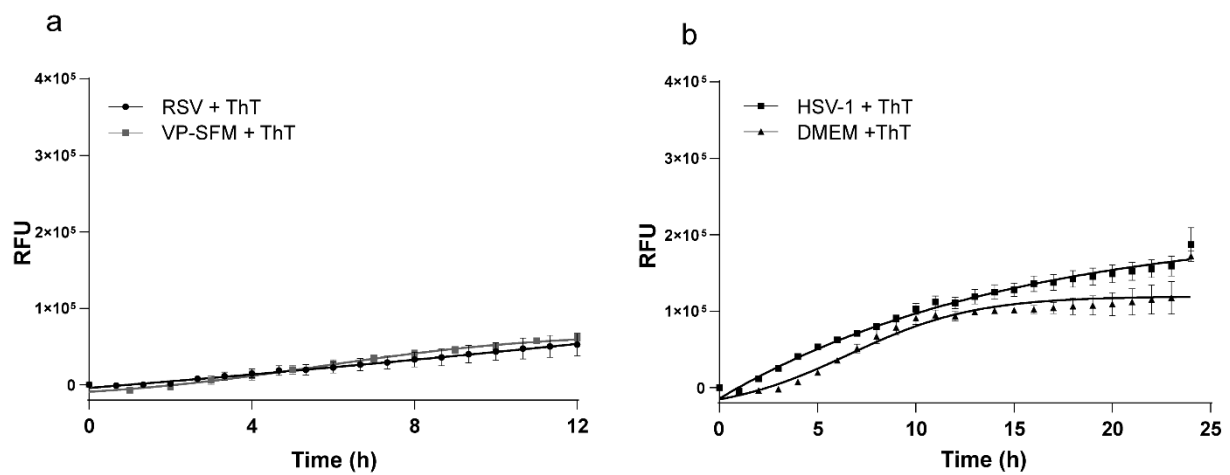
Supplementary Figure 4. Detection of specific corona proteins using western blot. Serum-free produced RSV was incubated with BALF or HP at a protein concentration of 0.1 mg/ml at a ratio of 1:1 v/v for 1h at 37 °C then harvested and washed in a procedure similar to the corona proteomic analysis. **(a)** Bands detected using anti SP-A antibody appear only in the BALF corona samples showing SP-A bands together with higher molecular weight SP-A containing complexes. No SP-A bands were detected in the HP corona samples. **(b)** Bands detected using anti-human IgG antibody appear in both BALF and HP corona, with higher intensity in the latter. **(c)** RSV viral proteins were used as loading controls. **(d)** IgG band intensity was quantified and normalized to the G protein loading control band intensity using ImageJ software. Data is represented as mean relative intensity $\pm$ SEM and significance was calculated using Student's T-test and is indicated by **P < .01.



Supplementary Figure 5. Methodology of FACS gating (a-c) Uninfected HEp-2 cells and (d-f) MoDCs. Cells were first gated based on FSC/SSC (a and d), followed by gating on living cells (b and e) and lastly the GFP gate (c and f). Autofluorescent, non-infected Hep-2 cells were excluded using the FL1 channel.



Supplementary Figure 6. Fluorescence microscopy of RSV infectivity. Representative pictures showing infectivity and syncytia formation in HEp-2 cells using virus stocks in serum-free conditions or with different coronae resulting from pre-incubation with HP, FBS, BALF or MP (n=3). The upper row presents images the fluorescence channel and the lower row represents images of combined fluorescence and phase contrast channels. Bar= 500 μ M.



Supplementary Figure 7. Curves representing the background interaction of ThT with the growth media in absence of peptides (a) for RSV and (b) for HSV-1. Means $\pm$ SEM of six replicates from two separate experiments are shown.