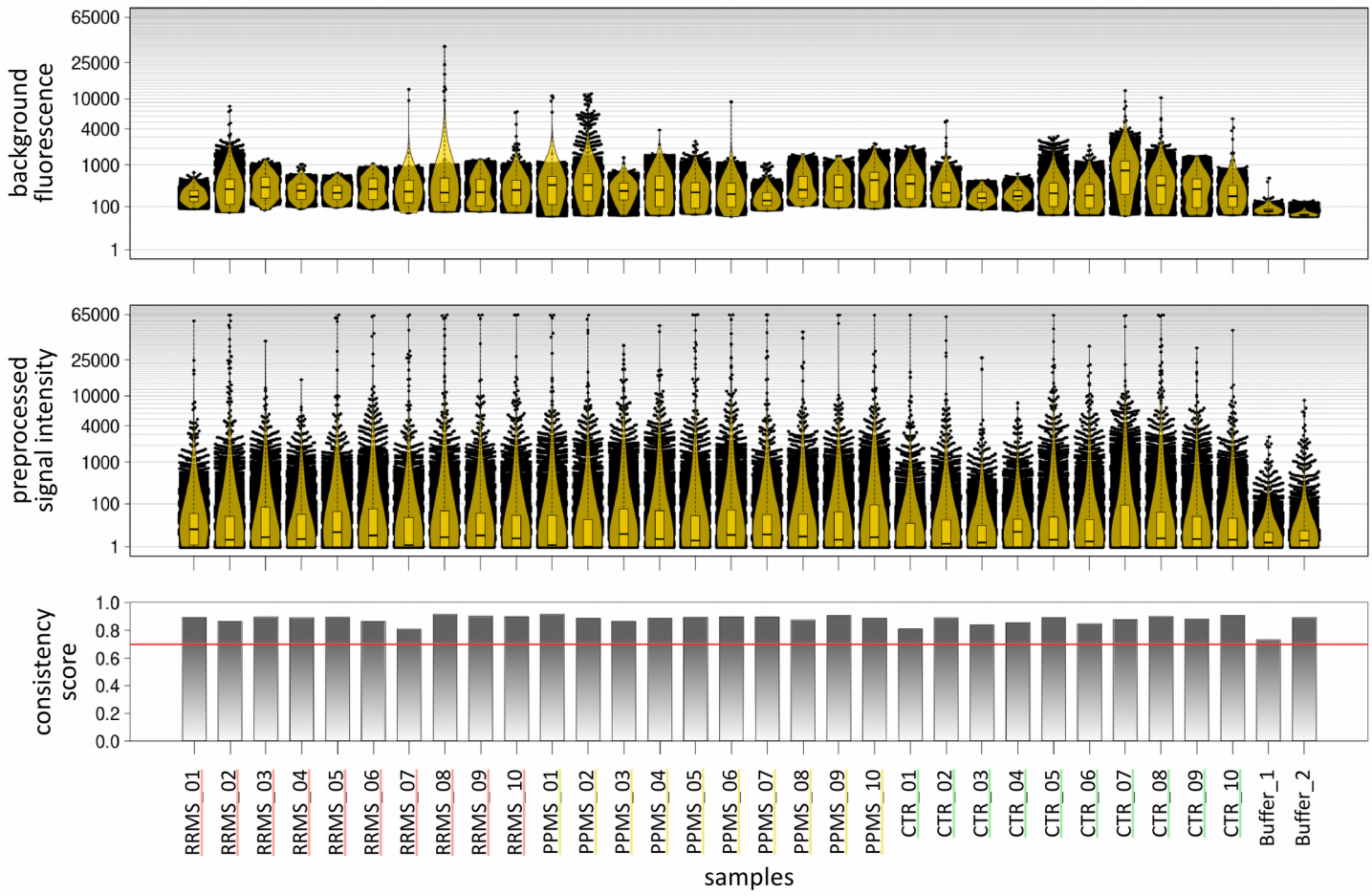
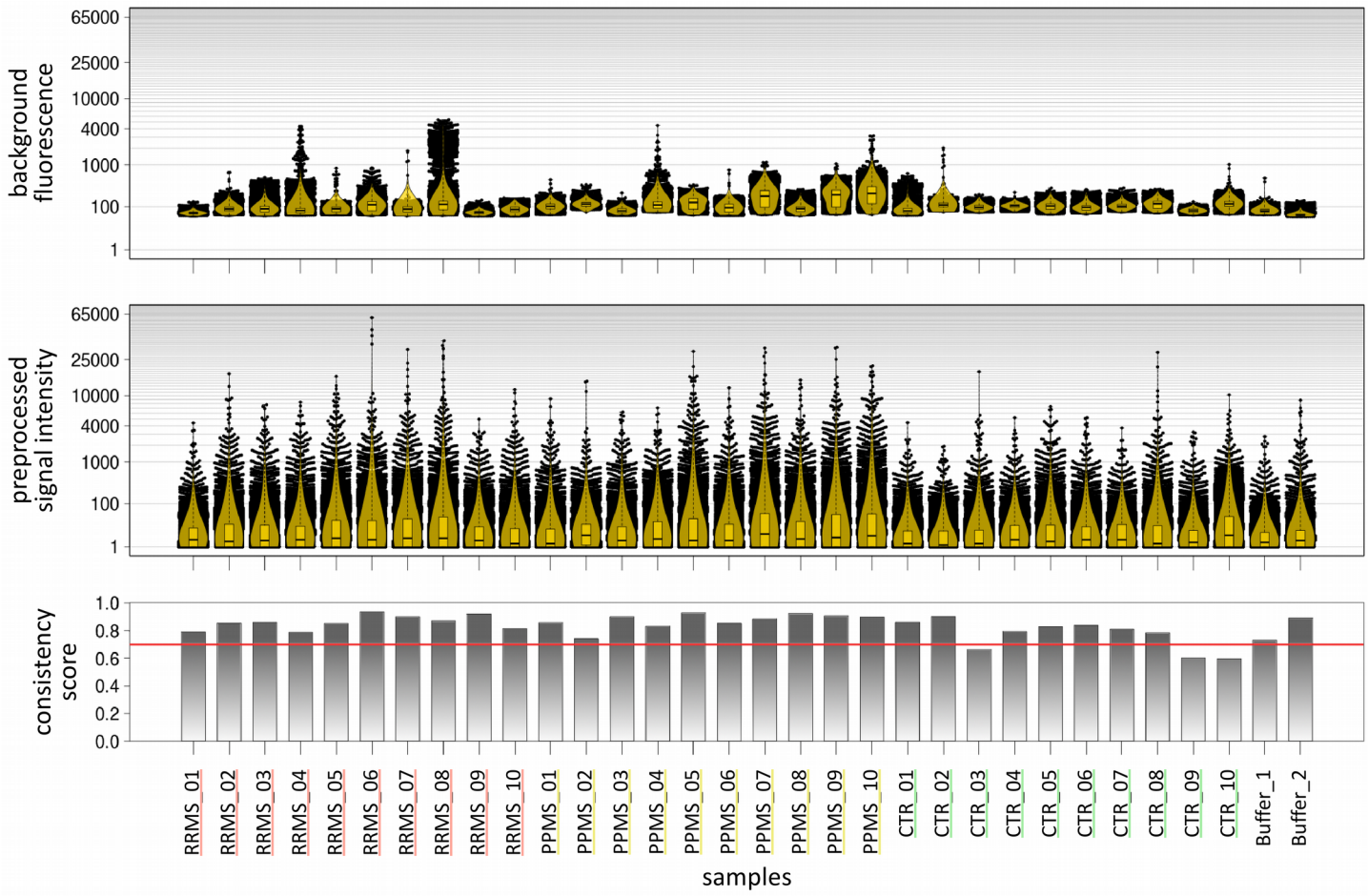
Supplementary File 3: Quality control of the high-density peptide microarray data.

Quality control of the serum data set.



Data distribution and data consistency are shown for the 30 peptide microarrays that were incubated with serum as well as for the two negative control microarrays ("Buffer"). Local background fluorescence intensities of the peptide spots (n=11973 per array) were usually <1000 (top panel). The preprocessed signals (n=3991 data points per array, supplementary file 4) superimposed with boxplots and violin plots are shown in the middle panel. The data distributions were highly skewed, with most signal intensities being <400. Based on the average of correlation coefficients comparing the data for each pairwise combination of the three subarrays, consistency scores were calculated for each peptide microarray according to Hecker *et al*. (Autoimmun Rev. 2012). This score was >0.7 (red line) for all arrays in this data set (bottom panel), which indicates good data quality.

Quality control of the CSF data set.



Data distribution and data consistency are presented for the 30 peptide microarrays that were incubated with CSF as well as for the two buffer controls. Overall, low background signals, comparable preprocessed signal intensities across the microarrays and high consistency scores were seen. However, the microarray for the CSF sample of RRMS_08 showed several high background signals (fluorescence >1000 for 496 / 11973 = 4.13% of the data points) due to a smear in one corner of the array (top panel). The preprocessed peptide signals were elevated for patients with a high CSF/serum quotient of total IgG (RRMS_08, PPMS_05, PPMS_07, PPMS_09 and PPMS_10, supplementary file 1), and they were in general lower for the CSF samples than for the serum samples (middle panel). Moreover, 3 peptide microarrays (for CTR_03, CTR_09 and CTR_10) had consistency scores <0.7 (bottom panel), possibly because this quality measure is impaired when only few peptides show high IgG reactivities. Therefore, despite these minor flaws, all microarrays were accepted for the further analysis of the data.