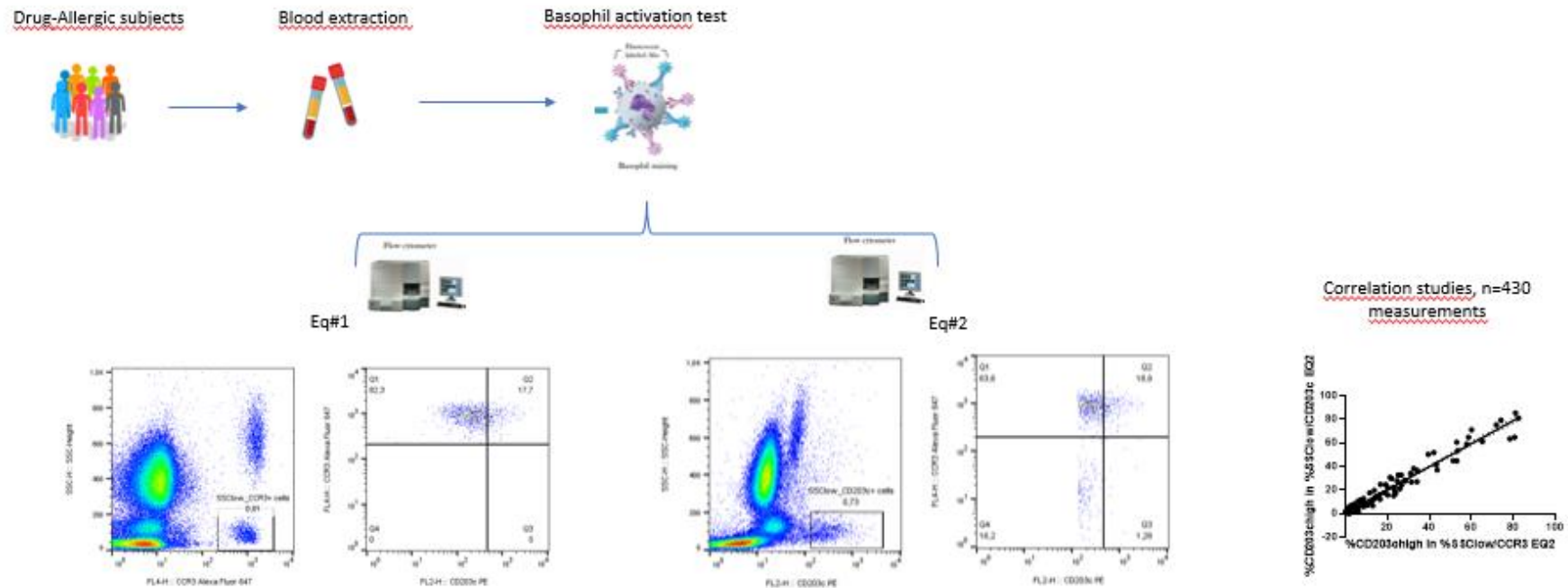
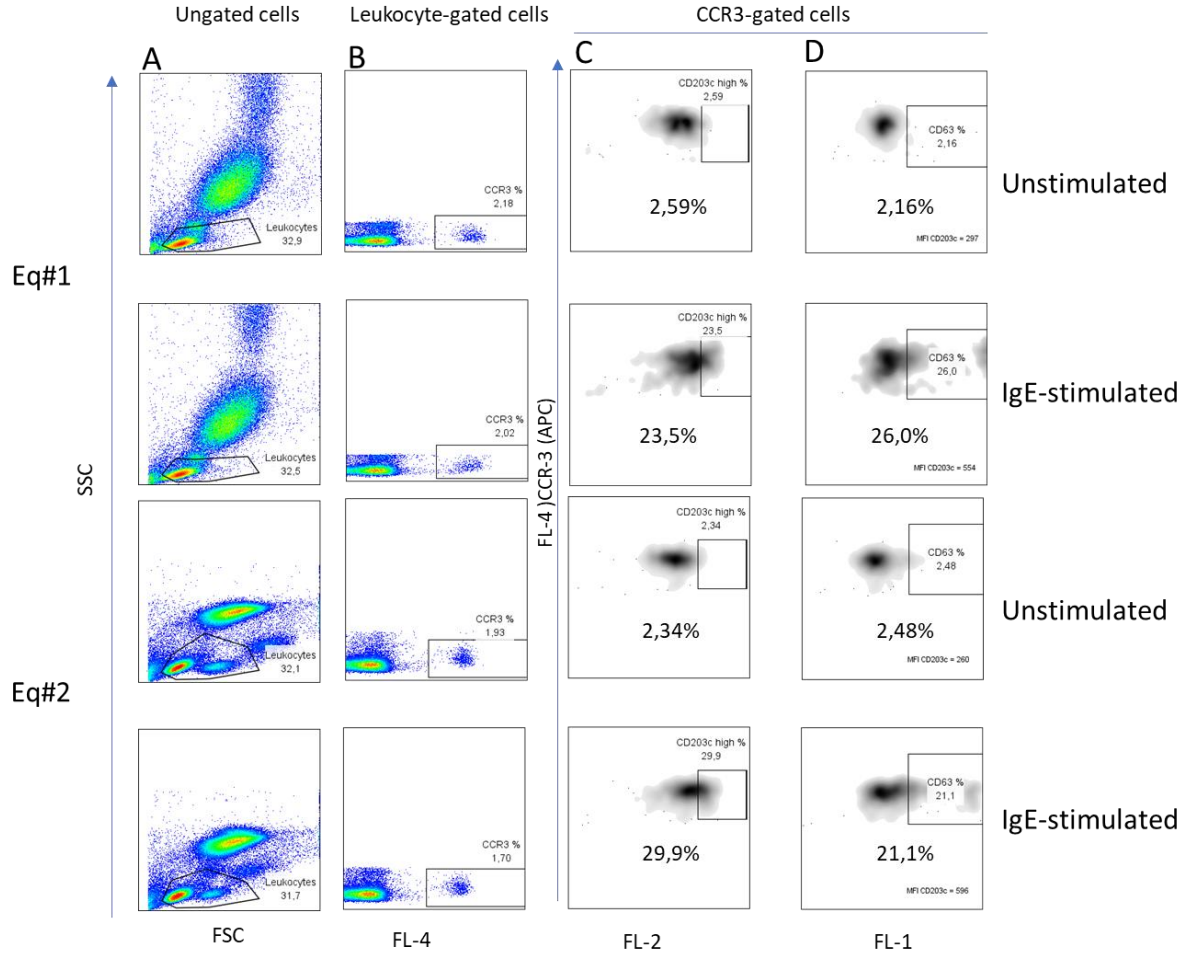


SUPPLEMENTARY MATERIAL

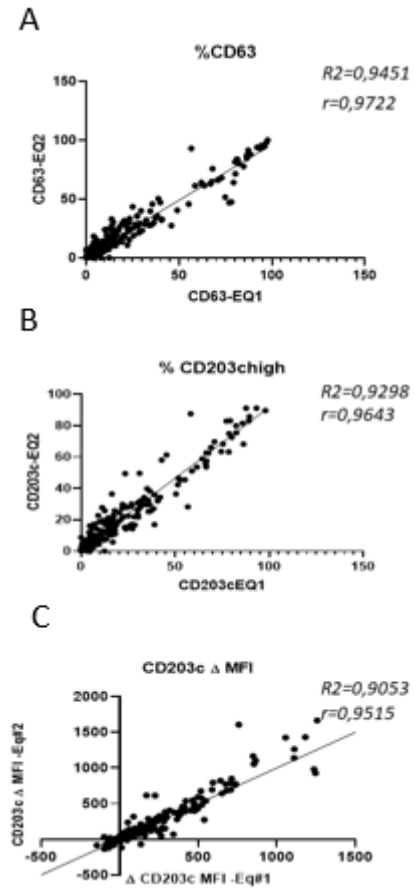
Supplementary Figure1. A. Study design. BAT was performed following SOPs in anonymized clinical samples from patients with confirmed allergy to amoxicillin, clavulanic acid, cefuroxime, ceftriaxone, dipyrone or azithromycin. Two basic flow cytometers were compared (FACS Calibur model 2012: (Eq#1) and FACS Calibur Model 2012: (Eq#2)), totalizing 430 measures. Activated basophils were assessed either by %CD63, CD203c mean fluorescence intensity (MFI) or %CD203chigh and by stimulation index (SI: % marker of stimulated / non stimulated cells). Correlation, concordance, and level of agreement were calculated.



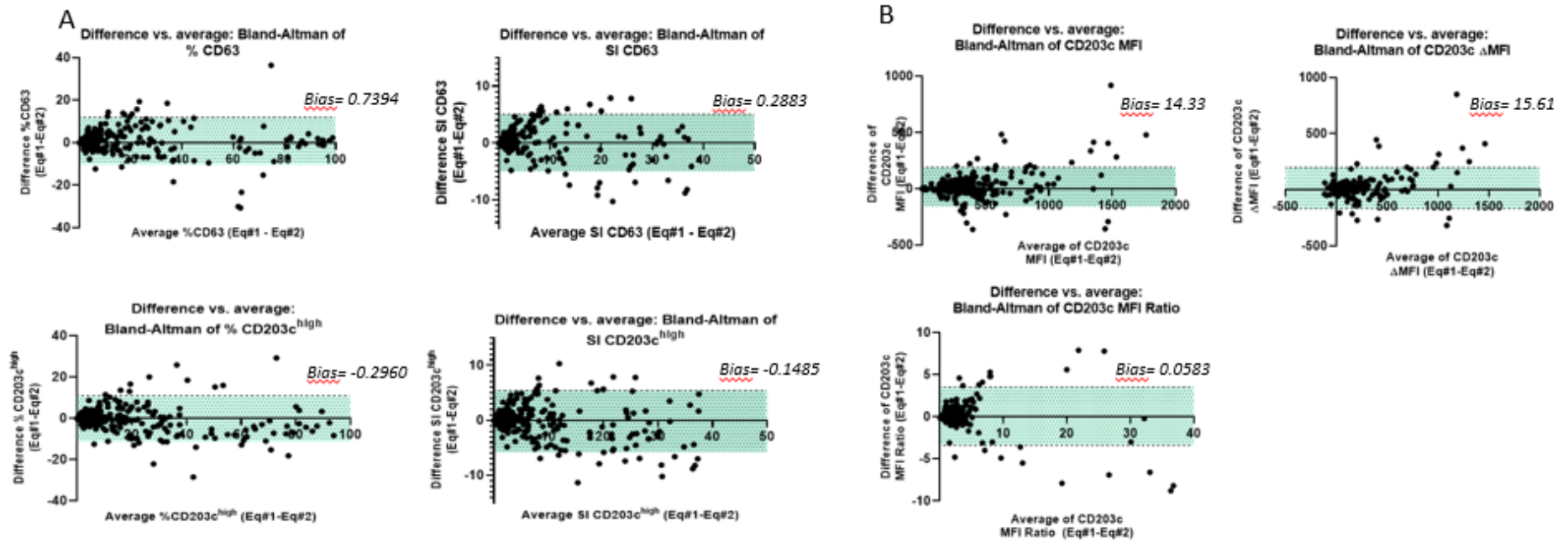
Supplementary Figure 2. Gating strategy for determining basophil activation.



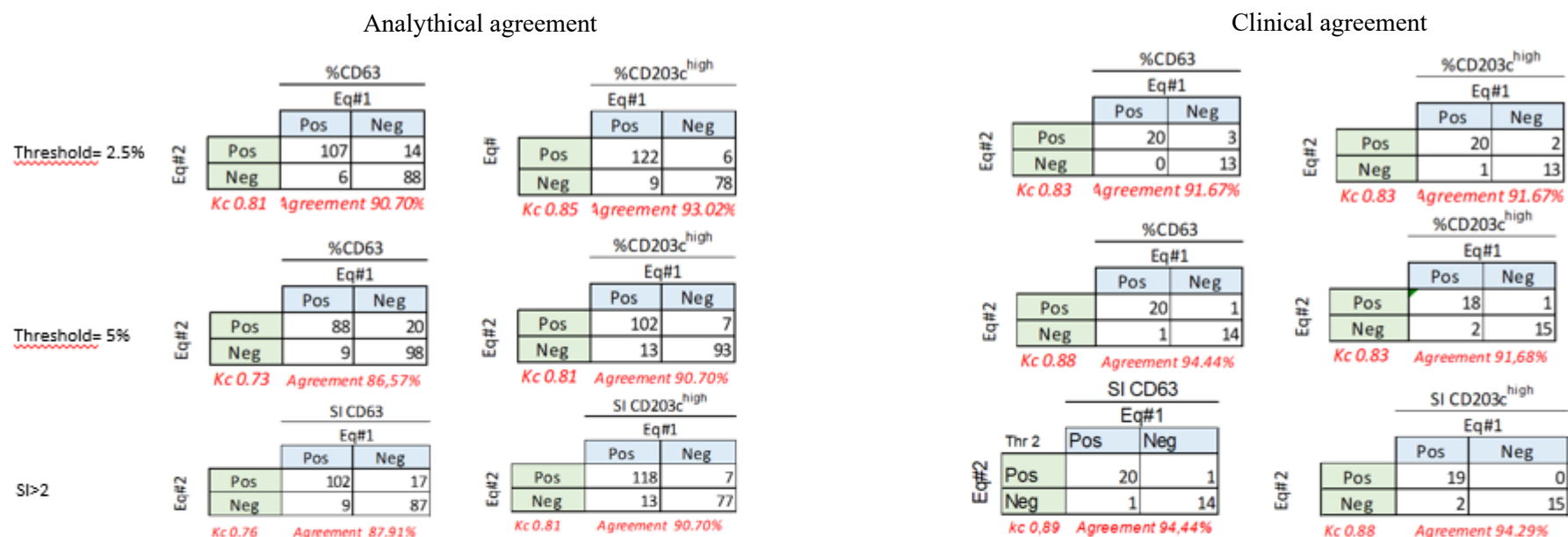
Supplementary Figure 3. Correlation of Eq#1 and Eq#2 with (A) %CD63, (B) %CD230c^{high} and (C) CD203c ΔMFI. r = Pearson's correlation.



Supplementary Figure 4. Analyses of concordance between Eq#1 and Eq#2 using Bland-Altman plots for (A) %CD63, %CD230c^{high} and (B) CD203c MFI (total, delta and MFI ratio). Green lines ± 1.96 times standard deviation, n= 430.



Supplementary Figure 5. Kappa coefficient calculation and agreement for each of the values obtained in each instruments (analytical agreement) or between the final test results (clinical agreement). Comparators: %CD63, % CD203chigh, Stimulation Index (SI). Different thresholds were tested (depicted on the left)



Comparator	kappa (ana)	kappa (clin)	SE kappa (ana)	SE kappa (clin)	CI 95 kappa (ana)	CI 95 kappa (clin)	% Agreement (ana)	% Agreement (clin)
% CD63 (thr 2.5%)	0.81	0.83	0.04	0.094	0.735-0.891	0.644-1.000	90.70	91.67
% CD203high (thr 2.5%)	0.85	0.83	0.036	0.95	0.783-0.925	0.640-1.000	93.02	91.67
% CD63 (thr 5%)	0.76	0.88	0.044	0.079	0.640-0.845	0.732-1.000	86.51	94.44
% CD203high (thr 5%)	0.81	0.83	0.040	0.093	0.736-0.891	0.651-1.000	90.70	91.68
SI CD63 (thr 2.5%)	0.75	0.89	0.044	0.079	0.670-0.845	0.732-1.000	87.91	94.44
SI CD203high (thr 2.5%)	0.81	0.88	0.041	0.077	0.727-0.877	0.738-1.000	90.70	94.29