

Supplemental Material to:

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Novel asymmetrically engineered antibody Fc variant with superior FcγR binding affinity and specificity compared with afucosylated Fc variant

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Supplementary tables

Supplementary Table 1.

Affinity for Fc γ Rs and T_M of C_H2 domain of Fc variants

	FcyRla	FcyRIIa ^{R131}	FcγRIIa ^{H131}	FcyRIIb	FcyRIIIa ^{F158}	FcγRIIIa ^{V158}	-	
Fc variants	K _D (nmol/L)	Κ _D (µmol/L)	K _⊅ (µmol/L)	K _⊅ (µmol/L)	K _⊅ (µmol/L)	K _⊅ (µmol/L)	(°C)	
lgG1	0.23	0.88	0.66	6.0	1.4	0.31	71	
control mAb2	0.25	1.0	0.93	4.2	2.6	0.39	70	
afucosyl mAb	0.43	0.49	0.78	2.6	0.079	0.0069	69	
homo-DLE	0.073	0.34	0.69	0.63	0.0091	0.0031	49	
homo- VLPYLL	0.70	3.1	0.43	7.8	0.041	0.012	69	
asym-mAb1	0.24	0.38	0.19	4.2	0.0012	0.00037	64	

 $T_M = T_M \text{ of } C_H 2 \text{ domain}$

Supplementary Table 2. The percentage of reduction from initial monomer peak area of Fc variants described in Table 1.

		2 weeks		4 weeks			
				1st			
	1st assay (%)	2nd assay (%)	Average (%)	assay (%)	2nd assay (%)	Average (%)	
control mAb1	0.40	0.48	0.44	1.2	1.4	1.3	
hemi-YWA	0.77	0.98	0.88	1.4	1.4	1.4	
hemi-DLE	6.5	6.5	6.5	9.7	9.5	9.6	
homo-YWA	1.0	1.0	1.0	1.8	1.8	1.8	
homo-DLE	8.9	8.8	8.9	15.5	15.7	15.6	
DLE/YWA	1.3	1.3	1.3	2.4	2.5	2.5	

Supplementary figure



Supplementary Figure 1. Sensorgrams of Fc variant binding to FcγRIIIa^{F158}.

SPR sensorgrams for a representative set of Fc variants as described in Table 1 are shown. Solid black line, control mAb1; solid red line, homo-DLE; dotted red line, hemi-DLE; solid blue line, homo-YWA; dotted blue line, hemi-YWA; and dotted green line, DLE/YWA.