Supplemental Figure 1

		HCVpp																	
		subtype 1a												subtype 1b					
		1a09	1a31	1a38	1a53	1a72	1a80	1a116	1a123	1a129	1a142	1a154	1a157	1b09	1b14	1b21	1b34	1b38	1b52	1b58
mAbs	HEPC82	43	52	36	91	36	35	30	38	27	-5	29	-25	28	41	13	28	52	43	-9
	HEPC90	46	50	9	59	66	15	90	25	-5	54	33	-54	27	12	22	25	29	25	-50
	HEPC96	19	-60	-0	68	62	-32	-5	4.9	-18	60	25	4.2	-18	0.4	65	7.7	-28	-20	4.8
	HEPC80	4.9	13	-14	59	57	-27	29	-1	-37	21	32	-10	-1	-8	49	-5	-18	-38	-50
	HEPC46	-5	12	25	7	40	20	29	-3	27	-3	2.2	10	20	20	15	33	22	8.8	11
	HEPC85	25	-3	16	93	20	-8	17	10	2.8	-32	-1	-34	-20	27	-35	-42	-4	29	-31
	HEPC50	27	15	16	22	23	-2	2.3	-0	12	-30	-10	-49	-30	2.2	38	14	5.5	28	-5
	HEPC87	-19	-92	-34	97	10	-80	-35	-62	-46	25	-69	-30	-28	-9	-8	7.4	-46	-41	-2
	HEPC91	-47	4.2	-20	84	-23	22	-43	-13	-21	-1	2.4	1	-9	-28	-26	-14	16	-26	-27
	HEPC97	-71	-7	-46	-6	-40	3.2	-8	1	18	-49	-11	5.9	-1	-39	-26	1.8	-9	-28	-22

% neutralization				
	>75	50-75	25-50	<25

Figure S1. Neutralizing breadth of the 10 mAbs not shown in Figure 2 against a diverse panel of genotype 1a or 1b HCV pseudoparticles (HCVpp). Neutralization patterns for the 5 most broadly-neutralizing mAbs are shown in Figure 2; data for the remaining 10 mAbs are shown here. MAbs marked with blue were isolated from Subject 117 and mAbs marked with green were isolated from Subject 110. Values shown are percent neutralization achieved by 50 µg/mL of mAb. Values are means of two replicate tests.

Supplemental Figure 2

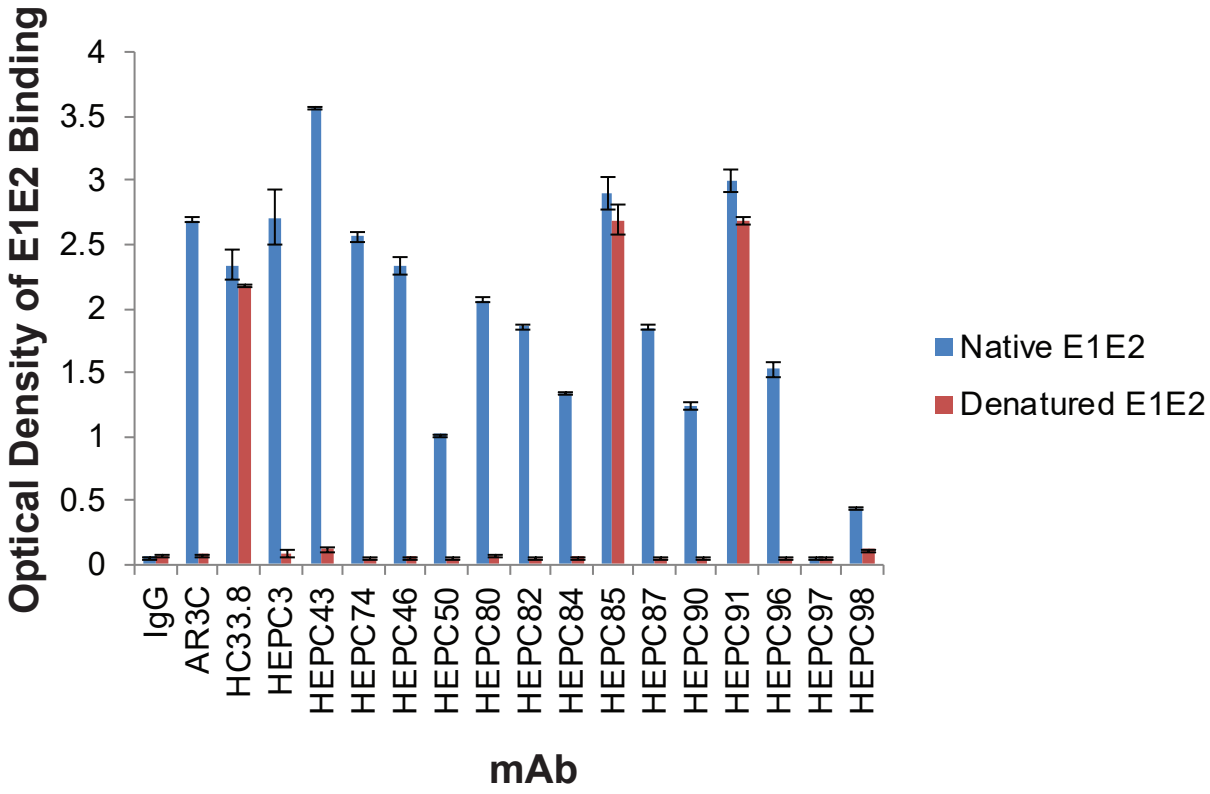
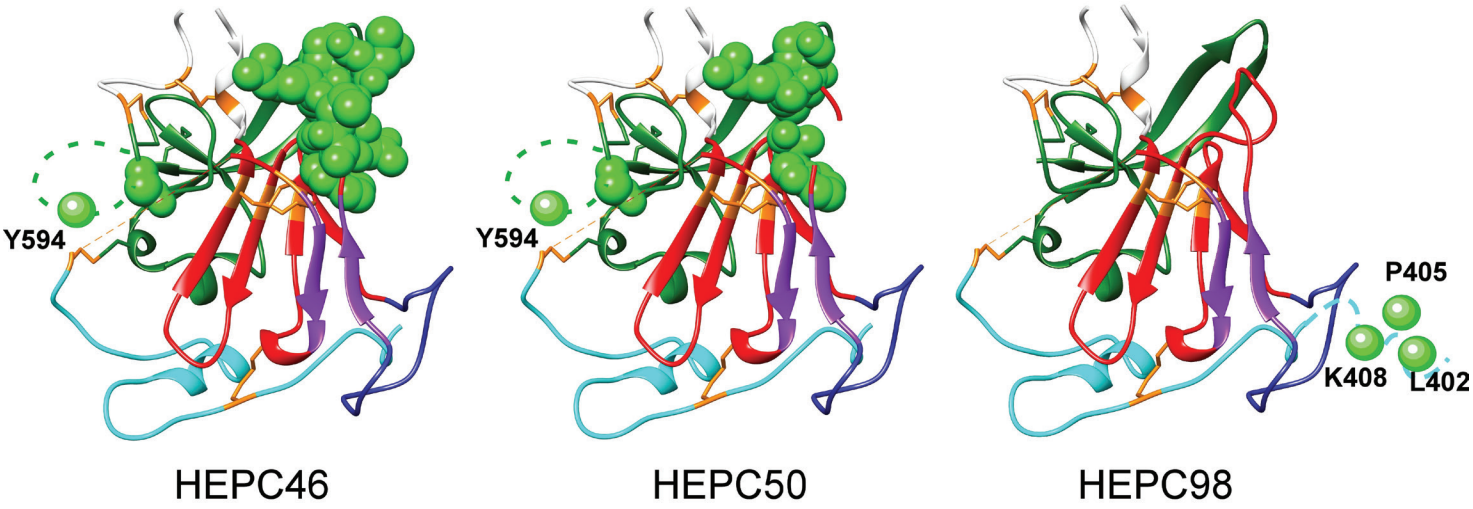


Figure S2. Binding of mAbs to native and denatured E1E2. Binding of 2 µg/mL of each mAb to native E1E2 (clone 1a53) or to the same E1E2 protein after boiling of E1E2 in detergent. Reference mAbs AR3C (conformational epitope) and HC33.8 (linear epitope) are included as controls. Values are the means of two replicate tests, and error bars indicate standard deviations.

Supplemental Figure 3



mAb	Binding residues
HEPC46	N541A, T542A, R543A, P544A, P545A, L546A, N548A, W549A, Y594A, G598A, V633A
HEPC50	R543A, P544A, P545A, W549A, Y594A, C597A, G598A
HEPC98	L402A, P405A, K408A

Figure S3. Binding epitopes of 3 mAbs not shown in Figure 4. Binding residues were identified by measuring relative binding of mAbs to strain H77 E1E2 or alanine scanning mutants spanning the full H77 E1E2 sequence. Critical binding residues are marked with green spheres superimposed on the H77 E2 core structure (31). MAbs not shown here or in Figure 4 did not have adequate affinity for strain H77 E1E2 to be mapped by this method.

Supplemental Figure 4

A

Biotinylated mAbs \ Blocking mAbs	IgG	HEPC3	HEPC43	HEPC74	HEPC87	HEPC85	HEPC91	HEPC82	HEPC84	HEPC90	HEPC50	HEPC46	HEPC80	HEPC98	HEPC96
HEPC3	1.01	0.16	0.30	0.16	0.83	0.98	0.90	0.88	1.01	1.00	0.99	0.94	0.91	1.01	0.96
HEPC43	0.99	0.23	0.21	0.15	1.02	1.00	1.23	0.83	1.01	1.11	0.96	0.92	1.12	0.99	1.19
HEPC74	0.99	0.29	0.43	0.21	0.99	0.99	1.07	0.93	1.01	1.04	0.98	0.97	1.02	0.98	1.04
HEPC87	1.01	0.54	0.34	0.25	0.50	0.33	0.31	0.30	0.59	0.52	0.96	0.98	1.01	0.97	0.97
HEPC85	1.06	1.04	1.09	1.09	0.99	0.33	0.27	1.02	1.03	0.94	1.11	1.03	0.91	1.14	1.00
HEPC91	1.02	1.03	1.01	1.02	1.00	0.56	0.51	1.01	1.01	1.01	1.02	1.00	1.00	1.02	1.01
HEPC82	1.09	0.46	0.30	0.24	0.36	0.27	0.20	0.17	0.62	0.60	1.01	0.96	1.34	1.08	0.86
HEPC84	1.09	1.56	1.36	1.30	0.61	1.55	1.17	0.50	0.24	0.32	1.06	0.62	0.78	1.19	0.82
HEPC90	1.01	1.22	1.18	1.15	0.84	1.22	1.20	0.48	0.29	0.30	1.07	0.74	0.84	1.04	1.02
HEPC50	1.03	0.54	0.56	0.50	0.85	1.21	1.29	0.90	0.87	0.93	0.43	1.10	0.92	1.09	0.80
HEPC46	1.07	0.78	0.66	0.64	0.88	0.92	0.64	0.88	0.84	0.77	1.14	0.37	1.12	1.09	1.06
HEPC80	1.03	0.69	0.56	0.66	1.07	0.85	0.88	1.07	0.72	0.86	1.01	1.05	0.61	1.02	0.88
HEPC98	1.07	0.42	0.93	1.13	0.75	0.08	0.04	0.61	1.09	1.08	1.10	1.03	1.10	0.96	1.03
HEPC96	1.05	0.92	0.89	0.89	1.01	0.91	0.75	0.95	0.92	1.12	0.95	1.12	0.59	1.06	0.73

B

Biotinylated mAbs \ Blocking mAbs	IgG	AR1A	AR2A	AR3A	AR3C	HC84.26	AR4A	AR5A	HC33.8	self
HEPC3	0.79	0.87	0.86	0.3	0.57	0.63	1.03	0.82	0.83	0.22
HEPC43	1.02	0.69	0.91	0.44	0.40	0.72	1.05	0.77	0.63	0.23
HEPC74	1.05	1.06	1.05	0.88	0.65	1.04	1.02	1.06	1.03	0.34
HEPC50	1.02	1.29	1.28	0.45	nd	0.8	0.82	0.91	0.79	0.36
HEPC98	0.94	1.34	1.2	0.51	0.70	1.1	1.09	1.1	0.1	0.65
HEPC46	0.98	0.91	0.93	0.58	nd	1.04	0.75	1.06	0.92	0.47
HEPC82	0.99	1.04	0.84	0.89	0.22	1.02	0.26	0.7	0.58	0.36
HEPC84	1.28	0.99	0.87	1.52	1.11	0.77	0.27	0.66	0.73	0.3
HEPC85	1.02	0.93	0.91	0.77	nd	0.87	0.73	0.84	0.79	0.32

Figure S4. Competition-binding between mAbs. Names of mAbs isolated from Subject 117 or Subject 110 are marked in blue or green, respectively. Binding of 2 µg/mL of the mAbs on the Y-axis ("Biotinylated mAbs") to strain 1a53 E1E2 was measured in the presence or absence of the mAbs on the X-axis ("Blocking mAbs") at a concentration of 20 µg/mL. Values shown are binding of the biotinylated mAb in the presence of blocking mAb, relative to binding in the absence of blocking mAb. Combinations resulting in relative binding <0.7 or <0.3 are marked in yellow or red, respectively. **(A)** Competition-binding between novel mAbs and each other. **(B)** Competition-binding between novel mAbs and a panel of previously published anti-HCV bNAbs.

Supplemental Figure 5

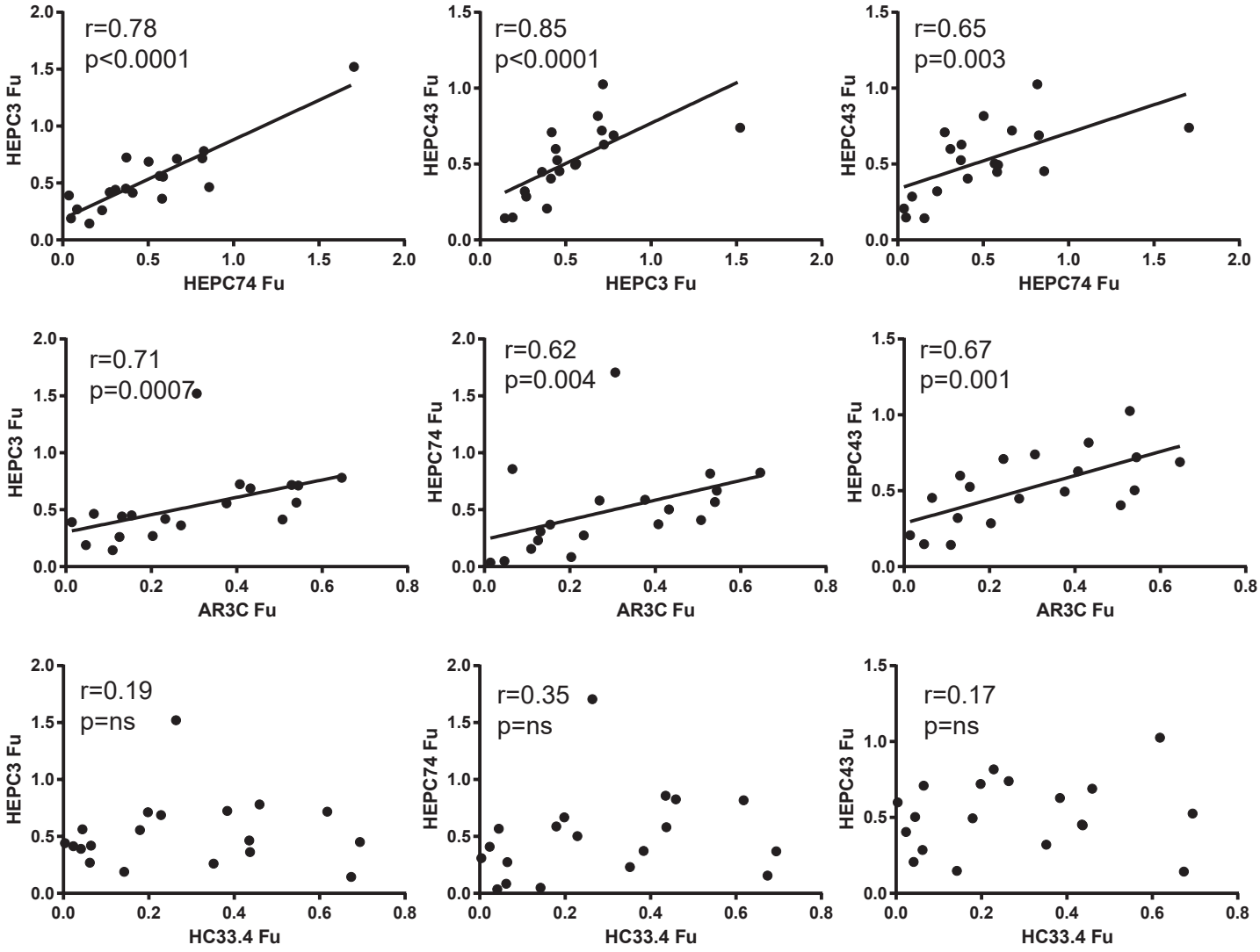
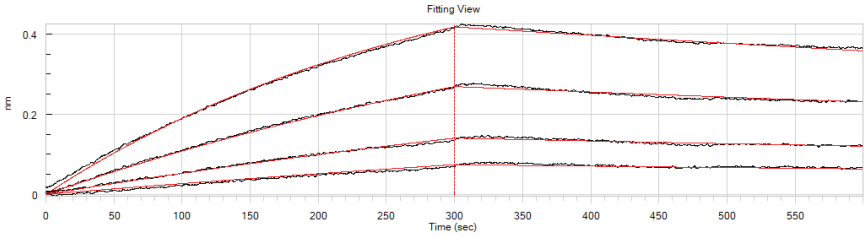


Figure S5. Examples of correlations between neutralization profiles used for hierarchical clustering of mAbs in **Figure 4**. Each point indicates neutralization of an individual HCVpp by one antibody on the x-axis and a second antibody on the y-axis. Neutralization values are fraction unaffected (Fu) at 10 µg/mL of each antibody. Fu=infection in the presence of mAb/infection in the presence of nonspecific IgG. Spearman correlations (r) and p values are indicated for each mAb pair. HEPC3, HEPC74, and HEPC43 show strong, statistically significant correlations with each other and with AR3C, but no correlation with HC33.4.

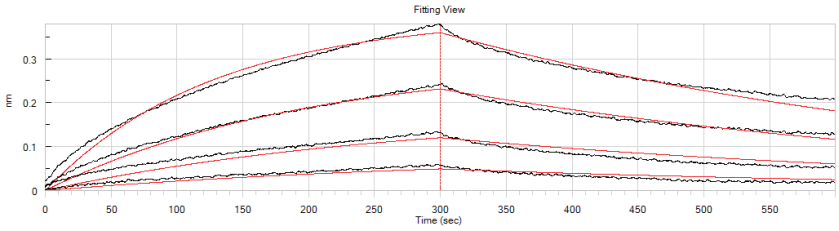
Supplemental Figure 6A

HEPC3



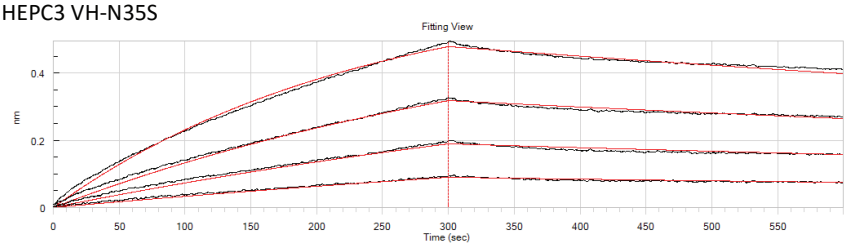
KD (M)	KD Error	Full X^2	Full R^2
5.18E-07	1.39E-08	0.11743	0.996778

HEPC3 VH-L30F



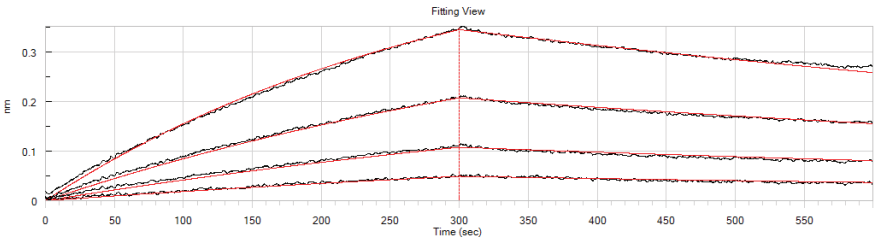
KD (M)	KD Error	Full X^2	Full R^2
1.24E-06	2.41E-08	0.236935	0.98931

HEPC3 VH-N35S



KD (M)	KD Error	Full X^2	Full R^2
5.11E-07	7.47E-09	0.095416	0.997848

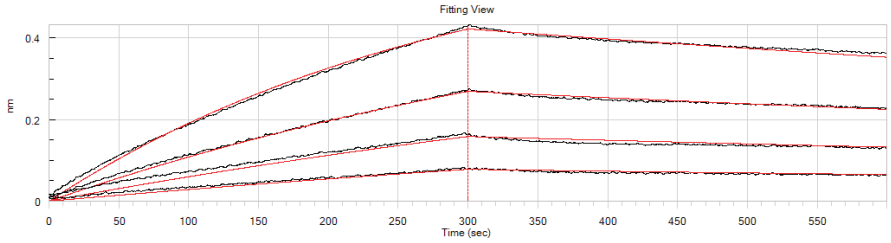
HEPC3 VH-E38A



KD (M)	KD Error	Full X^2	Full R^2
1.24E-06	1.87E-08	0.035795	0.998435

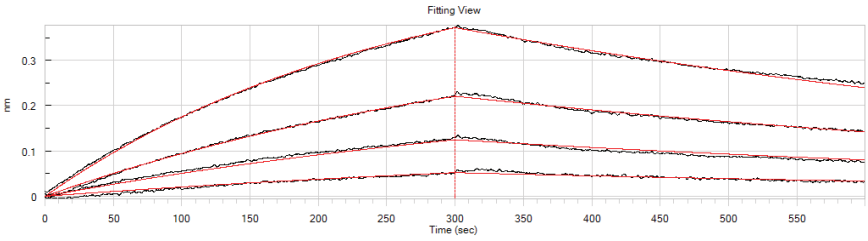
Supplemental Figure 6B

HEPC3 VH-T40S



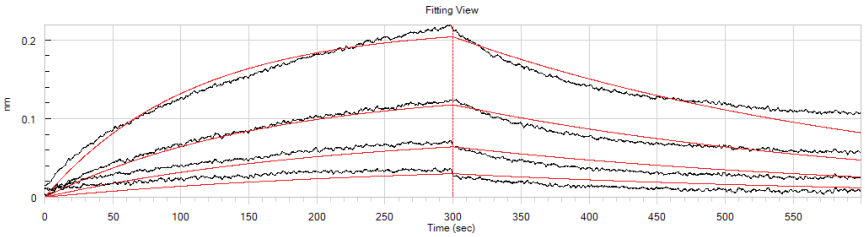
KD (M)	KD Error	Full X^2	Full R^2
6.46E-07	1.23E-08	0.091593	0.997301

HEPC3 VH-T57I



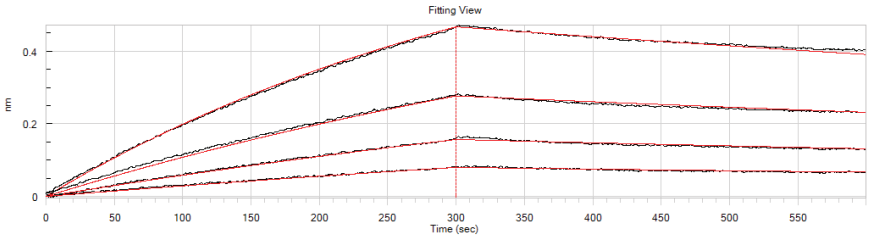
KD (M)	KD Error	Full X^2	Full R^2
1.82E-06	2.46E-08	0.035704	0.998535

HEPC3 VH-63G



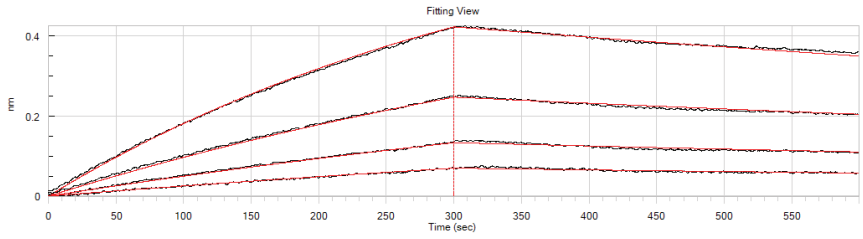
KD (M)	KD Error	Full X^2	Full R^2
1.49E-06	3.93E-08	0.149456	0.977922

HEPC3 VH-E64T



KD (M)	KD Error	Full X^2	Full R^2
8.77E-07	1.22E-08	0.038604	0.999096

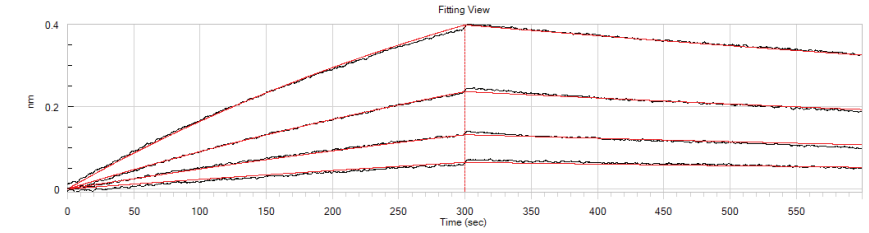
HEPC3 VH-T65A



KD (M)	KD Error	Full X^2	Full R^2
9.06E-07	1.10E-08	0.02515	0.999284

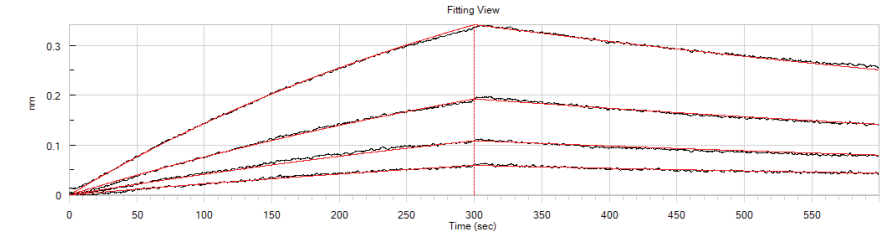
Supplemental Figure 6C

HEPC3 VH-T66N



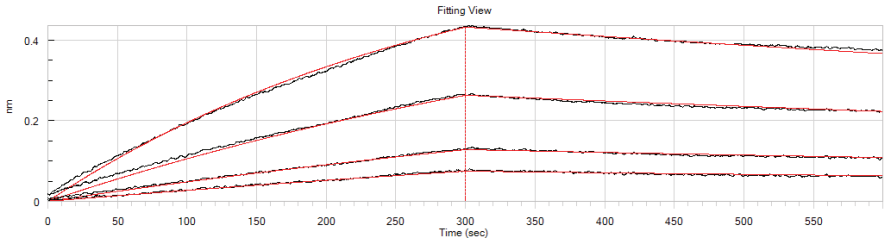
KD (M)	KD Error	Full X^2	Full R^2
1.28E-06	2.23E-08	0.032017	0.998985

HEPC3 VH-T87A



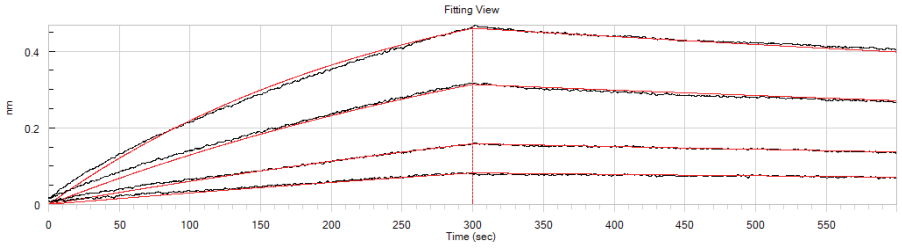
KD (M)	KD Error	Full X^2	Full R^2
2.12E-06	3.24E-08	0.015773	0.999253

HEPC3 VH-P96S



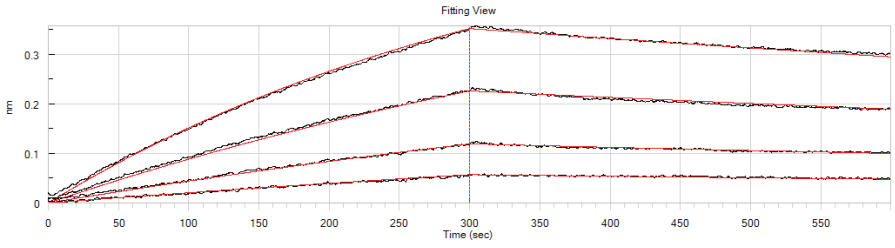
KD (M)	KD Error	Full X^2	Full R^2
5.89E-07	8.82E-09	0.054157	0.998561

HEPC3 VH-G111S



KD (M)	KD Error	Full X^2	Full R^2
4.10E-07	6.74E-09	0.087339	0.998009

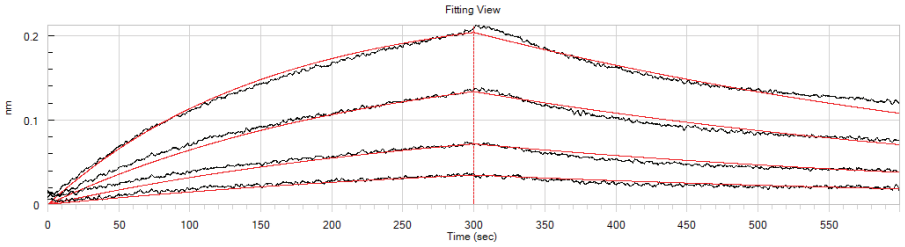
HEPC3 VH-R112S



KD (M)	KD Error	Full X^2	Full R^2
8.16E-07	1.09E-08	0.022449	0.999096

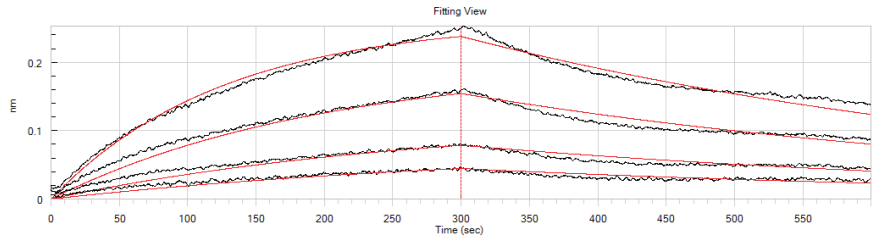
Supplemental Figure 6D

HEPC3 VH-CDR1all



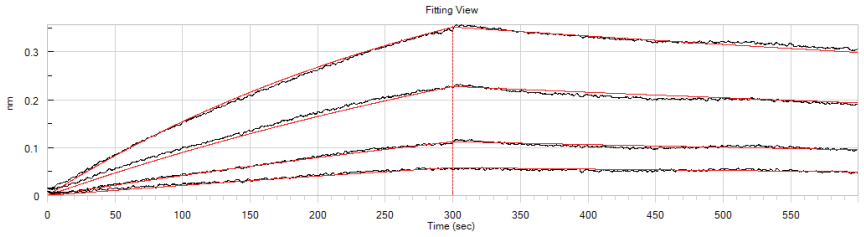
KD (M)	KD Error	Full X^2	Full R^2
1.49E-06	2.68E-08	0.046836	0.993063

HEPC3 VH-CDR2all



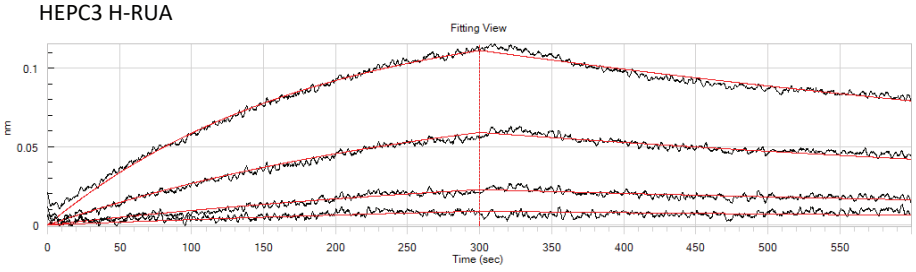
KD (M)	KD Error	Full X^2	Full R^2
1.15E-06	2.09E-08	0.095841	0.989629

HEPC3 VH-CDR3all



KD (M)	KD Error	Full X^2	Full R^2
6.66E-07	1.21E-08	0.046164	0.998161

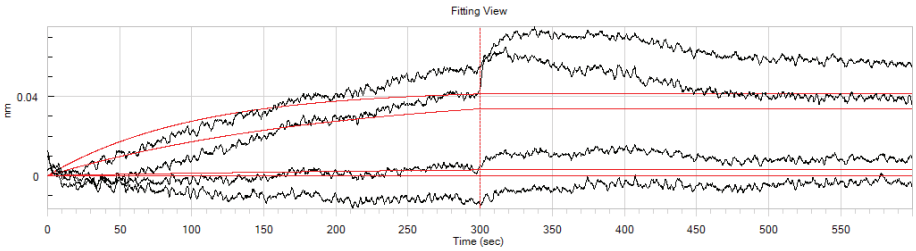
HEPC3 H-RUA



KD (M)	KD Error	Full X^2	Full R^2
7.92E-07	1.36E-08	0.01114	0.995862

isotype control mAb

MR73



KD (M)	KD Error	Full X^2	Full R^2
7.92E-07	1.36E-08	0.01114	0.995862

Figure S6. Octet association/dissociation curves with HEPC3 mAb variants and J6 (genotype 2a) soluble E2. KD values generated from these curves are summarized in Figure 6.

Supplemental Figure 7

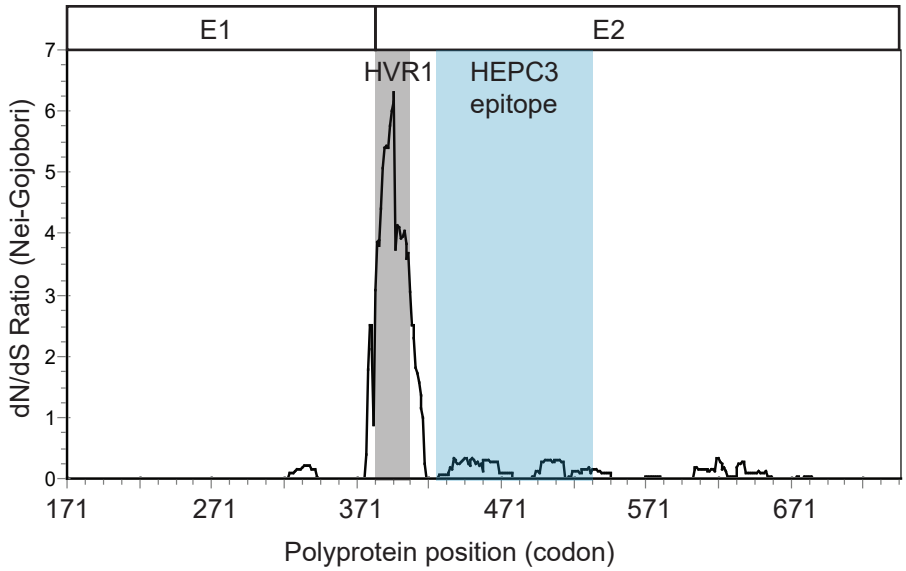


Figure S7. Ratio of nonsynonymous/synonymous mutations across all longitudinal E1E2 variants sequenced from subject 117. Analysis was performed with 20 codon windows and 1 codon steps. Hypervariable region 1 (HVR1) is shaded in gray, and the region spanning HEPC3 binding residues is shaded in blue.

Supplemental Figure 8

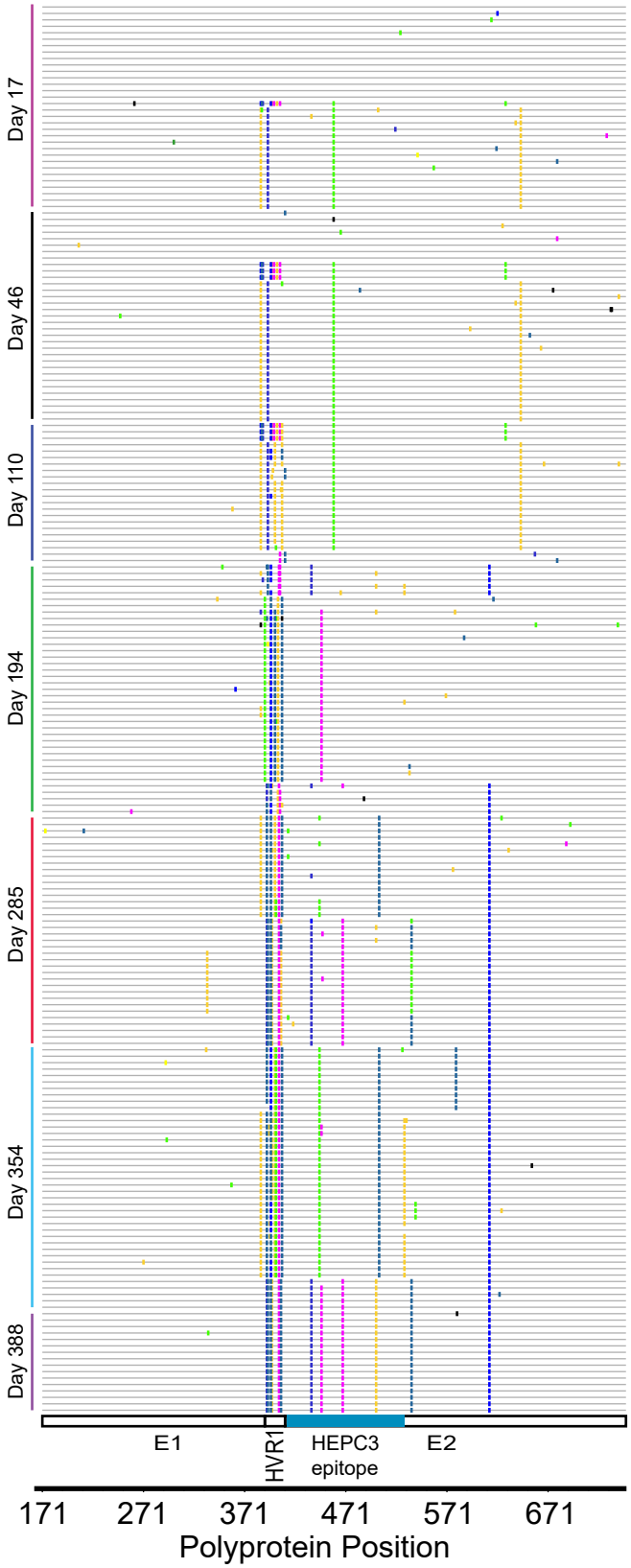


Figure S8. Highlighter plot indicating positions of amino acid differences in longitudinal E1E2 variants isolated from subject 117. Autologous variant transmitted/founder #3 (T/F#3) is used as the reference sequence, and sequences are arranged by their date of isolation. The locations of E1, E2, HVR1, and the region spanning HEPC3 binding residues are indicated.