Mapping human norovirus antigens during infection reveals the breadth of the humoral immune response

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Supplementary Figure 1. Schematic of HuNoV genomic phage library construction.

Supplementary Figure 2. Deep sequencing analysis of the inserts present in the NoV Jun-Fos library before and after affinity selection versus day 14-30 post-infection sera.

Supplementary Figure 3. In-frame fraction analysis of NoV Jun-Fos library versus post-infection sera before and after affinity selection.

Supplementary Figure 4. Alignment of peptides with high coverage after two rounds of affinity selection with sera of subject 715.

Supplementary Figure 5. Deep sequencing analysis of the inserts present in the GII.4 HOV Jun-Fos library before and after affinity selection versus GII.4 antisera.

Supplementary Figure 6. In-frame fraction analysis of GII.4 HOV Jun-Fos library before and after affinity selection versus GII.4 antisera.

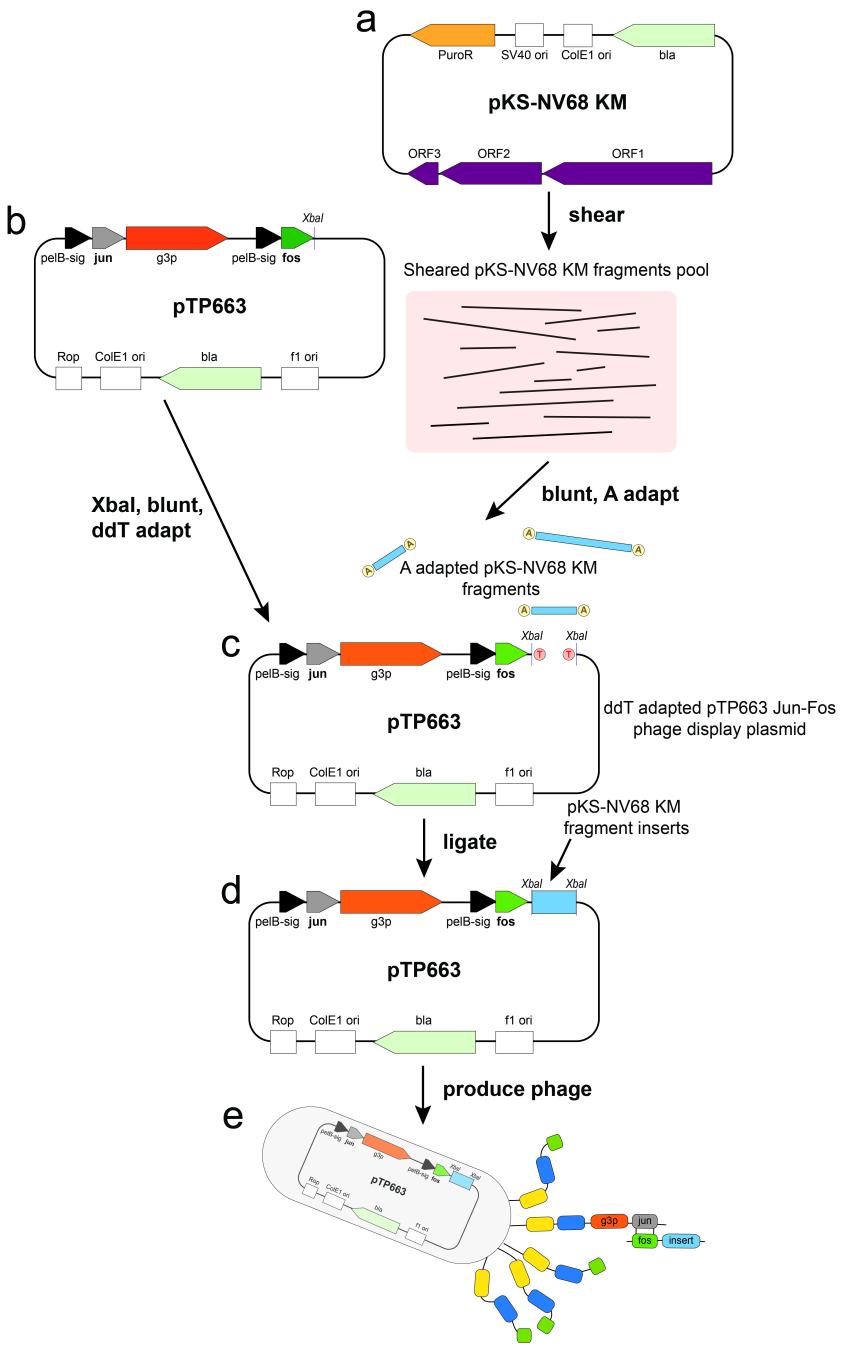
Supplementary Figure 7. Deep sequencing analysis of the inserts present in the GII.4 HOV Jun-Fos library before and after affinity selection versus GII.4 antisera.

Supplementary Figure 8. Dendrogram of GII.4 HOV epitope profiles.

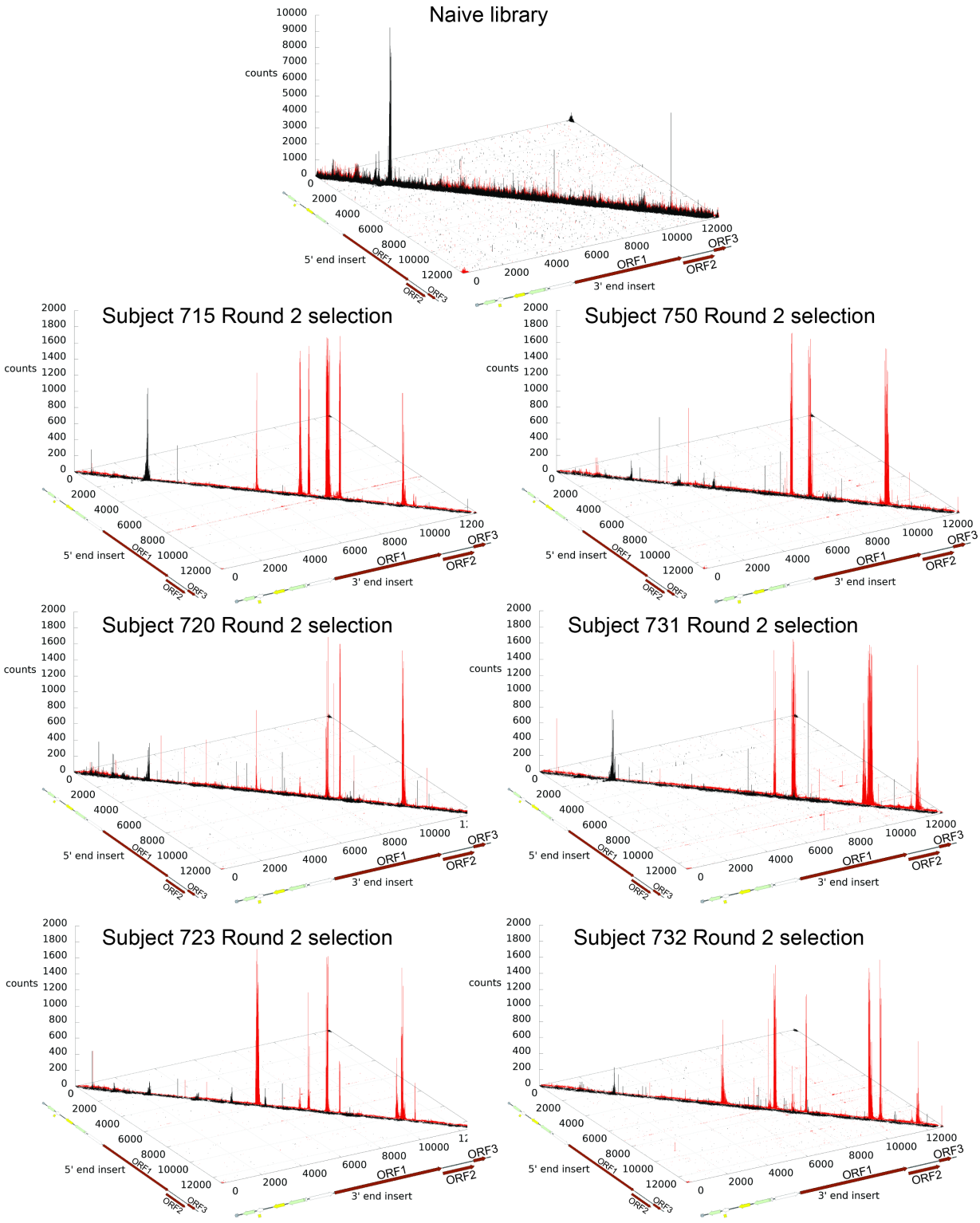
Supplementary Figure 9. Alignment of epitope sequences of GI.1 Norwalk to that of GII.4 HOV.

Supplementary Table 1. List of epitopes from all the sera identified after two rounds of affinity selections with the NoV Jun-Fos library.

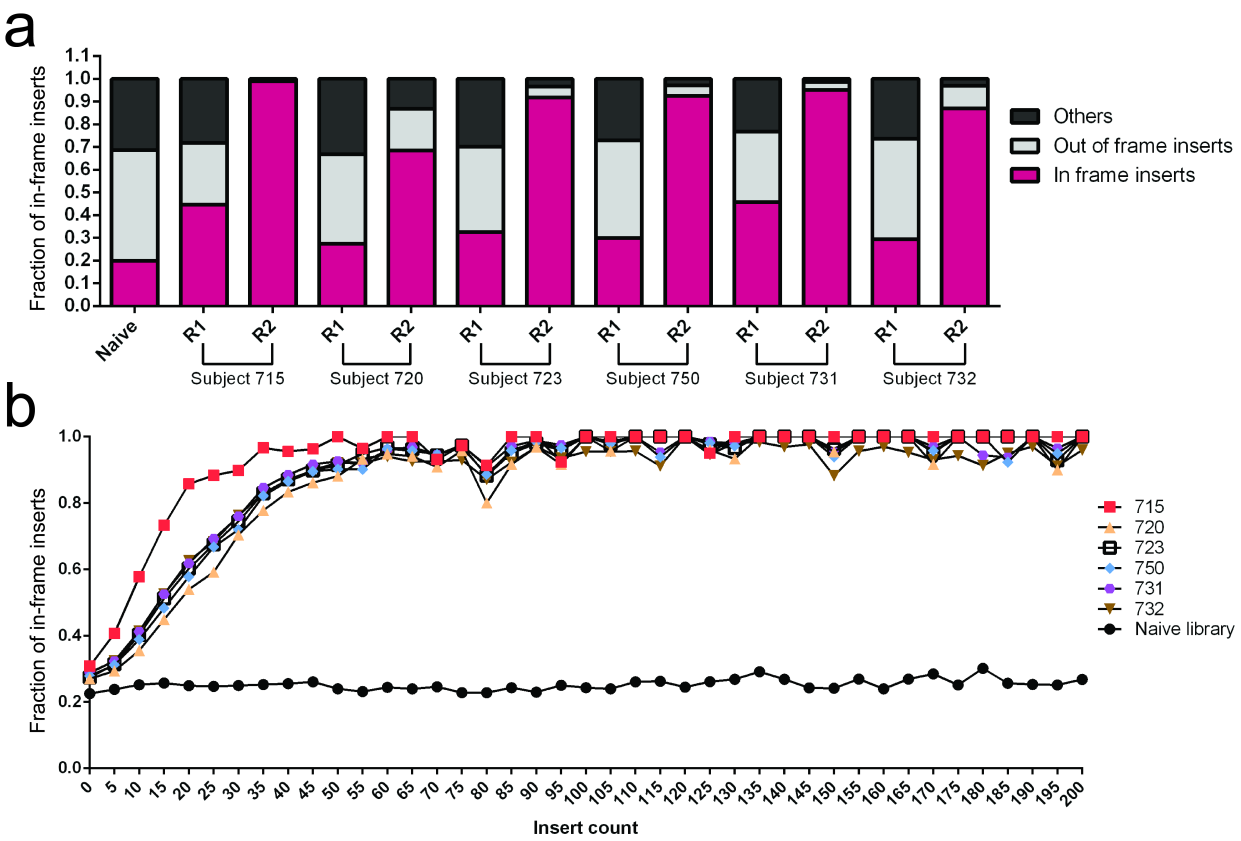
Supplementary Table 2. List of epitopes from all the sera identified after two rounds of affinity selections with the GII.4 HOV Jun-Fos library.



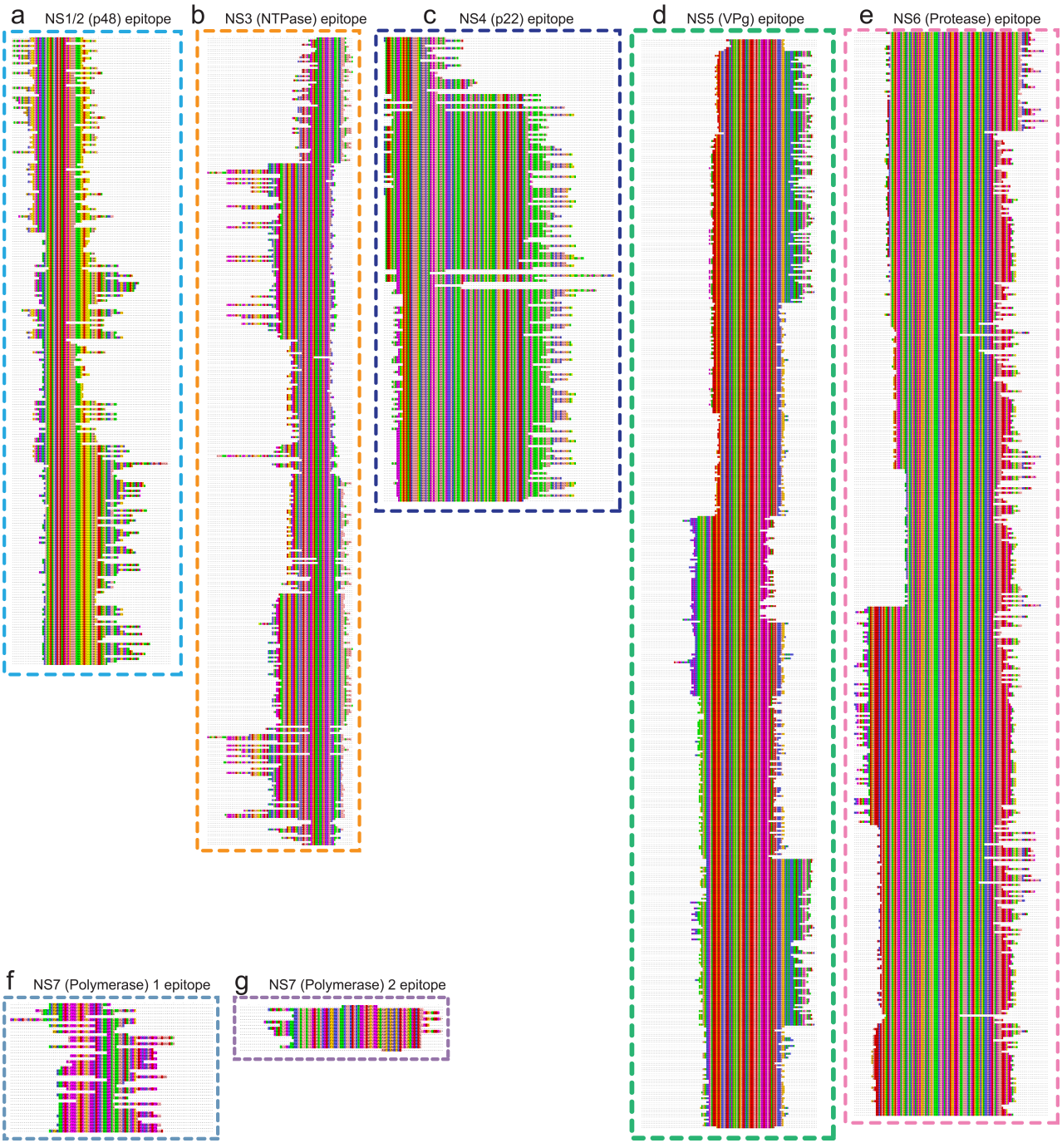
Supplementary Figure 1. Schematic of HuNoV genomic phage library construction. **a** Schematic illustration of pKS-NV68 KM plasmid, which contains the GI.1 open reading frames (ORFs) as indicated in purple, that was sheared to construct the HuNoV phage library. Since the entire plasmid was sheared, the library contains inserts from all regions of the plasmid in addition to ORFs 1 to 3. **b** Schematic illustration of the pTP663 Jun-Fos phage display plasmid. **c** Sheared DNA of 100 to 500 bp was adapted with A. **d** The pTP663 plasmid was digested with XbaI, the ends were adapted with T, and the fragments were ligated with the A-adapted sheared DNA to form the library. **e** The library clones were transformed into *E. coli*, pooled, and helper phage was added to propagate phages for the phage library to be used for affinity selection.



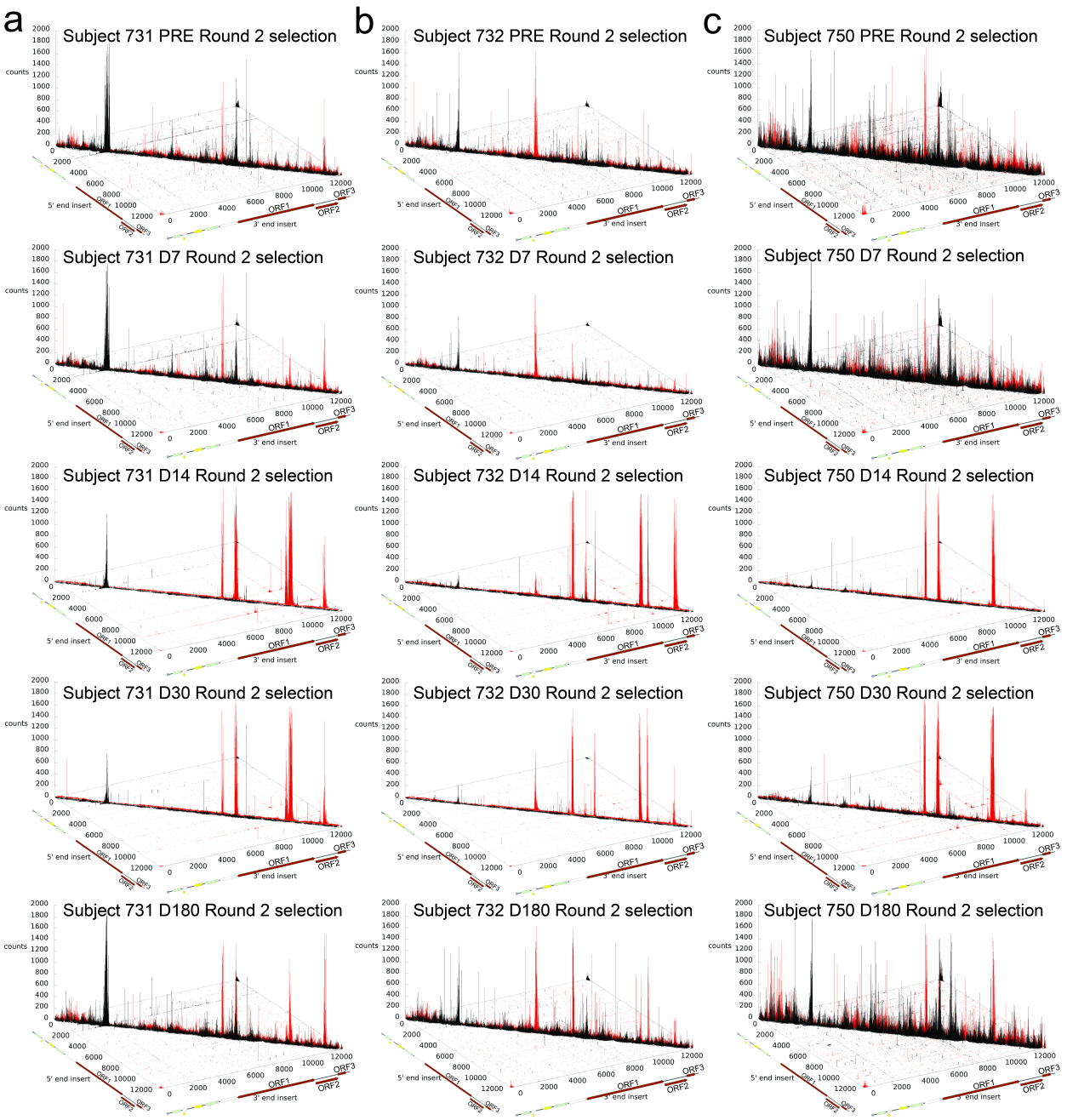
Supplementary Figure 2. Deep sequencing analysis of the inserts present in the NoV Jun-Fos library before and after affinity selection versus day 14-30 post-infection sera. Distribution of inserts after two rounds of affinity selection with sera of subjects 715, 720, 723, 750, 731, and 732.



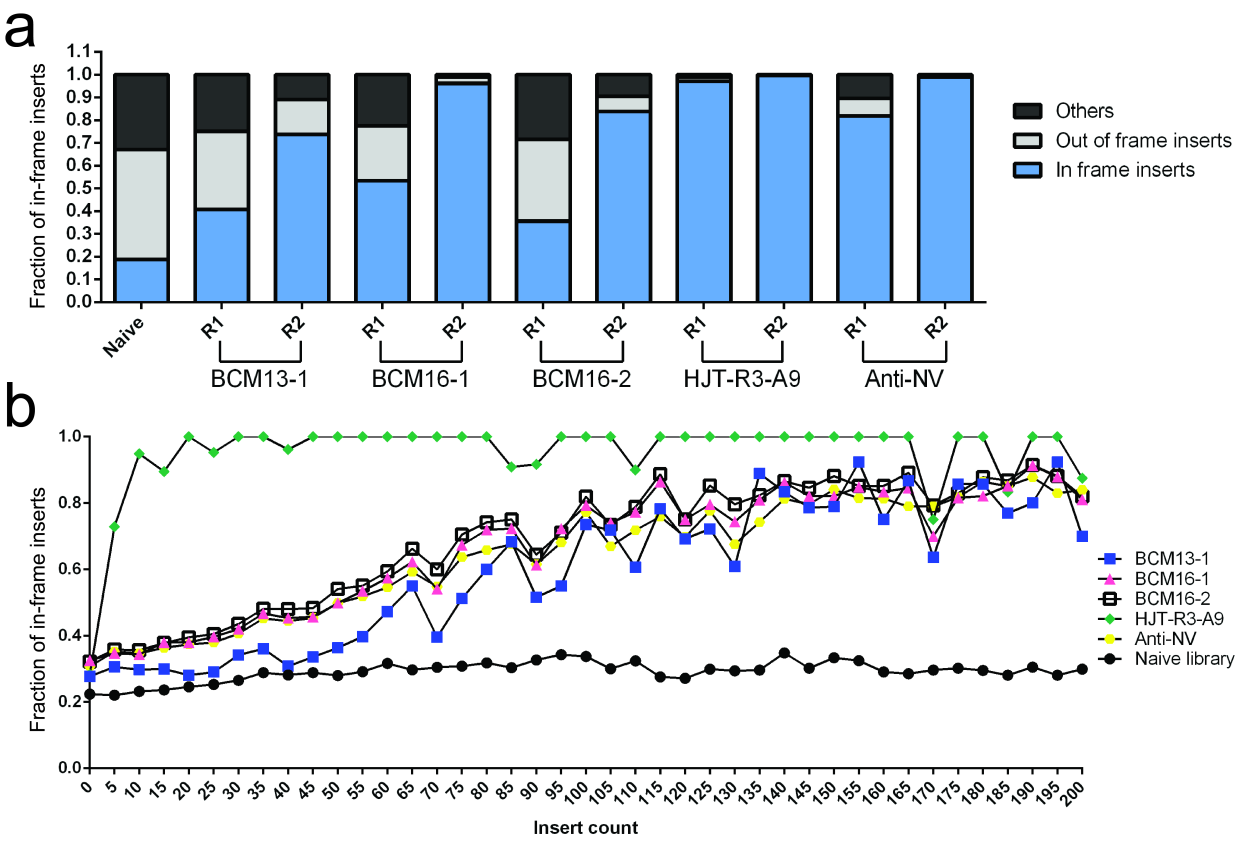
Supplementary Figure 3. In-frame fraction analysis of NoV Jun-Fos library versus post-infection sera before and after affinity selection. **a** The fraction of all the in-frame inserts in the naïve library and each round of affinity selection with subjects 715, 720, 723, 750, 731, and 732 sera. The fraction of the in-frame inserts is shown on the y-axis while the naïve library and the individual sera names are shown on the x-axis. The fraction of the inserts that are in-frame to the HuNoV ORFs are shown in magenta while the fraction of the out-of-frame inserts and the inserts that are not within HuNoV ORFs are shown in gray and black, respectively. The fraction of the in-frame inserts was determined by dividing the number of in-frame inserts by the total number of inserts in that experiment. **b** The fraction of in-frame reads for each insert count group in the naïve library and the libraries after the second round of affinity selection. The fraction of in-frame inserts is shown on the y-axis while the insert count groups are shown on the x-axis. The fraction of the in-frame inserts per insert count group is determined by dividing the number of in-frame inserts by the total number of inserts in that insert count group.



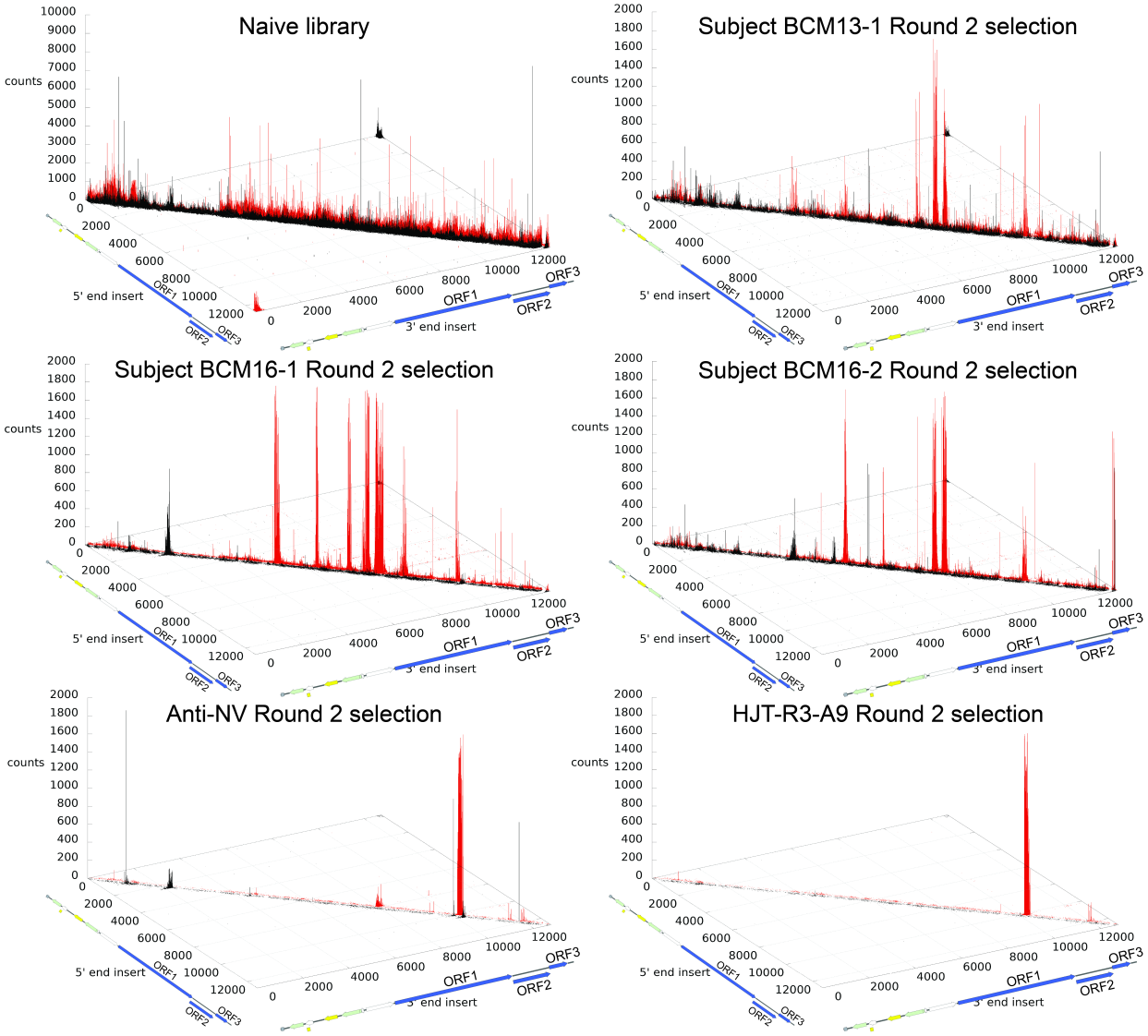
Supplementary Figure 4. Alignment of peptides with high coverage after two rounds of affinity selection with sera of subject 715. The numbering at the top indicates the amino acid residue positions in the nonstructural proteins. The Jalview Zappo color scheme is used, in which aliphatic/hydrophobic residues (I, L, V, A, and M) are peach, aromatic residues (F, W, and Y) are gold, positively charged residues (K, R, and H) are blue, negatively charged residues (D and E) are red, hydrophilic residues (S, T, N, and Q) are green, conformationally special residues (G and P) are magenta, and cysteine is yellow. The peptide alignments define the anti-GI.1 nonstructural protein epitopes in **a** NS1/2 (p48), **b** NS3 (NTPase), **c** NS4 (p22), **d** NS5 (VPg), **e** NS6 (protease), and **(f, g)** NS7 (RdRp).



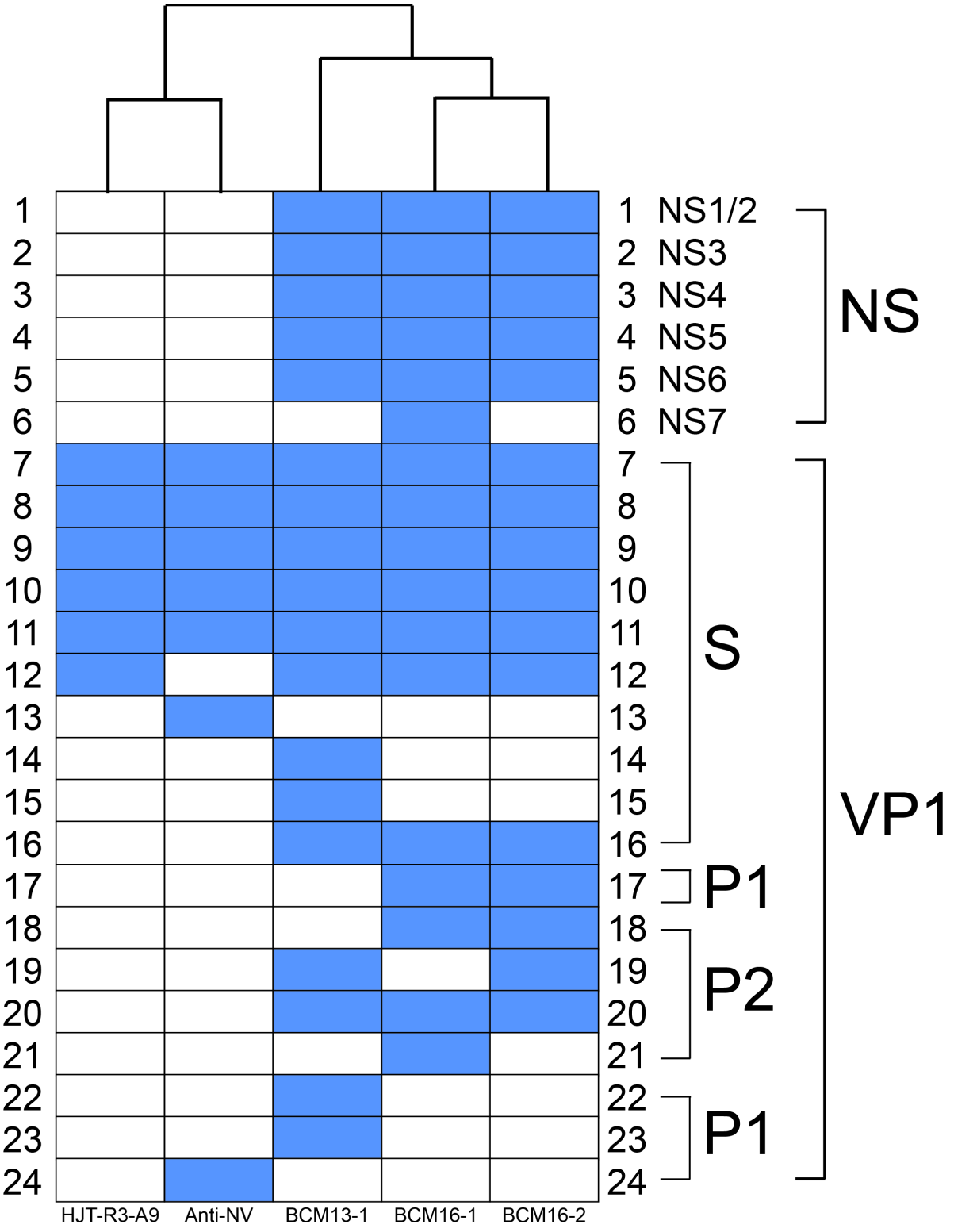
Supplementary Figure 5. Deep sequencing analysis of the inserts present in the HuNoV Jun-Fos library before and after affinity selection versus pre- and post-infection sera. **a** Distribution of inserts after two rounds of affinity selection with subject 731 sera before challenge, 7 days after challenge, 14 days after challenge, 30 days after challenge, and 180 days after challenge. **b** The distribution of inserts after two rounds of affinity selection with subject 732 sera before challenge, 7 days after challenge, 14 days after challenge, 30 days after challenge, and 180 days after challenge. **c** The distribution of inserts after round 2 of affinity selection with subject 750 sera before challenge, 7 days after challenge, 14 days after challenge, 30 days after challenge, and 180 days after challenge.



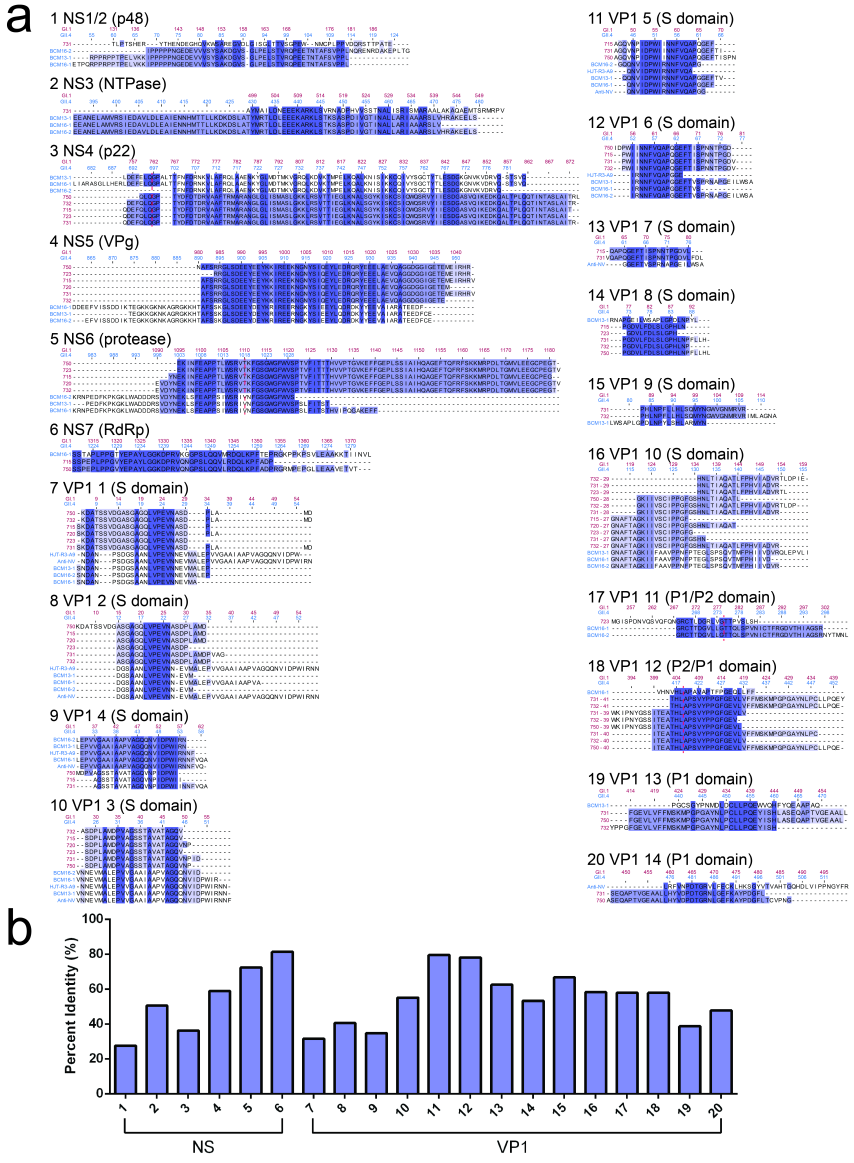
Supplementary Figure 6. In-frame fraction analysis of GII.4 HOV Jun-Fos library before and after affinity selection versus GII.4 antisera. **a** The fraction of all the in-frame inserts in the naïve library and each round of affinity selection with subjects BCM16-1, BCM13-1, and BCM16-2 sera, HJT-R3-A9 antibody, and Anti-NV. The fraction of the in-frame inserts is shown on the y-axis while the naïve library and the individual sera names are shown on the x-axis. The fraction of the inserts that are in-frame to the HuNoV ORFs are shown in blue while the fraction of the out-of-frame inserts and the inserts that are not within HuNoV ORFs are shown in gray and black, respectively. The fraction of the in-frame inserts is determined by dividing the number of in-frame inserts by the total number of inserts in that experiment. **b** The fraction of in-frame reads for each insert count group in the naïve library and the libraries after affinity selections. The fraction of in-frame inserts is shown on the y-axis while the insert count groups are shown on the x-axis. The fraction of the in-frame inserts per insert count group is determined by dividing the number of in-frame inserts by the total number of inserts in that insert count group.



Supplementary Figure 7. Deep sequencing analysis of the inserts present in the GII.4 HOV Jun-Fos library before and after affinity selection versus GII.4 antisera. Distribution of inserts after two rounds of affinity selection with BCM16-1 sera, BCM13-1 sera, BCM16-2 sera, anti-NV rabbit polyclonal antibodies, HJT-R3-A9 scFv antibody.



Supplementary Figure 8. Dendrogram of GII.4 HOV epitope profiles. A dendrogram comparing the epitope profiles shared among the four sera and HJT-R3-A9 scFv antibody. Blue blocks indicate the epitope that an individual has in the specified nonstructural or structural protein domain.



Supplementary Figure 9. Alignment of epitope sequences of GI.1 Norwalk to that of GII.4 HOV. **a** Jalview Percent Identity (PID) is used to align sequences from GI.1 Norwalk and GII.4 HOV and visualize the percent identity the sequences share. The numbering at the top indicates the amino acid residue positions in the corresponding nonstructural and structural proteins. GI.1 Norwalk positions are shown in red while the GII.4 HOV positions are shown in blue. The leftmost column indicates the name of the sera or antibody while the different shades of purple indicate the percent identity in the alignments. Percent identity is calculated according to the percentage of the residues in each column that match the consensus sequence. The darkest shade purple indicates 80% conservation between the GI.1 Norwalk and GII.4 HOV sequences. The next lighter grade of purple indicates 60%, and 40% and 20% for the subsequently lighter shades. **b** The NCBI Multiple Sequence Alignment Viewer (MSA) is used to calculate the percent identity between the GI.1 Norwalk and GII.4 HOV epitope sequences. Percent identity is calculated as the number of matches in an alignment row relative to the alignment length, where the alignment length is the aligned sequence minus any gaps for all other alignments.

		715 epitope sequence	715 epitope position	720 epitope sequence	720 epitope position	723 epitope sequence	723 epitope position	731 epitope sequence	731 epitope position	732 epitope sequence	732 epitope position	750 epitope sequence	750 epitope position	shared epitope sequence	shared epitope position	epitope length	shared among
1	1-p48									TLPTSHERYTHENDEGHQVKWSAREGVDLGISGLTTVSGPEWNMCPLPPVDQRSTTPATE	131 190			TLPTSHERYTHENDEGHQVKWSAREGVDLGISGLTTVSGPEWNMCPLPPVDQRSTTPATE	131 190	60	1
2	2-p48	VPQFKQRLTYDQLKELENEPWPYAAVTNNCFEFCCQVMCLEDT	245 287	PKQRLTYDQLKELENEPWPYAAVTNNCFEFCCQVM	248 282	VCVVPQPKQRLTYDQLKELENEPWPYAAVTNNCFEFCCQVMCLEDTWLQRKLIS	242 295							PKQRLTYDQLKELENEPWPYAAVTNNCFEFCCQVM	248 282	35	3
3	3-p48							QDSKLEMVRDAVLLGKLKPLHVLNLLS	315 355					QDSKLEMVRDAVLAAINGLVSRPFKDLLGKLKPLHVLNLLS	315 355	41	1
4	1-NTPase							DLVPVLGGIGLAIGFTRDKVSKMMKNAVDGLRAATQLGQYGL	406 448					DLVPVLGGIGLAIGFTRDKVSKMMKNAVDGLRAATQLGQYGL	406 448	43	1
5	2-NTPase							AYMAILDNEEEKARKLSVRNADPHVVSSTNALISRISMARALAKAQAEMTSRMRPV	498 554					AYMAILDNEEEKARKLSVRNADPHVVSSTNALISRISMARALAKAQAEMTSRMRPV	498 554	57	1
6	3-NTPase	DTTALKDCFKPDFSHLKMELAPQGGFDNQGNTPFGKGVMKPTTINRLL	697 744	DTTALKDCFKPDFSHLKMELAPQGGFDNQGNTPFGKGVM KPTTINRL	697 743	GDTTALKDCFKPDFSHLKMELAPQGGFDNQGNTPFGKGVMKPTTINRL	696 743							DTTALKDCFKPDFSHLKMELAPQGGFDNQGNTPFGKGVMKPTTINRL	697 743	47	1
7	1-p22	QDEFQLQGPTYDFDTDRVAAFTRMARANGLGLISMASLGKKLRSVTTIEGLKNALSGYKISKCSIQIWQSRVYIIESDGASVQIKEDKQALTPLQQTINTASLAIT	755 859	QDEFQLQGPTYDFDTDRVAAFTRMARANGLGLISMASLGKKLRSVTTIEGLKNALSGYKISKCSIQIWQSRVYIIESDGASV QIKEDKQALTPLQ QTINTASLAI	755 858	QDEFQLQGPTYDFDTDRVAAFTRMARANGLGLISMASLGKKLRSVTTIEGLKNALSGYKISKCSIQIWQSRVYIIESDGASVQIKEDKQALTPLQ QTINTASLAITR	755 860	QDEFQLQGPTYDFDTDRVAAFTRMARANGLGLISMASLGKKLRSVTTIEGLKNALSGYKISKCSIQIWQSRVYIIESDGASVQIKEDKQALTPLQ QTINTASLAITR	756 860	QLQGPTYDFDTDRVAAFTRMARANGLGLISMASLGKKLRSVTTIEGLKNALSGYKISKCSIQIWQSRVYIIESDGASVQIKEDKQALTPLQ QTINTASLAITRL	759 861	QLQGPTYDFDTDRVAAFTRMARANGLGLISMASLGKKLRSVTTIEGLKNALSGYKISKCSIQIWQSRVYIIESDGASVQIKEDKQALTPLQQTINTASLAI	759 858	100	5
8	1-VPg	APSRRGLSDEEYEYKKIREEKNGNYSIQEYLEDRQRYEELAEVDAGGDGGIGETEMEIRHRV	981 1044	AFSRRGLSDEEYEYKKIREEKNGNYSIQEYLEDRQRYEELAEVDAGGDGGIGETEM	981 1044	RRGLSDEEYEEYKKIREEKNGNYSIQEYLEDRQRYEELAEVDAGGDGGIGETEMEIRHR	984 1043	AFSRRGLSDEEYEYKKIREEKNGNYSIQEYLEDRQRYEELAEVDAGGDGGIGETEMEIRHR	981 1044	AFSRRGLSDEEYEYKKIREEKNGNYSIQEYLEDRQRYEELAEVDAGGDGGIGETE	981 1037	NAFSRRGLSDEEYEEYKKIREEKNGNYSIQEYLEDRQRYEELAEVQAGDGGIGETEMEIRHR	980 1043	RRGLSDEEYEEYKKIREEKNGNYSIQEYLEDRQRYEELAEVQAGDGGIGETE	984 1037	54	6
9	1-protease	YNEKINFEAPPTLWSRVTKFGSGWGFWGFSPTVFITTHMVPTGVKEFFGEPLSIAIHQAGEFTQFRFSKKMRPDLTGMVLEEGCPEG	1093 1180	EVDYNEKINFEAPPTLWSRVTKFGSGWGFWVSPTVFITTHMVPTGVKEFFG EPLSSIAIHQAGEF TQFRFSKKMRPDL TGMVLEEGCPEGTV	1090 1186	EKINFEAPPTLWSRVTKFGSGWGFWVSPTVFITTHMVPTGVKEFFGEPLSSIAIHQAGEFTQFRFSKKMRPDLTGMVLEEGCPEGTV	1095 1182	EVDYNEKINFEAPPTLWSRVTKFGSGWGFWVSPTVFITTHMVPTGVKEFFGEPLSSIAIHQAGEFTQFRFSKKMRPDLTGMVLEEGCPEGT	1090 1181	EKINFEAPPTLWSRVTKFGSGWGFWVSPTVFITTHMVPTGVKEFFGEPLSSIAIHQAGEFTQFRFSKKMRPDLTGMVLEEGCPEGT	1095 1181	EKINFEAPPTLWSRVTKFGSGWGFWVSPTVFITTHMVPTGVKEFFGEPLSSIAIHQAGEFTQFRFSKKMRPDLTGMVLEEGCPEG	1095 1180	86	3
10	1-RdRp	SSPEPLPPGVYEPAYLGGKDPRVQNGPSLQQVLRDQLKPFADP	1311 1353									SSPEPLPPGVYEPAYLGGKDPRVQNGPSLQQVLRDQLKPFADPGRMMEPGLLEAAVETVT	1311 1371	SSPEPLPPGVYEPAYLGGKDPRVQNGPSLQQVLRDQLKPFADP	1311 1353	43	2
11	2-RdRp							RLLWGADLGTVVRAARAFGPFCDAIKSHVIKLPIKVGMNTIEDGPLIYAEHAKYKNHFDADYTAWDSTQNRQIMTESF	1462 1539	SHVIKLPIKVGMNTIEDGPLIYAEHAKYKNHFDADYTAWDSTQNRQIMTESF	1488 1539			SHVIKLPIKVGMNTIEDGPLIYAEHAKYKNHFDADYTAWDSTQNRQIMTESF	1488 1539	52	2
12	3-RdRp							GLPSGPFCTSQVNSINHWIITLCALSEATGLSPDVVQSMSYFSFYGDDEIVSTDIDFDPARLTQILKEYGLKPTRPD	1577 1653					GLPSGPFCTSQVNSINHWIITLCALSEATGLSPDVVQSMSYFSFYGDDEIVSTDIDFDPARLTQILKEYGLKPTRPD	1577 1653	77	1
13	4-RdRp					PSETLVPHTQRKIQLISLLGEASLHDEKYRKISSKVIHEIKTGGLEMYVPGWQAMFRWMRFHDLGLWTGDRDLLPE	1706 1782	SDPSETLVPHTQRKIQLISLLGEASLHGEKFYRKISSKVIHEIKTGGLEMYVPGWQAMFRWMRFHDLGLWTGDRDLLPEFVNDD	1704 1787			HSDPSETLVPHTQRKIQLISLLGEASLHGEKFYRKISSKVIHEIKTGGLEMYVPGWQAMFRWMRFHDLGLWTGDRDLLPEF	1703 1783	TLVPHTQRKIQLISLLGEASLHEDKYRKISKVIHEIKTGGLEMYVPGWQAMFRWMRFHDLGLWTGDRDLL	1709 1780	72	4
14	1-VP1 (S domain)	SKDATSSVDGASGAGQLVPEVNASDPLAMD	5 30	SKDATSSVDGASGAGQLVPEVNASDPLA	6 32	KDATSSVDGASGAGQLVPEVNASDPLAMD	6 29	KDATSSVDGASGAGQLVPEVNASDPLAMD	5 34	KDATSSVDGASGAGQLVPEVNASDPLAMD	6 34	KDATSSVDGASGAGQLVPEVNASDPLAMD	6 34	KDATSSVDGASGAGQLVPEVNASD	6 29	24	6
15	1-VP1 (S domain)	ASGAGQLVPEVNAVSDPLAMD	15 34	ASGAGQLVPEVNASDPLAMD	15 34	ASGAGQLVPEVNASDP	15 30	ASGAGQLVPEVNASDPLAMDPVAG	15 38	ASGAGQLVPEVNASDPLAMDP	15 35	ASGAGQLVPEVNASDPLAMDPVAG	15 38	ASGAGQLVPEVNASDP	15 30	16	4
16	1-VP1 (S domain)	PEVNASDPLAMDPVA	23 36	PEVNASDPLAMDPVAG	23 38	GQLVPEVNASDPLAMDPVA	19 37	GQLVPEVNASDPLAMDPVAGSSTAVAT	19 45	GQLVPEVNASDPLAMDPVAGS	19 39	VPEVNASDPLAMDPVAGS	22 39	PEVNASDPLAMDPVA	23 37	15	4
17	1-VP1 (S domain)	SDPLAMDPVAGSSTAVATAGQV	28 49	SDPLAMDPVAGSSTAVATAGQVNP	28 51	SDPLAMDPVAGSSTAVATAGQV	28 49	SDPLAMDPVAGSSTAVATAGQVNPID	28 53	SDPLAMDPVAGSSTAVATAGQV	27 49	SDPLAMDPVAGSSTAVATAGQVNP	28 51	SDPLAMDPVAGSSTAVATAGQV	28 49	22	6
18	1-VP1 (S domain)	AGSSTAVATAGQVNPLDPWII	37 57					AGSSTAVATAGQVNPIDPWIINNFVQAPQGEF	37 63			MDPVAGSSTAVATAGQVNPIDPWI	33 56	AGSSTAVATAGQVNPIDPWI	37 56	20	3
19	1-VP1 (S domain)	AGQVNPIDPWIINNFVQAPQGEF	46 68					AGQVNPIDPWIINNFVQAPQGEFTI	46 70			AGQVNPIDPWIINNFVQAPQGEFTISPN	46 73	AGQVNPIDPWIINNFVQAPQGEF	46 68	23	3
20	1-VP1 (S domain)	PWIINNFVQAPQGEFTISPNNTPGD	54 78					PWIINNFVQAPQGEFTISPNNTPGDV	54 79	PWIINNFVQAPQGEFTISPNNTPGD	54 79	IDPWIINNFVQAPQGEFTISPNNTPGD	52 78	PWIINNFVQAPQGEFTISPNNTPGD	54 78	25	4
21	1-VP1 (S domain)	QAPQGEFTISPNNTPGDVL	62 80					VQAPQGEFTISPNNTPGDVLFDL	61 83					QAPQGEFTISPNNTPGDVL	62 80	19	2
22	1-VP1 (S domain)	PGDVLFDLSLGPHLN	76 90			GDVLFDLSLGPH	77 88	PGDVLFDLSLGPHLNPFLLH	76 95	PGDVLFDLSLGPHLNP	76 91	PGDVLFDLSLGPHLNPFLLHL	76 96	GDVLFDLSLGPH	77 88	12	3
23	1-VP1 (S domain)							PHLNPFLLHLSQMYNGWVGNMRVRIMLAGNA	87 110	PHLNPFLLHLSQMYNGWVGNMRVRIMLAGNA	87 117			PHLNPFLLHLSQMYNGWVGNMRVR	87 110	24	3
24	1-VP1 (S domain)	LHLSQMYNGWVGNMRVRIMLAGN	94 116					LHLSQMYNGWVGNMRVRIMLAGN	94 116	LHLSQMYNGWVGNMRVRIMLAGNAFTAGKIV	94 125			LHLSQMYNGWVGNMRVRIMLAGN	94 116	23	3
25	1-VP1 (S domain)							GWVGNMRVRIMLAGNAFTAGKIV	102 125	MYNGNWGNMRVRIMLAGNAFTAGKIVSCIPPGFGSHNLTIAQAT	99 143	GWVGNMRVRIMLAGNAFTAGKIVSCIPPG	102 131	GWVGNMRVRIMLAGNAFTAGKIV	102 125	24	3
26	1-VP1 (S domain)	VRIMLAGNAFTAGKIVSCIPPGFGSHNLTIAQATLPPHV	109 148			VRIMLAGNAFTAGKIVSCIPPGFGSHNLTIAQAT	109 143	VRIMLAGNAFTAGKIVSCIPPGFGSHNLTIAQATLPPHVI	109 149	VRIMLAGNAFTAGKIVSCIPPGFGSHNLTIAQATLPPHV	109 148	VRIMLAGNAFTAGKIVSCIPPGFGSHNLTIAQAT	109 148	VRIMLAGNAFTAGKIVSCIPPGFGSHNLTIAQAT	109 143	35	5
27	1-VP1 (S domain)	GNAFTAGKIVSCIPPGF	115 132	GNAFTAGKIVSCIPPGFGSHNLTIAQAT	115 143	GNAFTAGKIVSCIPPGFG	115 133	GNAFTAGKIVSCIPPGFGSHN	115 136	GNAFTAGKIVSCIPPGFGSHNLTIAQATLPPHVIADVR	115 153			GNAFTAGKIVSCIPPGF	115 132	18	1
28	1-VP1 (S domain)							GKIIVSCIPPGFGSHNLTIAQATLPPHVIADVR	121 153	GKIIVSCIPPGFGSHNLTIAQATLPPHVIADVRTLDP	121 157	GKIIVSCIPPGFGSHNLTIAQATL	121 144	GKIIVSCIPPGFGSHNLTIAQAT	121 144	24	3
29	1-VP1 (S domain)					HNLTIAQATLPPHVIADVRTL	135 155	HNLTIAQATLPPHVIADVR	135 153	HNLTIAQATLPPHVIADVRTLDPIE	135 159			HNLTIAQATLPPHVIADVR	135 153	19	3
30	1-VP1 (S domain)	ADVRTLDPIEVPLEDVR	150 166											ADVRTLDPIEVPLEDVR	150 166	17	1
31	1-VP1 (S domain)	RTLDPIEVPLEDVRNV	153 168	RTLDPIEVPLEDVRNV	153 168	RTLDPIEVPLEDVRNV	153 168	LDPIEVPLEDVRN	155 167	VRTLDPIEVPLEDVRN/	152 168	VRTLDPIEVPLEDVRNVRNQGTM	152 180	LDPIEVPLEDVRN	155 167	13	6
32	1-VP1 (S domain)									RNVLFHNNDRNQQTMRLVCMLYTPLRTGG	166 194	RNVLFHNNDRNQQTMRLVCMLYTPLR	166 191	RNVLFHNNDRNQQTMRLVCMLYTPLR	166 191	26	2
33	1-VP1 (P1 domain)											FNFLFLVPPTVEQKTRPFTLPNLPSSL	214 241	FNFLFLVPPTVEQKTRPFTLPNLPSSL	214 241	28	1
34	1-VP1 (P1 domain)					MGISPDNVQSVQFQNGRCTLDGRLVGTTPVSLSH	253 286							MGISPDNVQSVQFQNGRCTLDGRLVGTTPVSLSH	253 286	34	1
35	1-VP1 (P2 domain)									IQANGIGSGNYVGVLSWISPPSHPS	359 383			IQANGIGSGNYVGVLSWISPPSHPS	359 383	25	1
36	1-VP1 (P2 domain)							GNYVGVLSWISPPSHPSGSQVDLWKIPNYGSSITEAT	367 403	GNYVGVLSWISPPSHPSGSQVDLWKIP	367 393	GNYVGVLSWISPPSHPSGSQVDLWKI	367 392	GNYVGVLSWISPPSHPSGSQVDLWKI	367 392	26	3
37	1-VP1 (P2 domain)									LSWISPPSHPSGSQVDLWK	373 391	SWISPPSHPSGSQVD	374 388	SWISPPSHPSGSQVD	374 388	15	2
38	1-VP1 (P2 domain)							PSHPSGSQVDLWKIPNYGSSITEATHLAPSVYPP	379 411	PSHPSGSQVDLWKIPNYGSSITEAT	379 403			PSHPSGSQVDLWKIPNYGSSITEAT	379 403	25	2
39	1-VP1 (P1 domain)							WKIPNYGSSITEATHLAPSVYPPGFGEVLV	390 419	WKIPNYGSSITEATHLAPSVYPPGFGEV	390 417			WKIPNYGSSITEATHLAPSVYPPGFGEV	390 417	28	3
40	1-VP1 (P1 domain)							ITEATHLAPSVYPPGFGEVLVFFMSKMPGPGAYNLPC	399 435	ITEATHLAPSVYPPGFGEVLV	399 419			ITEATHLAPSVYPPGFGEVLV	399 419	21	3
41	1-VP1 (P1 domain)							THLAPSVYPPGFGEVLVFFMSKMPGPGAYNLPCLLPQEY	403 441	THLAPSVYPPGFGEVLVFFMSKMPGPGAYNLPCLLPQE	403 440			THLAPSVYPPGFGEVLVFFMSKMPGPGAYNLPCLLPQE	403 440	38	2
42	1-VP1 (P1 domain)							FGEVLVFFMSKMPGPGAYNLPCLLPQEYISHLASEQAPTVGEAALL	414 459	YPPGFGEVLVFFMSKMPGPGAYNLPCLLPQEYISH	410 444	FGEVLVFFMSKMPGPGAYNLPCLLPQEYISHLASEQAPTVGEAAL	414 452	FGEVLVFFMSKMPGPGAYNLPCLLPQEYISH	414 444	31	3
43	1-VP1 (P1 domain)							SEQAPTVGEAALLHYVDPDTGRNLGEPKAYPDGFL	447 481			ASEQAPTVGEAALLHYVDPDTGRNLGEPKAYPDGFLTCVPNGASSPNG	446 487	SEQAPTVGEAALLHYVDPDTGRNLGEPKAYPDGFL	447 481	35	2
44	1-VP1 (P1 domain)											DGFLTCVPNGASSGPQQLPING	478 499	DGFLTCVPNGASSGPQQLPING	478 499	22	3
45	1-VP1 (P1 domain)			PQQLPINGVFV	492 502			GPQQLPINGVFVFVSWVSRFYQLK	491 514			QQLPINGVFVFVSWVS	493 508	QQLPINGVFV	493 502	10	3
46	1-VP1 (P1 domain)							INGVFVFVSWVSRFYQLKP	497 515			INGVFVFVSWVS	497 508	INGVFVFVSWVS	497 508	12	2
47	1-VP1 (P1 domain)			SRFYQLKPVGTASSARG	508 523			YQLKPVGTASSARG	511 524	YQLKPVGTASSARG	511 524			YQLKPVGTASSARG	511 523	13	3
48	1-VP1 (P1 domain)							GTASSARG	517 524	GTASSARGR	517 525	LKPVGTASSARG	513 524	GTASSARG	517 524	8	3

Supplementary Table 1. List of epitopes from all the sera identified after two rounds of affinity selections with the NoV Jun-Fos library. A total of 48 unique epitopes were identified. The sequences and the positions in the GI.1 Norwalk ORF1 and ORF2 genome of each epitope are included. The shared epitopes amongst the six individuals as well as the epitopes' sequences, positions in the GI.1 Norwalk genome, length, and the number of sera that contain the epitopes are also included.

		HJT-R3-A9 epitope sequence	HJT-R3-A9 epitope position	BCM131 epitope sequence	BCM131 epitope position	BCM161 epitope sequence	BCM161 epitope position	BCM162 epitope sequence	BCM161 epitope position	Anti-NV epitope sequence	Anti-NV epitope position	shared epitope sequence	shared epitope position	epitope length	shared among
1	1-p48			RPPRRPTPELVKKI PPPPPNGEDEVVV SYSAKDGVSGLPE LSTVRQPEETNTAF SVPPL	55 131	ETPQRPPRPPTPE LVKKIPPPPNGED EVVVSYSAKDGVS GLPELSTVRQPEE TNTAFSVPPL	51 113	IPPPPPNGEDEVVV SYSAKDGVSGLPE LSTVRQPEETNTAF SVPPLNQRENRDA KEPLTG	68 127			IPPPPPNGEDEVVV SYSAKDGVSGLPE LSTVRQPEETNTAF SVPPL	68 113	46	3
2	2-NTPase			EEANELAMVRSIED AVLDLEAIENNHMT TLKKDKDSLATYM RTLDLEEEKARKLS TKSASPDIVGTINA LLARIAAARSLVHR AKEELS	395 483	EEANELAMVRSIED AVLDLEAIENNHMT TLLKDKDSLATYM RTLDLEEEKARKLS TKSASPDIVGTINA LLARIAAARSLV	395 475	EEANELAMVRSIED AVLDLEAIENNHMT TLKKDKDSLATYM RTLDLEEEKARKLS TKSASPDIVGTINA LLARIAAARSLVHR AKEELS	395 483			EEANELAMVRSIED AVLDLEAIENNHMT TLKKDKDSLATYM RTLDLEEEKARKLS TKSASPDIVGTINA LLARIAAARSLV	395 475	81	3
3	3-p22			LDEFELOGPALTTF NFDRNKVLAFRQL AAENKYGLMDTMK VGRQLKDVKTMPE LKQALKNISIKKCQI VYSGCTYTLESDG KGNVKVDRVQSTS VQ	690 785	LIARASGLLHERLD EFELQGPALTTFNF DRNKVLAFRQLAA ENKYGLMDTMKV GRQLKDVKTMPEL KQALKNISIKKCQIV YSGCTYTLESDGK GNVKVDRVQSTSV Q	678 785	TFNFDRNKVLAFR QLAAENKYGLMDT MKVGRQLKDVKT MPELKQALKNISIK KCQIVYSGCTYTL ESDGKGNVKVDRV Q	702 780			TFNFDRNKVLAFR QLAAENKYGLMDT MKVGRQLKDVKT MPELKQALKNISIK KCQIVYSGCTYTL ESDGKGNVKVDRV	702 779	78	3
4	4-VPg			TEGKKGKNKAGR GKKHTAFSSKGLS DEEYDEYKRIREE RNGKYSIEEYLQD RDKYYEEVAIARAT EEDFCE	874 944	DDEEFVISSDDIKT EGKKGKNKAGRG KKHTAFSSKGLSD EEYDEYKRIREER NGKYSIEEYLQDR DKYYEEVAIARATE EDF	861 942	EFVISSDDIKTEGK KGKNKAGRGKKHT AFSSKGLSDEEYD EYKRIREERNGKY SIEEYLQDRDKYY EEVAIARATEEDFC E	864 944			TEGKKGKNKAGR GKKHTAFSSKGLS DEEYDEYKRIREE RNGKYSIEEYLQD RDKYYEEVAIARAT EEDF	872 942	69	3
5	5-protease			PEDFKPKGKLWAD DDRSVDYNEKLSF EAPPSIWSRIVNFG SGWGFWVSPSLFI TST	982 1037	KRNPEDFKPKGKL WADDDRSVDYNE KLSFEAPPSIWSRI VNFGSGWGFWVS PSLFITSTHVIPGG AKEFF	979 1048	KRNPEDFKPKGKL WADDDRSVDYNE KLSFEAPPSIWSRI VNFGSGWGFWVS	979 1029			PEDFKPKGKLWAD DDRSVDYNEKLSF EAPPSIWSRIVNFG SGWGFWVS	982 1029	48	3
6	6-RdRp					SSTAPLPPGTYEP AYLGGKDPRVKG GPSLQQVMRDQL KPFTEPRGKPPKP SVLEAAKKTIINVL	1220 1283					SSTAPLPPGTYEP AYLGGKDPRVKG GPSLQQVMRDQL KPFTEPRGKPPKP SVLEAAKKTIINVL	1220 1283	64	1
7	1-VP1 (S domain)	NDANPSDGSAANL VPEVNNEVMALEP VVGAAIAAPVAGQ QNVIDPW	6 51	SNDANPSDGSAAN LVPEVNNEVMALE P	5 31	SNDANPSDGSAAN LVPEVNNEVMALE P	5 31	SNDANPSDGSAAN LVPEVNNEVMA	5 28	NDANPSDGSAANL VPEVNNEVMALEP VVGAAIAAPVAGQ QNVIDPWIRN	6 54	NDANPSDGSAANL VPEVNNEVMALEP	6 31	26	5
8	2-VP1 (S domain)	DGSAANLVPEVNN EVMALEPVVGAAIA APVAGQQNVIDPW IRNN	12 55	DGSAANLVPEVNN EVM	12 27	DGSAANLVPEVNN EVMALEPVVGAAIA APVA	12 42	DGSAANLVPEVNN EVM	12 27	DGSAANLVPEVNN EVMALEPVVGAAIA APVAGQQNVIDPW IRNN	12 55	DGSAANLVPEVNN EVM	12 27	16	5
9	3-VP1 (S domain)	NNEVMALEPVVGA AIAAPVAGQQNVID PWIRNN	23 55	VNNEVMALEPVVG AAIAAPVAGQQNVI DPWIRNN	22 55	VNNEVMALEPVVG AALAAPVAGQQNVI DPWIR	22 53	VNNEVMALEPVVG AAIAAPVAGQQNVI D	22 49	VNNEVMALEPVVG AAIAAPVAGQQNVI DPWIRNNF	22 56	NNEVMALEPVVGA AIAAPVAGQQNVID	23 49	27	5
10	4-VP1 (S domain)	EPVVGAAIAAPVA GQQNVIDPWIRNN F	30 56	LEPVVGAAIAAPVA GQQNVIDPWIRN	29 54	LEPVVGAAIAAPVA GQQNVIDPWIRN FVQA	29 59	LEPVVGAAIAAPVA GQQNVIDPWIRN	29 54	EPVVGAAIAAPVA GQQNVIDPWIRNN FVQ	30 58	EPVVGAAIAAPVA GQQNVIDPWIRN	30 54	25	5
11	5-VP1 (S domain)	QNVIDPWIRNNFV QA	45 59	QQNVIDPWIRNNF VQAPGGEFTV	44 66	QNVIDPWIRNNFV QAPGGEF	45 64	QQQNVIDPWIRNN FVQAPG	43 61	QNVIDPWIRNNFV QAPGG	45 62	QNVIDPWIRNNFV QA	45 69	15	5
12	6-VP1 (S domain)	IRNNFVQAPGGE	52 63	IRNNFVQAPGGEF TVSPRNAPGEILW SA	52 79	IRNNFVQAPGGEF TVS	52 67	IRNNFVQAPGGEF TVSPRNAPGEILW SA	52 79			IRNNFVQAPGGE	52 63	12	4
13	7-VP1 (S domain)									GGEFTVSPRNAPG EILWSA	61 79	GGEFTVSPRNAPG EILWSA	61 79	19	1
14	8-VP1 (S domain)			RNAPGEILWSAPL GPDLNPYL	69 89							RNAPGEILWSAPL GPDLNPYL	69 89	21	2
15	9-VP1 (S domain)			LWSAPLGPDLNPY LSHLARMYN	76 97							LWSAPLGPDLNPY LSHLARMYN	76 97	22	1
16	10-VP1 (S domain)			GNAFTAGKIIFAAV PPNFPTEGLSPSQ VTMFPHIIVDVRQL EPVLI	111 156	GNAFTAGKIIFAAV PPNFPTEGLSPSQ VTMFPHIIVD	111 147	GNAFTAGKIIFAAV PPNFPTEGLSPSQ VTMFPHIIVDVR	111 149			GNAFTAGKIIFAAV PPNFPTEGLSPSQ VTMFPHIIVD	111 147	37	4
17	11-VP1 (P1 domain)					GRCTTDGVLLGTT QLSPVNICTFRGDV THAGSR	264 297	GRCTTDGVLLGTT QLSPVNICTFRGDV THAGSRNYTMNL	264 303			GRCTTDGVLLGTT QLSPVNICTFRGDV THAGSR	264 297	34	2
18	12-VP1 (P2 domain)					NNYDPTEEIPAPLG TPDFVGKIQGLLT QTTKGDGSTRGHK AT	309 350	NNYDPTEEIPAPLG TPDFVGKIQGLLT QTTKGDGSTRGHK	309 348			NNYDPTEEIPAPLG TPDFVGKIQGLLT QTTKGDGSTRGHK	309 348	40	2
19	13-VP1 (P2 domain)			TQTTKGDGSTRGH KATVYTGSAP	335 357			TQTTKGDGSTRGH KATVYTGSAP	335 357			TQTTKGDGSTRGH KATVYTGSAP	335 357	23	2
20	14-VP1 (P2domain)			TKGDGSTRGHKAT VYTGSAPFTPKLG S	338 364	TKGDGSTRGHKAT VYTGSAPFTPKLG S	338 364	TKGDGSTRGHKAT VYTGSAPFTPKLG S	338 364			TKGDGSTRGHKAT VYTGSAPFTPKLG S	338 364	27	3
21	15-VP1 (P2 domain)					VHNVHLAPAVAPTF PGEQLLFF	413 434					VHNVHLAPAVAPTF PGEQLLFF	413 434	22	1
22	16-VP1 (P1 domain)			PGCSGYPNMDLD CLLPQEWVQHFY QEAAPAQ	439 469							PGCSGYPNMDLD CLLPQEWVQHFY QEAAPAQ	439 469	31	1
23	17-VP1 (P1 domain)			PNMDLDCLLPQEW VQHFYQEAAPAQS DVALLR	445 476							PNMDLDCLLPQEW VQHFYQEAAPAQS DVALLR	445 476	32	1
24	18-VP1 (P1 domain)									LRFVNPDTGRVLF ECKLHKSGYVTVA HTGQHDLVIPPNG YFR	475 516	LRFVNPDTGRVLF ECKLHKSGYVTVA HTGQHDLVIPPNG YFR	475 516	42	1

Supplementary Table 2. List of epitopes from all the sera identified after two rounds of affinity selections with the GII.4 HOV Jun-Fos library. A total of 24 unique epitopes were identified. The sequences and the positions in the GII.4 HOV ORF1 and ORF2 genome of each epitope are included. The shared epitopes amongst the six individuals as well as the epitopes' sequences, positions in the GII.4 HOV genome, length, and the number of sera that contain the epitopes are also included.