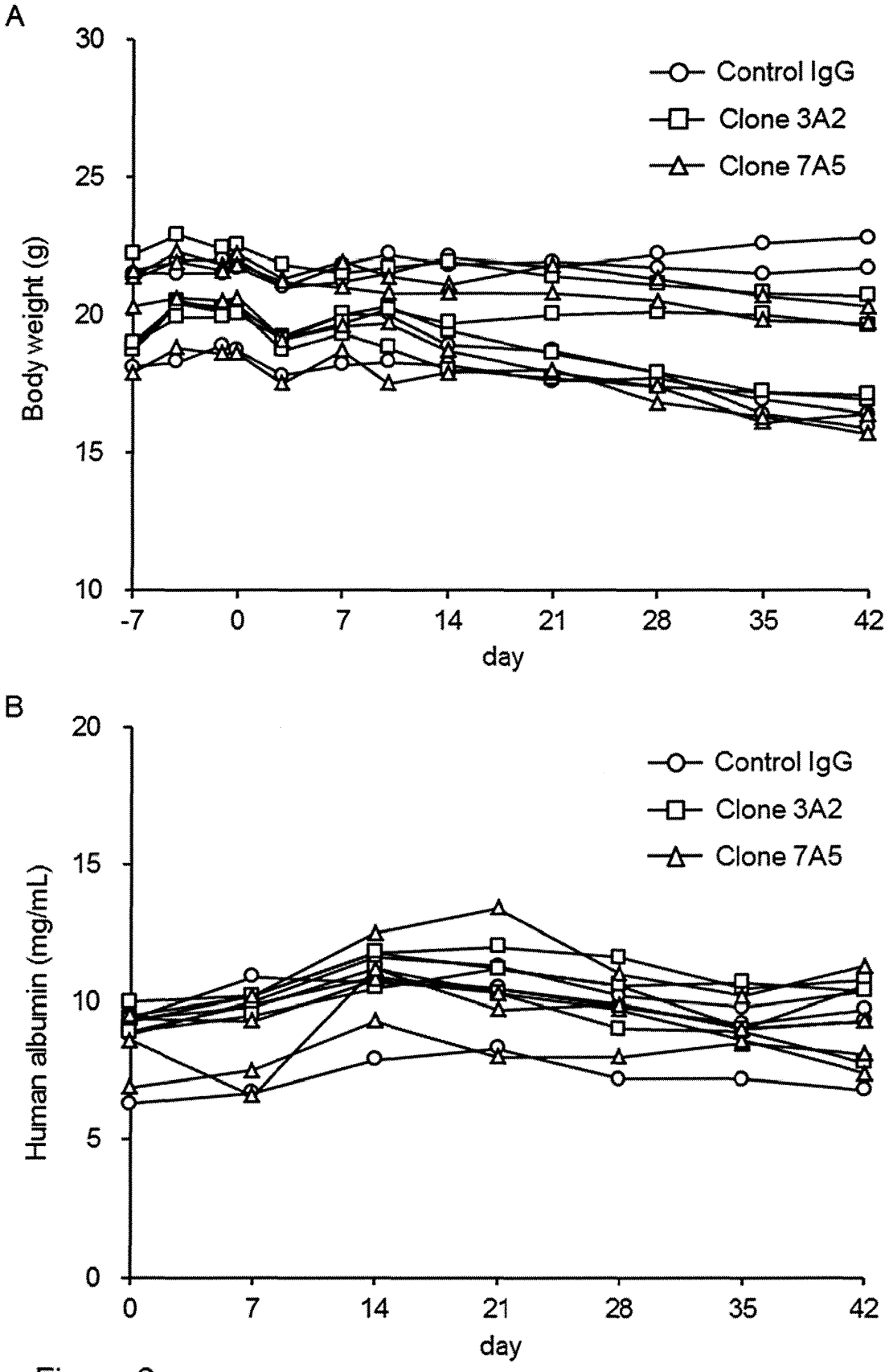
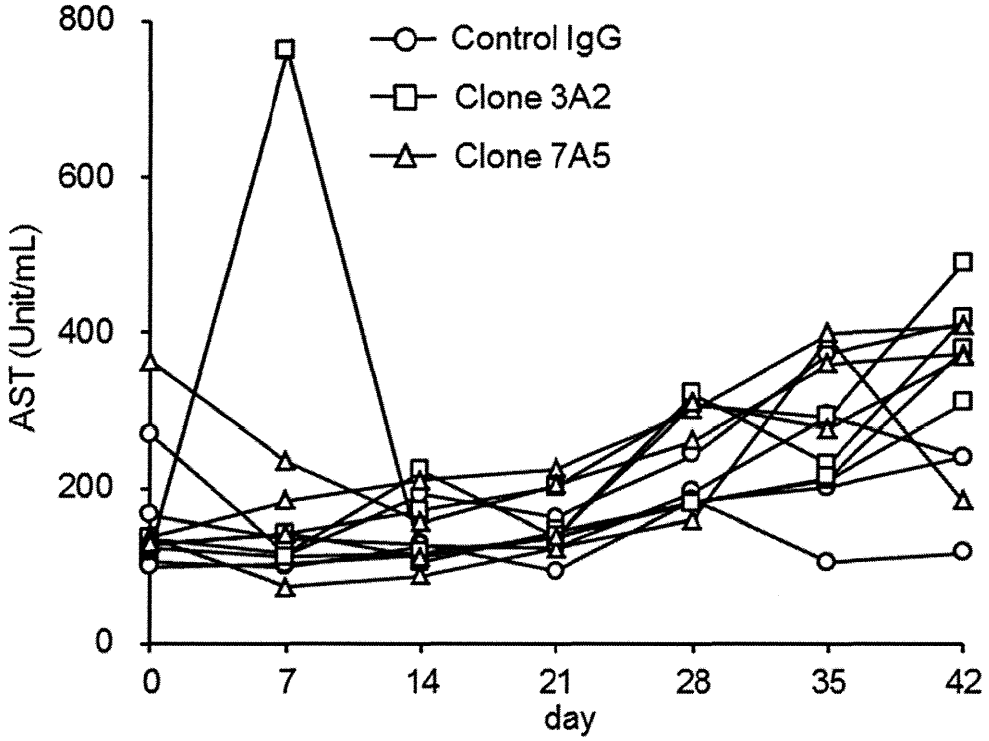


Figure 6
To be continued

C



D

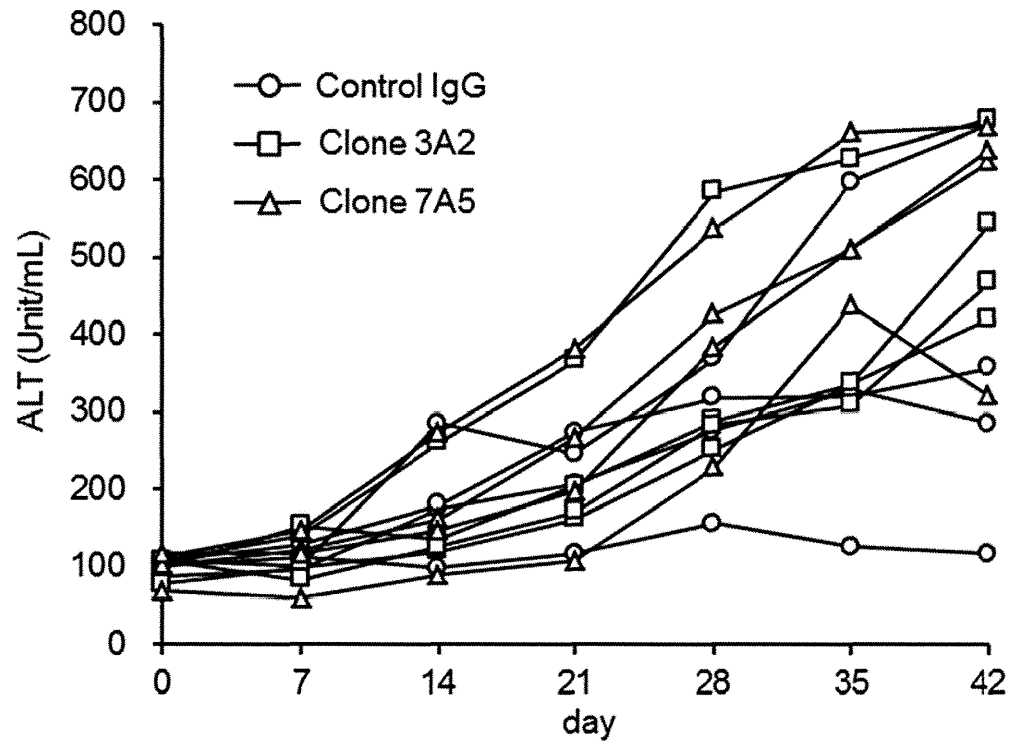


Figure 6 Toxicological tests of uPA-SCID mice administered anti-CL-1 antibodies.

Body weight (A), human albumin (B), AST (C) and ALT (D) were chronologically measured.

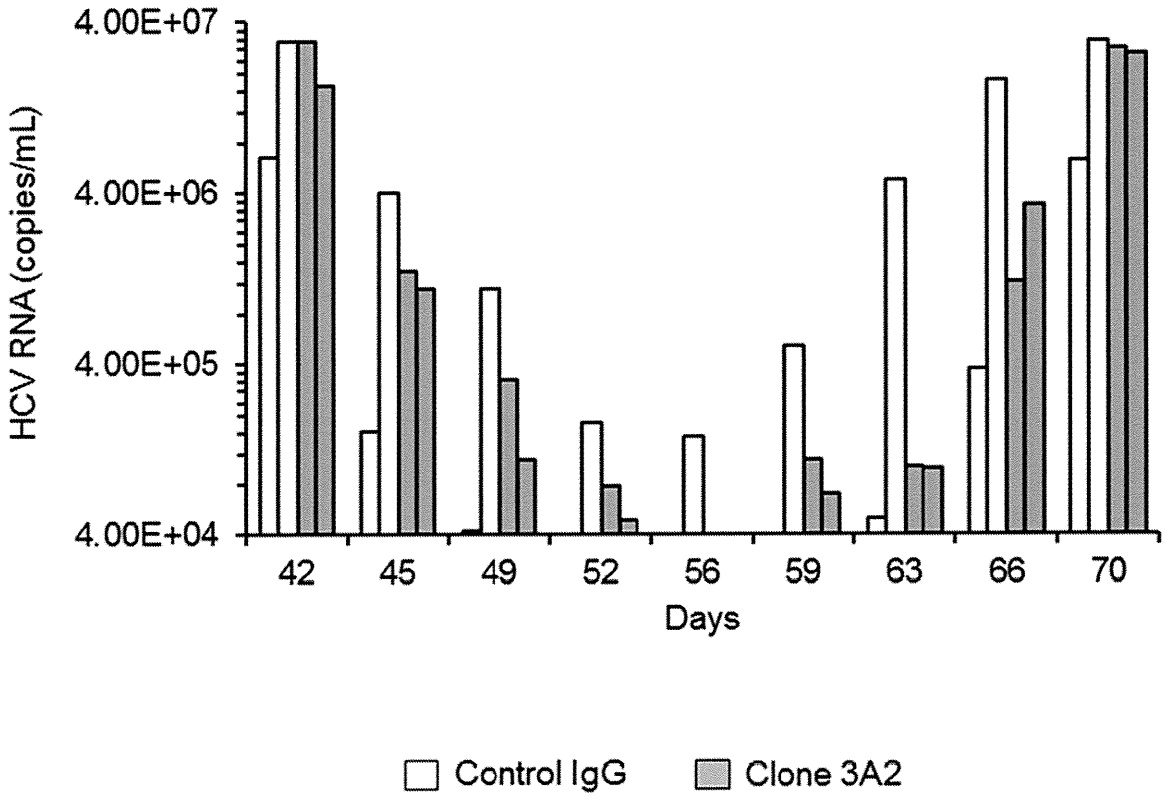
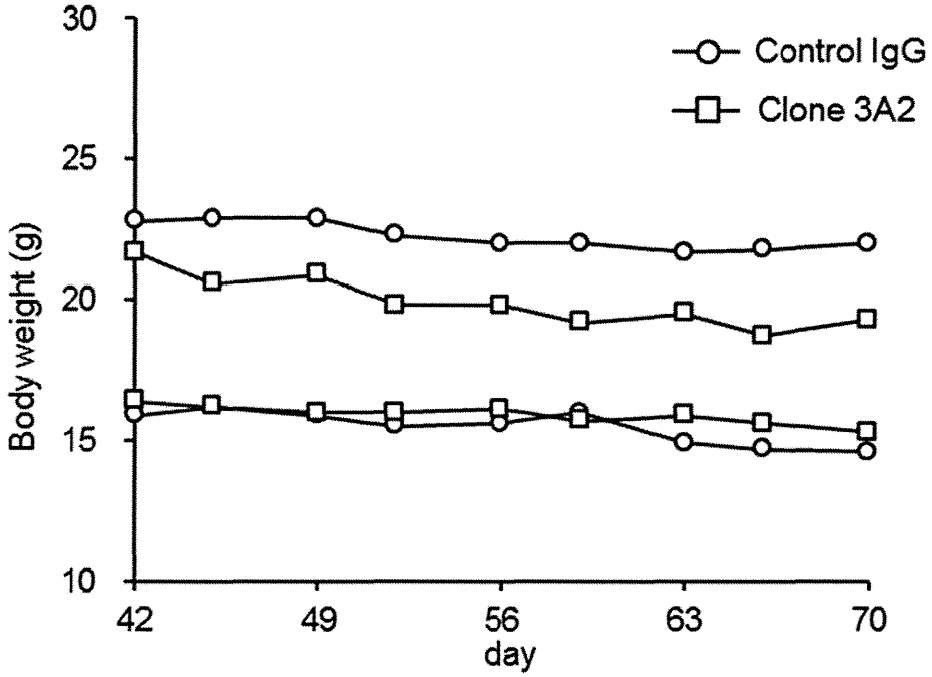


Figure 7 Measurement of HCV RNA in HCV genotype 1b infected uPA-SCID mice administered pegasys and anti-CL-1 antibodies.

A



B

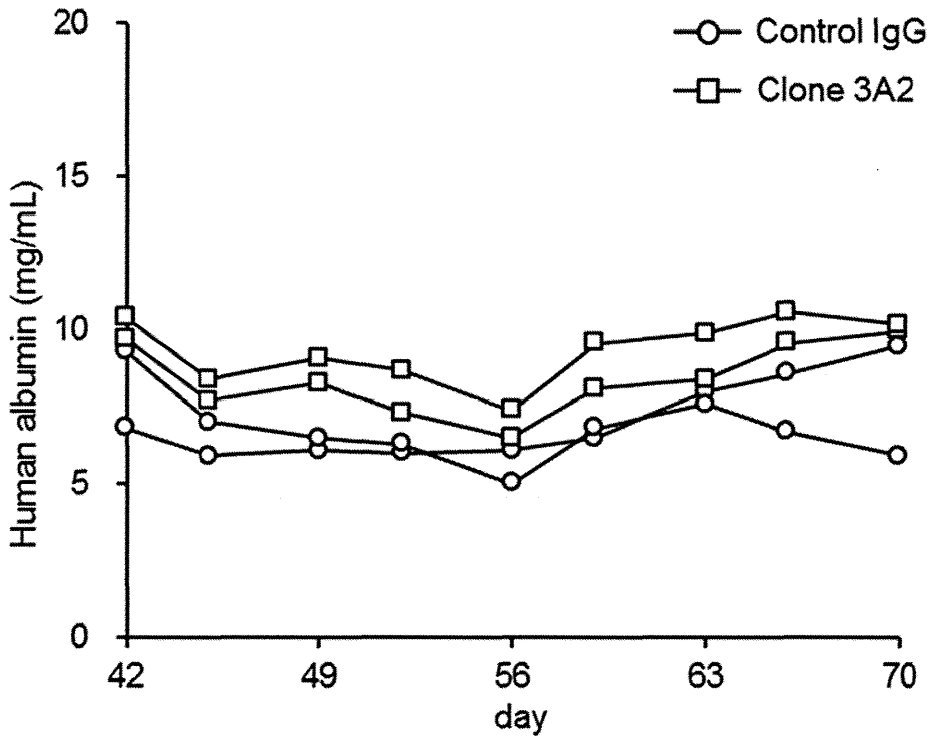
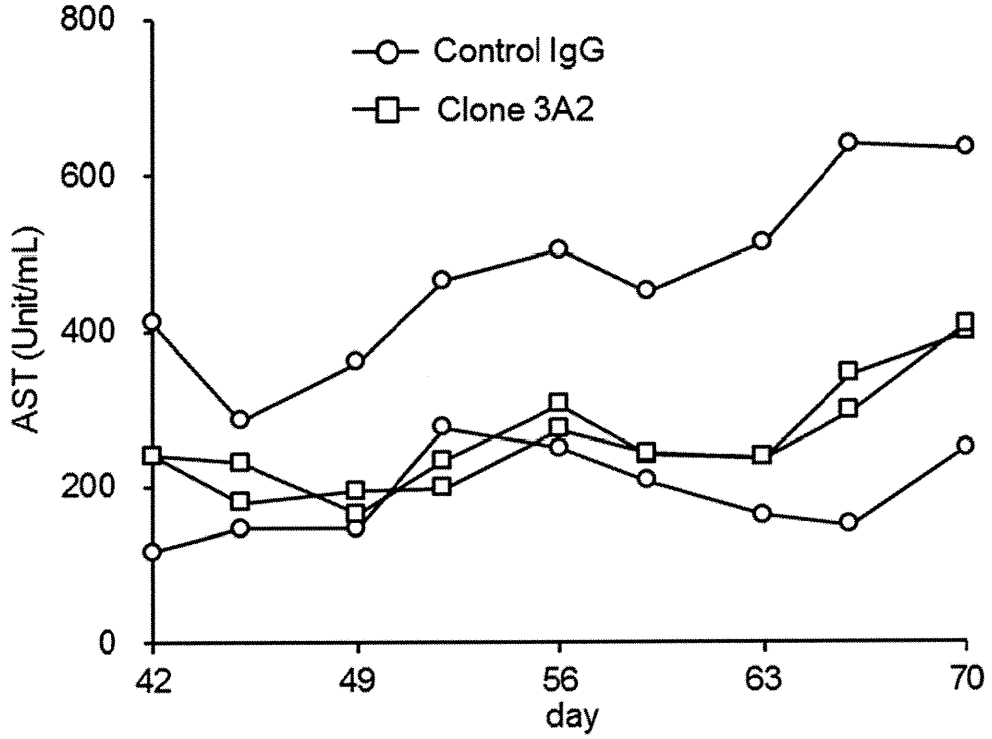


Figure 8
To be continued

C



D

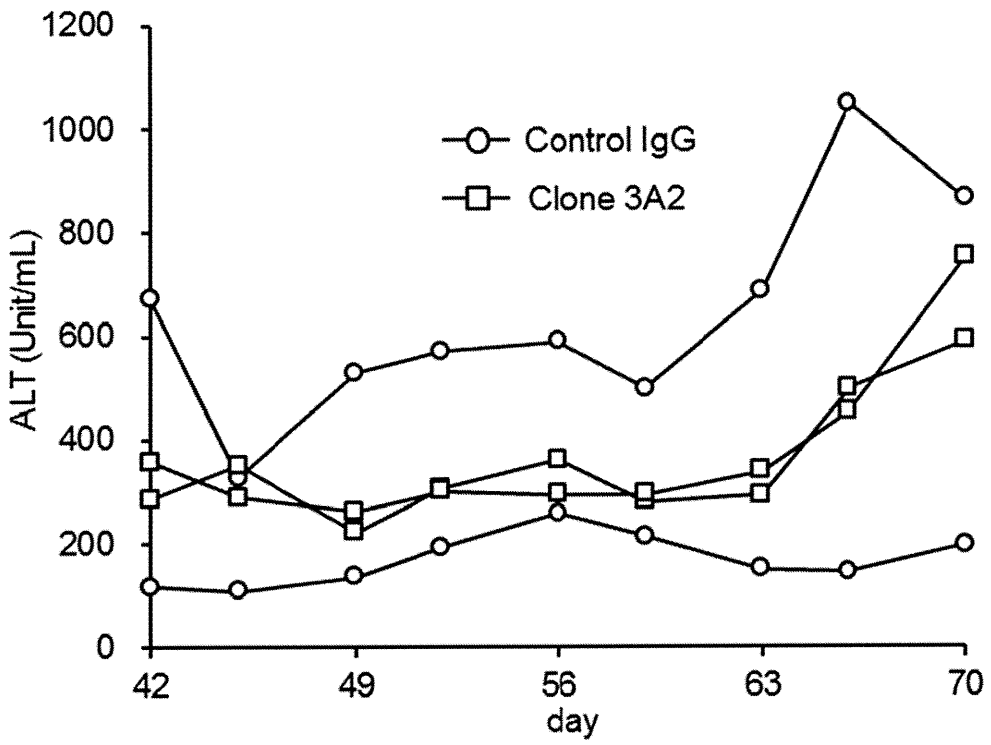


Figure 8 Toxicological tests of uPA-SCID mice administered pegasys and anti-CL-1 antibodies.
Body weight (A), human albumin (B), AST (C) and ALT (D) were chronologically measured.

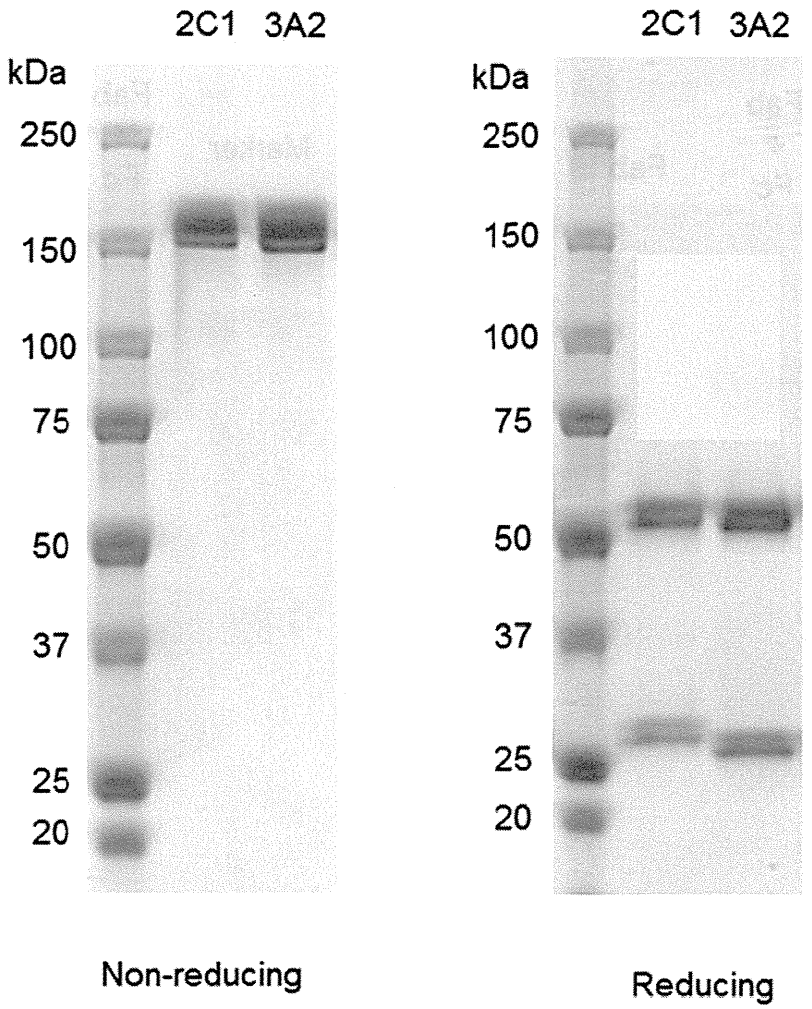


Figure 9 Preparation of anti-CL-1 human–mouse chimeric IgG1 antibodies.

SDS-PAGE under non-reducing or reducing conditions. The molecular weight of the purified product of anti-CL-1 human–mouse chimeric IgG1 antibodies were confirmed.

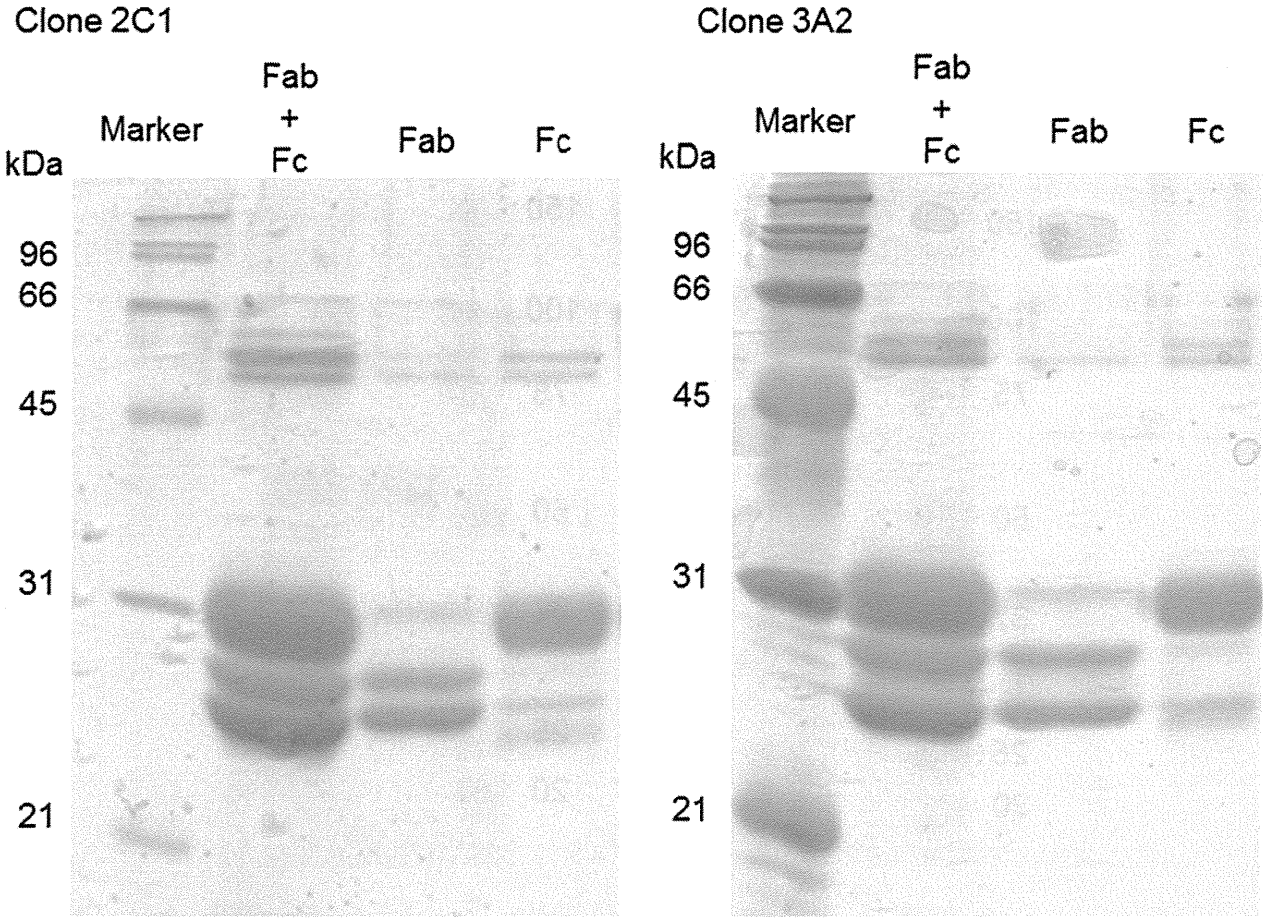


Figure 10 Preparation of Fab fragments.
Purified IgG was digested with papain in Fab digestion buffer. The purity of papain digested IgG into Fab fragments was evaluated by 15% gradient SDS-PAGE. Lane 1: Protein molecular weight marker; Lane 2: Digested IgG; Lane 3: Purified Fab fragments; Lane 4: Fc fragments.

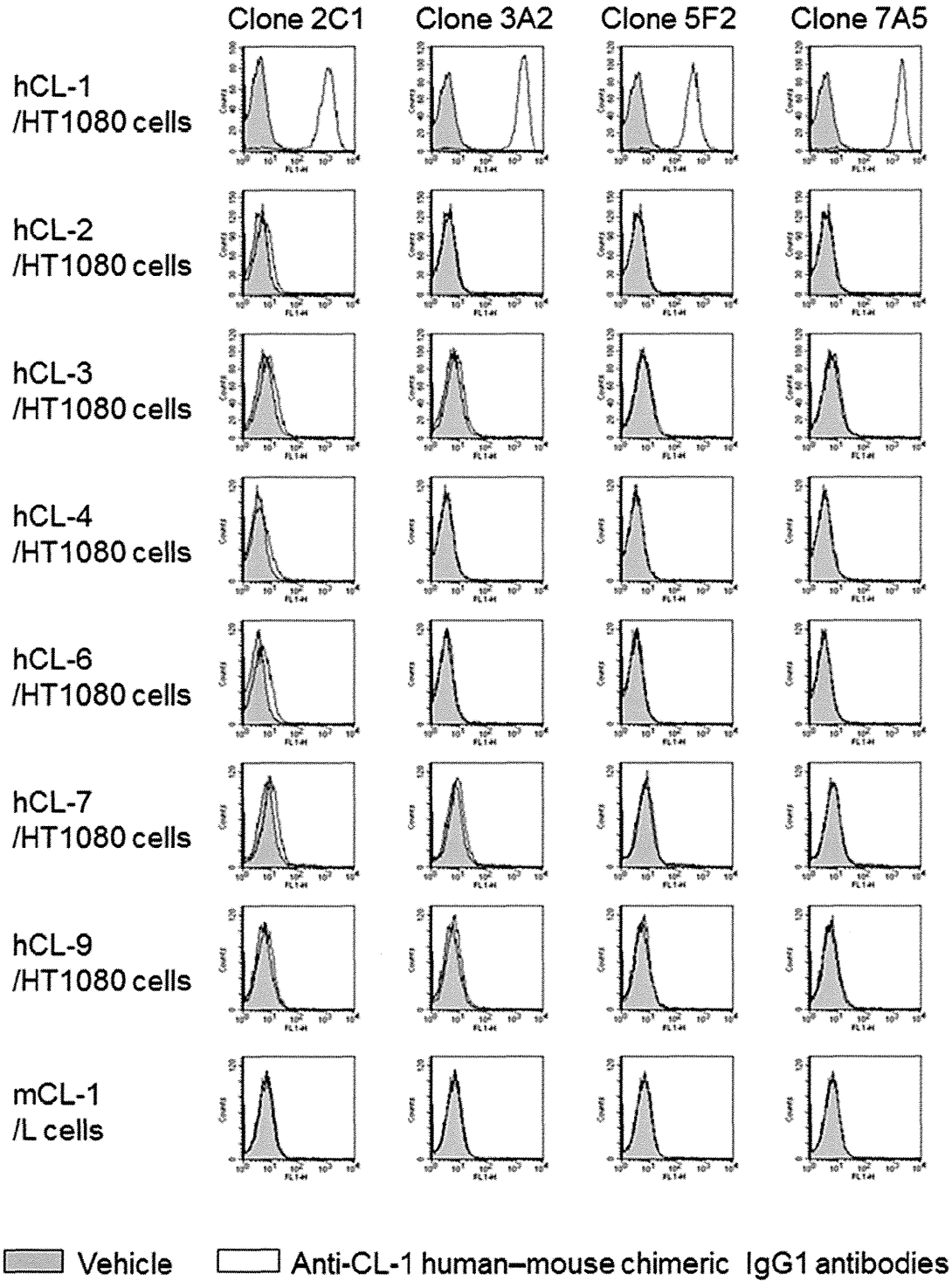


Figure 11 Flow cytometry analysis of anti-CL-1 human–mouse chimeric IgG1 antibodies against CLs-expressing cells.
Each cells were incubated with 5 μg/mL of anti-CL-1 human–mouse chimeric IgG1 antibodies (white histogram) or control (gray histogram), and binding was detected by FITC-conjugated goat anti-human IgG (H+L). As a control, cells were incubated with phosphate buffered saline (PBS).

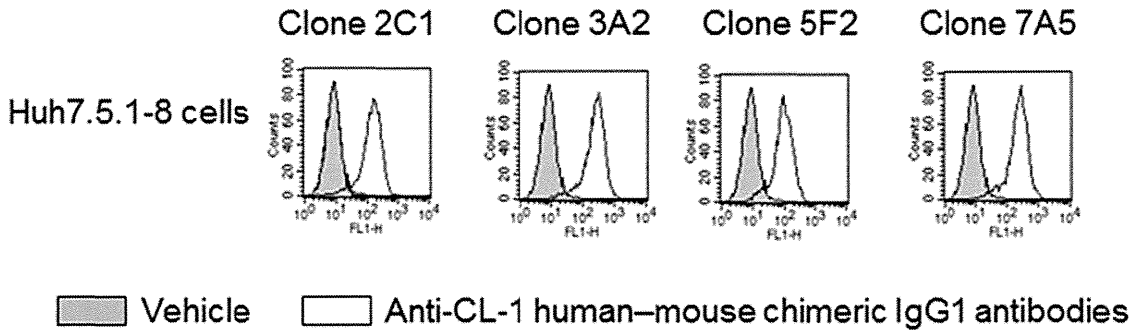


Figure 12 Flow cytometry analysis of anti-CL-1 human–mouse chimeric IgG1 antibodies against Huh7.5.1-8 cells.
Huh7.5.1-8 cells were incubated with 5 μg/mL of anti-CL-1 human–mouse chimeric IgG1 antibodies (white histogram) or control (gray histogram), and binding was detected by FITC-conjugated goat anti-human IgG (H+L). As a control, cells were incubated with phosphate buffered saline (PBS).

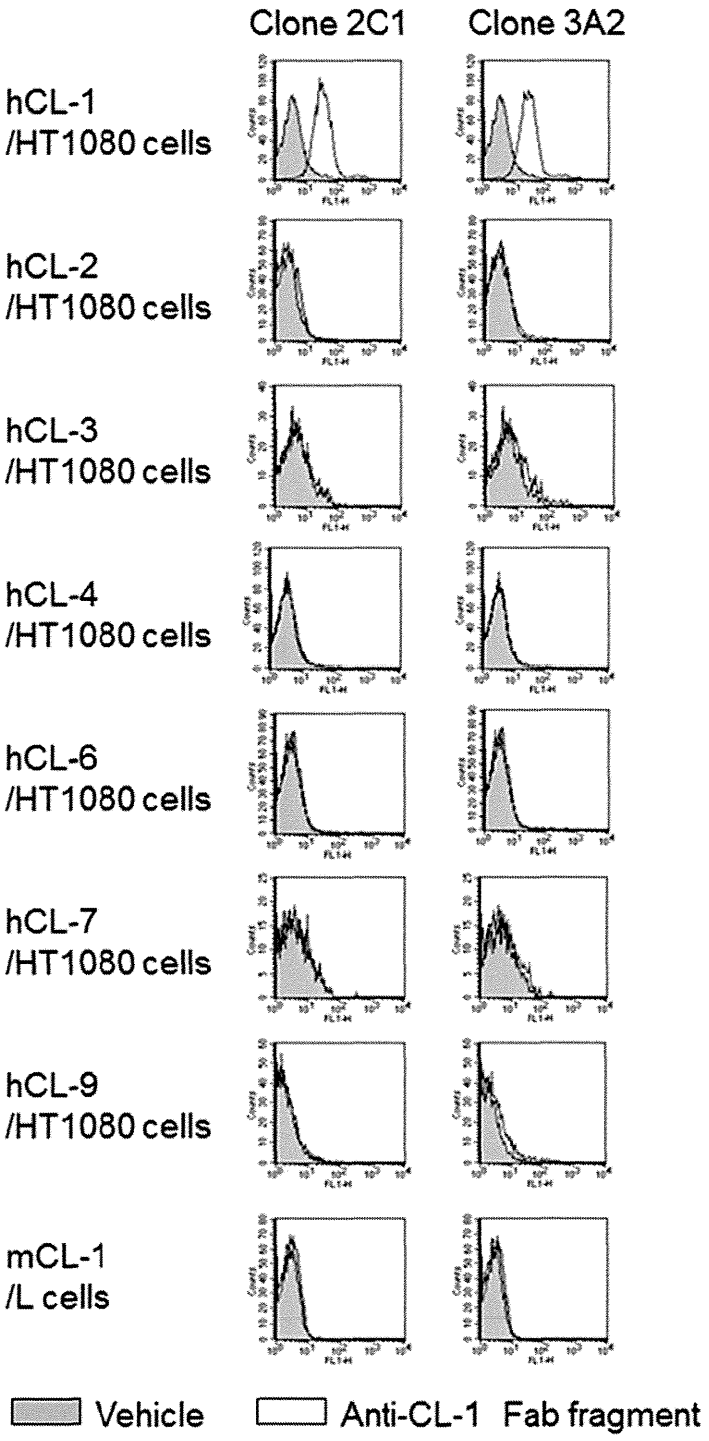


Figure 13 Flow cytometry analysis of anti-CL-1 Fab fragment against CLs-expressing cells.

Each cells were incubated with 5 μg/mL of anti-CL-1 Fab fragment (white histogram) or control (gray histogram), and binding was detected by FITC-conjugated goat anti-human IgG (Fab specific). As a control, cells were incubated with phosphate buffered saline (PBS).

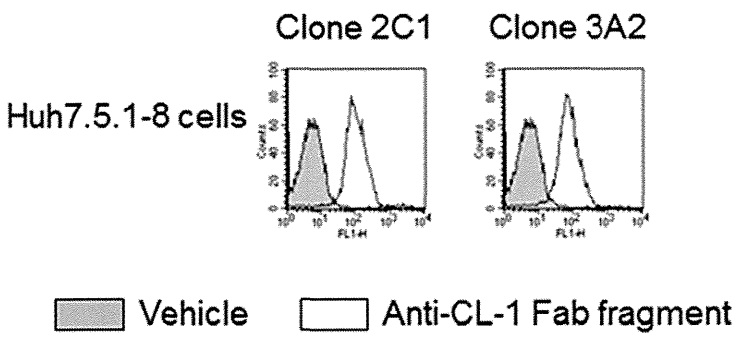


Figure 14 Flow cytometry analysis of anti-CL-1 Fab fragment against Huh7.5.1-8 cells.

Huh7.5.1-8 cells were incubated with 5 μg/mL of anti-CL-1 human–mouse chimeric IgG1 antibodies (white histogram) or control (gray histogram), and binding was detected by FITC-conjugated goat anti-human IgG (H+L). As a control, cells were incubated with phosphate buffered saline (PBS).

A

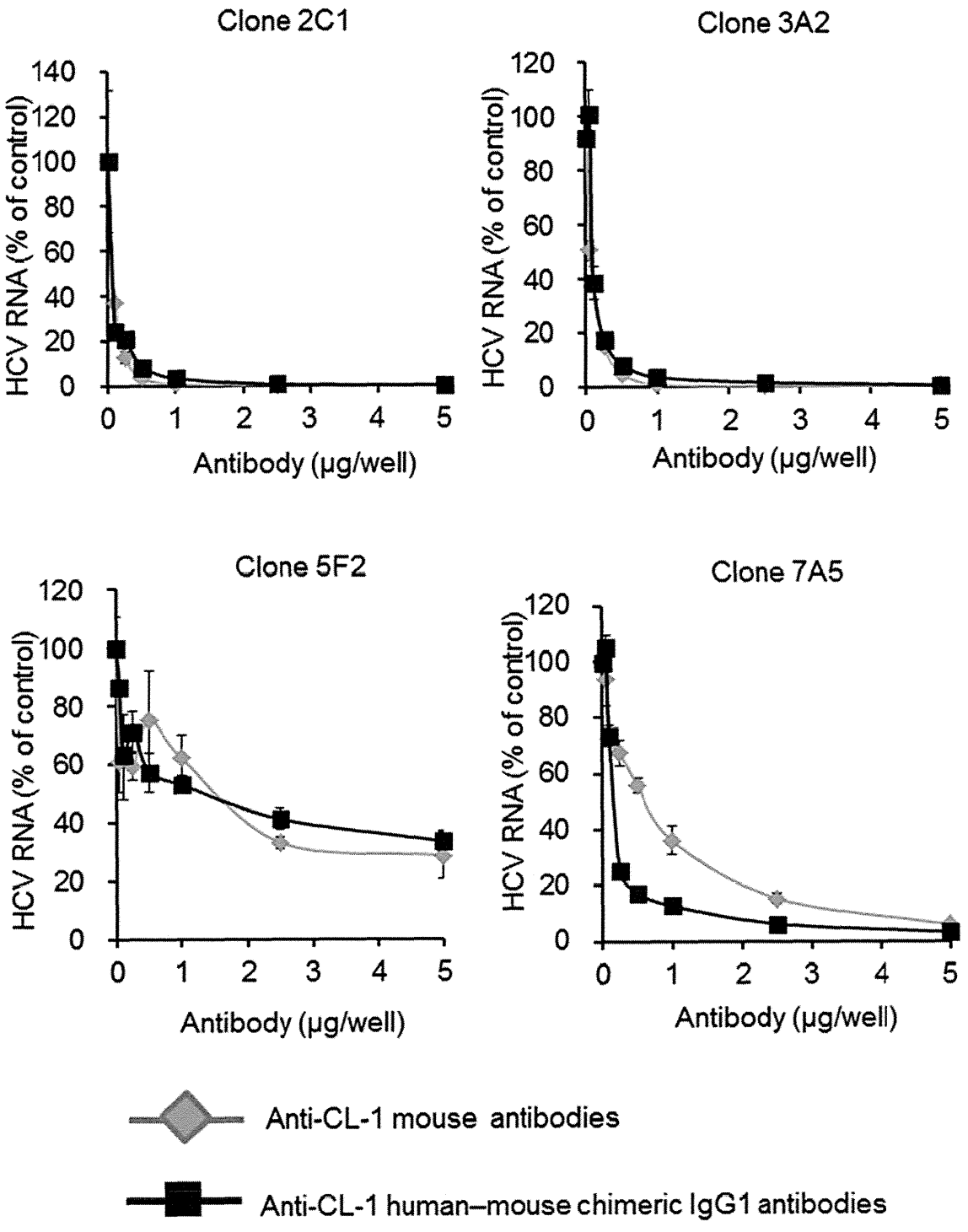


Figure 15
To be continued

B

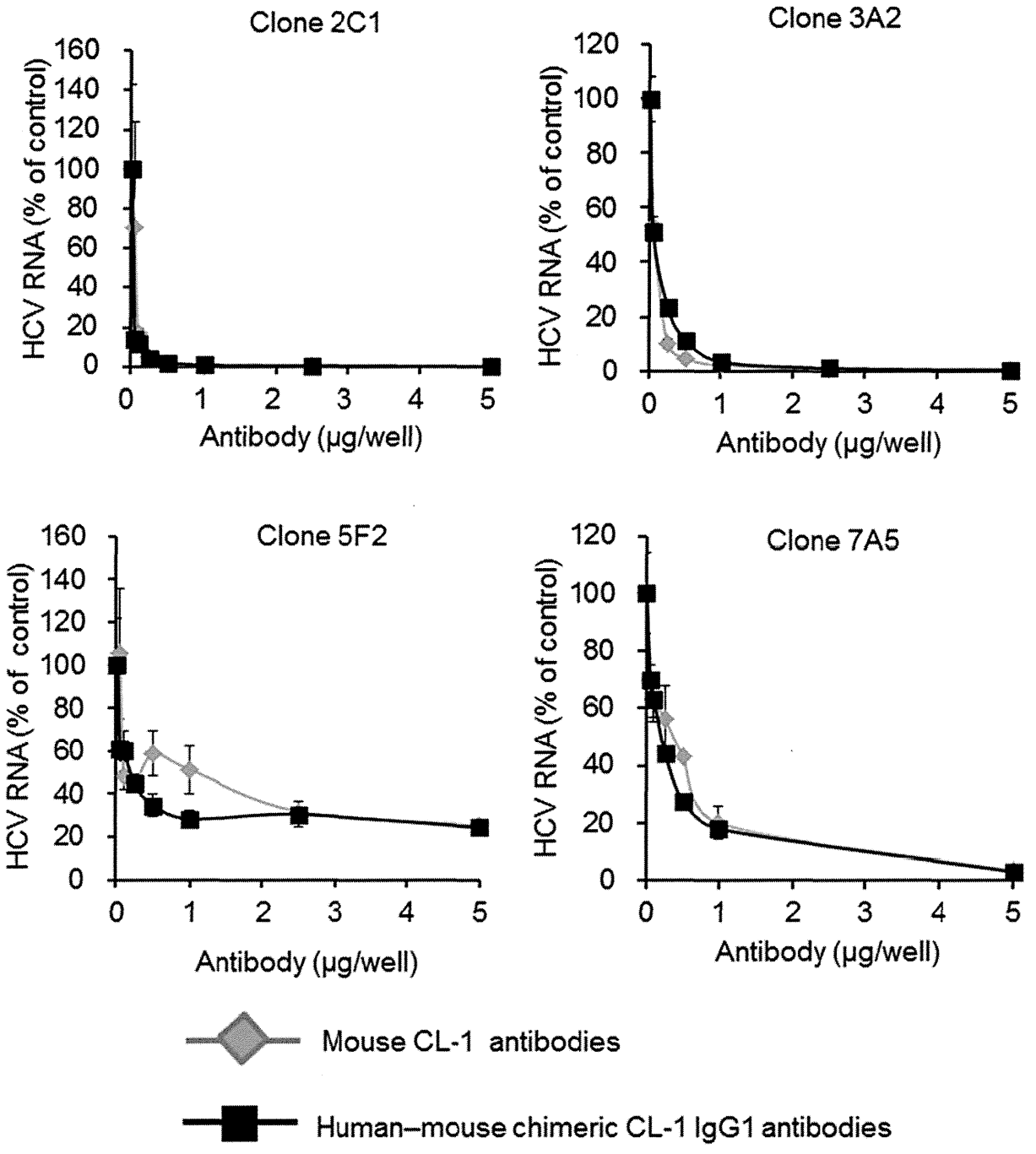


Figure 15 Inhibition of HCVcc (genotype 2a) infection by anti-CL-1 mouse antibodies or human–mouse chimeric IgG1 antibodies. Huh7.5.1-8 cells were pre-incubated with increasing concentration of anti-CL-1 antibodies for 30 minutes at room temperature before infection with HCVcc. After culturing for 4 days, HCV RNA within the cell (A) and in the culture supernatant (B) were measured. Data are shown as mean ± SD (n=4), percentage of HCV RNA copies of non-treated group.

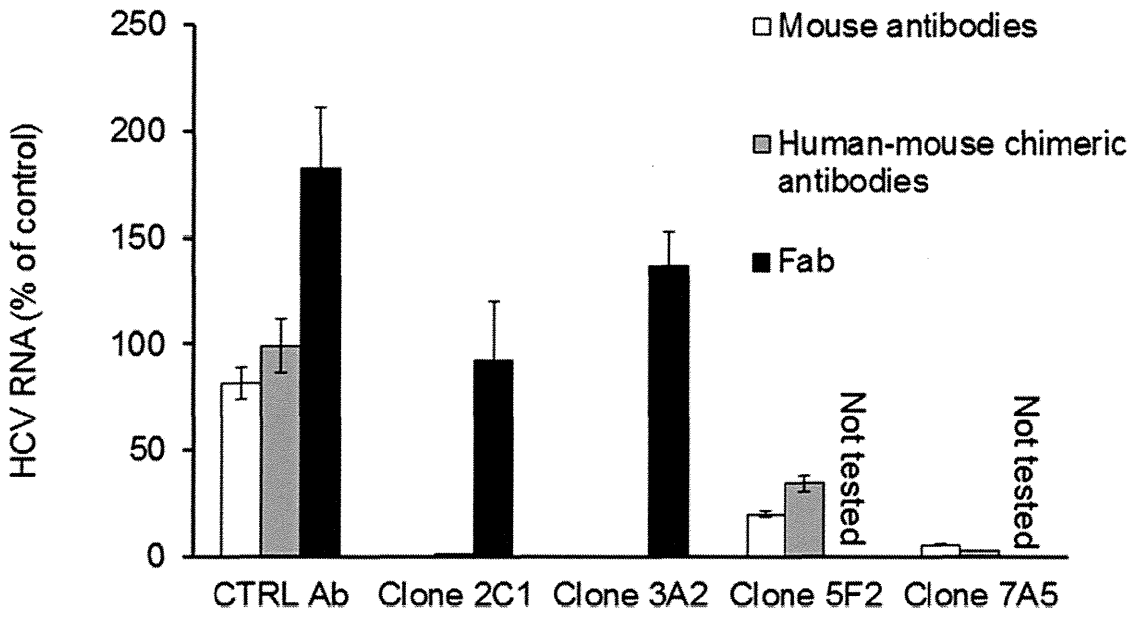


Figure 16 Inhibition of HCVcc (genotype 2a) infection by anti-CL-1 human–mouse chimeric IgG1 antibodies or anti-CL-1 Fab.
Huh7.5.1-8 cells were pre-incubated with 5 μg/well (12.5 μg/mL) of anti-CL-1 antibodies or control antibodies for 30 minutes at room temperature before infection with HCVcc. After culturing for 4 days, HCV RNA within the cell were measured. Data are shown as mean ± SD (n=4), percentage of HCV RNA copies of non-treated group.

A

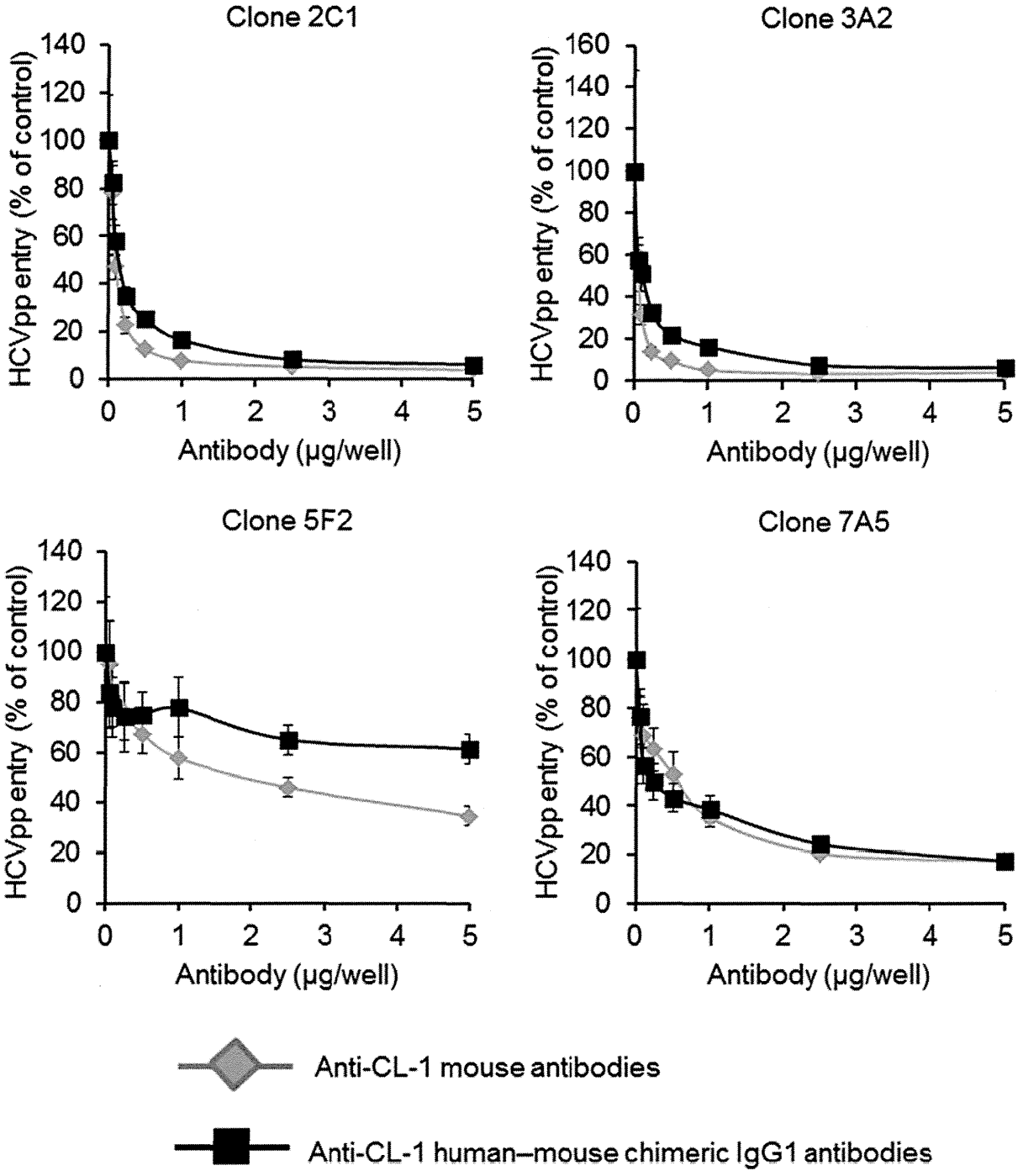


Figure 17
To be continued

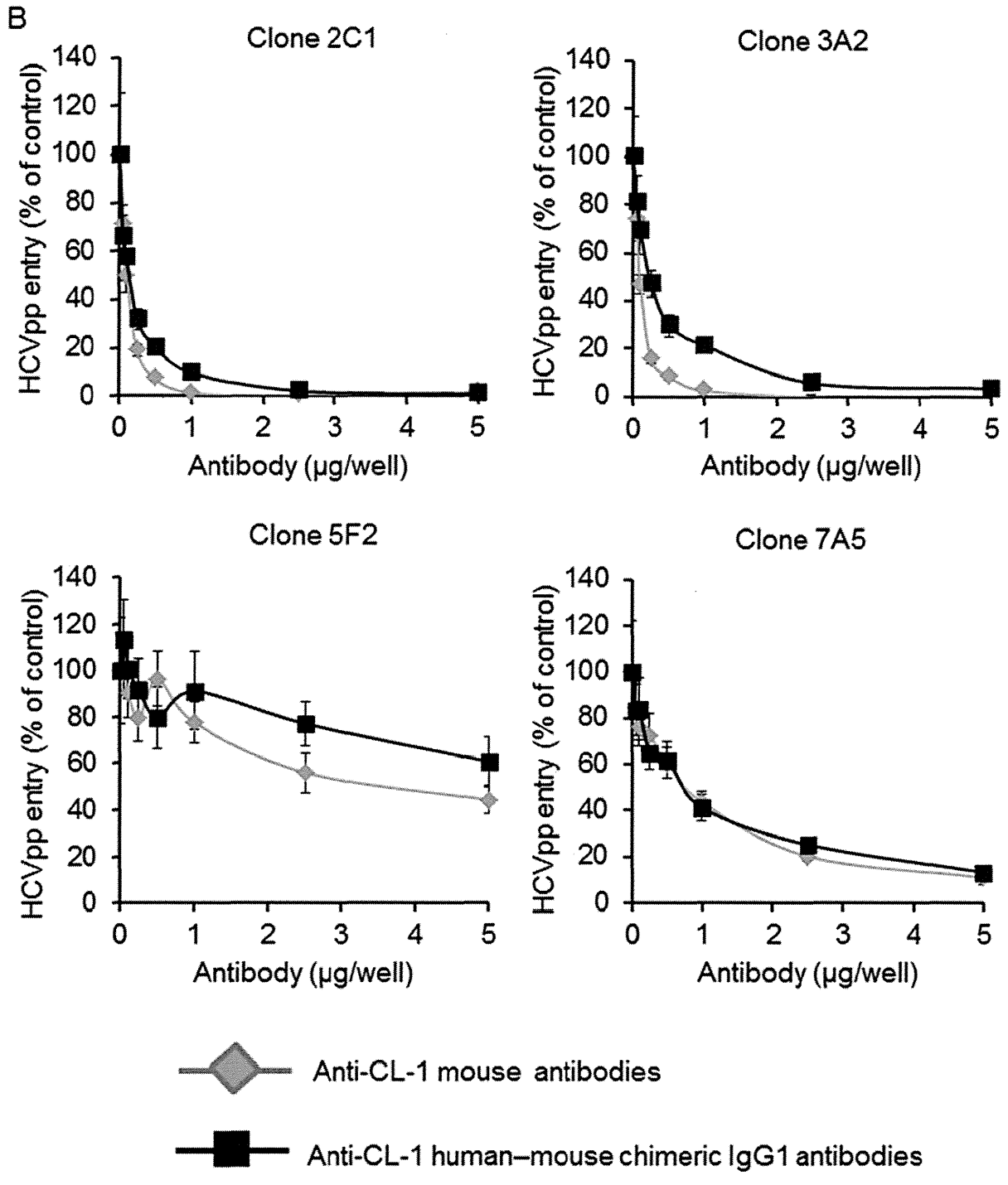


Figure 17 Inhibition of HCVpp infection by anti-CL-1 mouse antibodies or human–mouse chimeric IgG1 antibodies.
Huh7.5.1-8 cells were pre-incubated with increasing concentration of anti-CL-1 antibodies for 30 minutes at room temperature before infection with HCVpp (genotype 1b (A), 2a (B)). After culturing for 3 days, luciferase activity was determined. Data are shown as mean ± SD (n=4), percentage of amount observed of non-treated group.

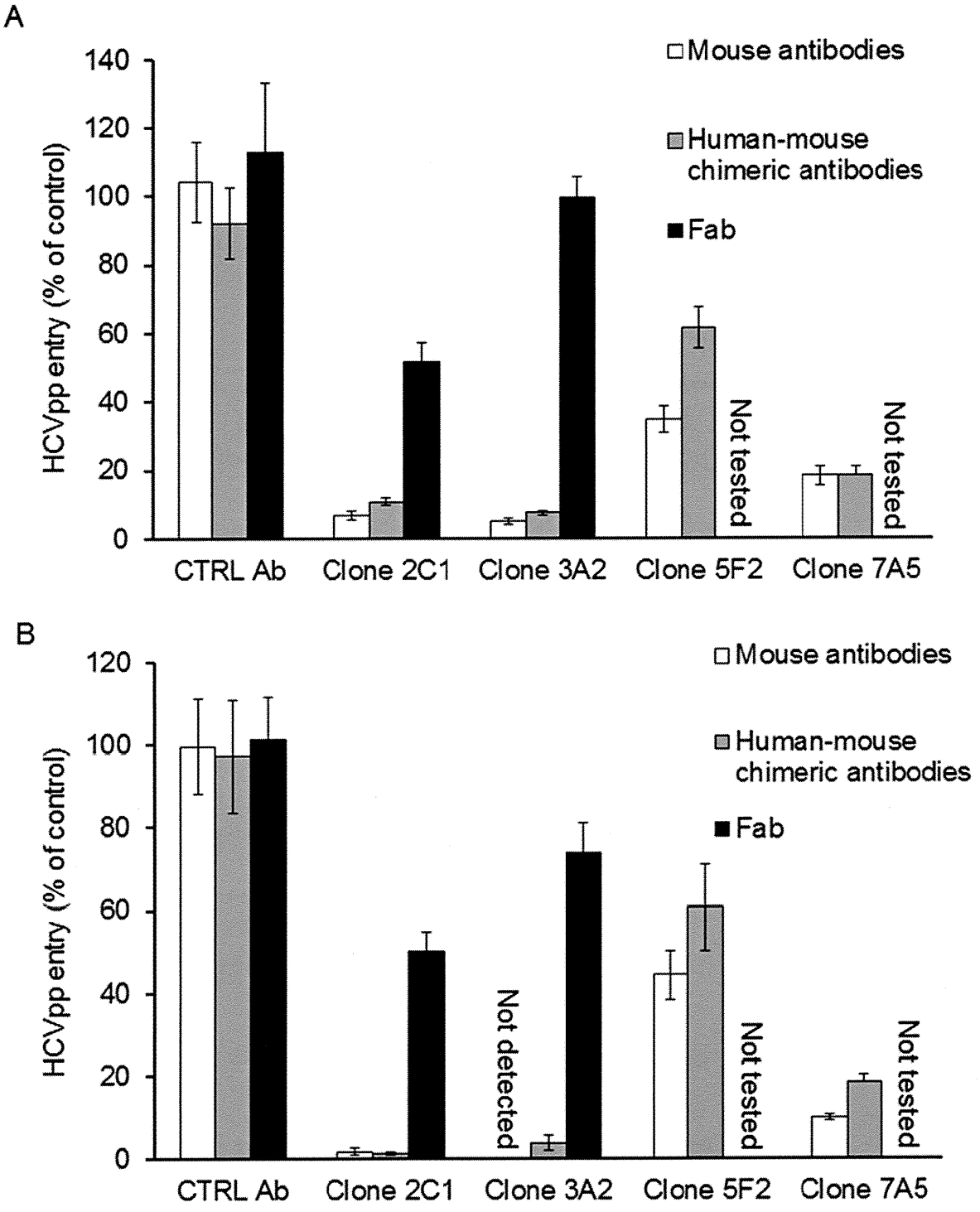


Figure 18 Inhibition of HCVpp infection by anti-CL-1 human–mouse chimeric IgG1 antibodies or anti-CL-1 Fab.

Huh7.5.1-8 cells were pre-incubated with 5 μg/well (12.5 μg/mL) of anti-CL-1 antibodies or control antibodies for 30 minutes at room temperature before infection with HCVpp (genotype 1b (A), 2a (B)). After culturing for 3 days, luciferase activity was determined. Data are shown as mean ± SD (n=4), percentage of amount observed of non-treated group.

Table 3. Amino acid sequence of peptide spot array of human claudin-4.

Spot number	Position of human claudin-4	Sequence
D1	1-15	MASMGLQVMGIALAV
D2	4-18	MGLQVMGIALAVLGW
D3	7-21	QVMGIALAVLGWLAV
D4	10-24	GIALAVLGWLAVMLC
D5	13-27	LAVLGWLAVMLCCAL
D6	16-30	LGWLAVMLCCALPMW
D7	19-33	LAVMLCCALPMWRVT
D8	22-36	MLCCALPMWRVTAFI
D9	25-39	CALPMWRVTAFIGSN
D10	28-42	PMWRVTAFIGSNIVT
D11	31-45	RVTAFIGSNIVTSQT
D12	34-48	AFIGSNIVTSQTIWE
D13	37-51	GSNIVTSQTIWEGLW
D14	40-54	IVTSQTIWEGLWMNC
D15	43-57	SQTIWEGLWMNCVVQ
D16	46-60	IWEGLWMNCVVQSTG
D17	49-63	GLWMNCVVQSTGQMQ
D18	52-66	MNCVVQSTGQMQCKV
D19	55-69	VVQSTGQMQCKVYDS
D20	58-72	STGQMQCKVYDSLLA
D21	61-75	QMQCKVYDSLLALPQ
D22	64-78	CKVYDSLLALPQDLQ
D23	67-81	YDSLLALPQDLQAAR
D24	70-84	LLALPQDLQAARALV

Table 3
To be continued

Table 3. Amino acid sequence of peptide spot array of human claudin-4.

Spot number	Position of human claudin-4	Sequence
E1	73-87	LPQDLQAARALVIIS
E2	76-90	DLQAARALVIISIIV
E3	79-93	AARALVIISIIVAAL
E4	82-96	ALVIISIIVAALGVL
E5	85-99	IISIIVAALGVLLSV
E6	88-102	IIVAALGVLLSVVGG
E7	91-105	AALGVLLSVVGGKCT
E8	94-108	GVLLSVVGGKCTNCL
E9	97-111	LSVVGGKCTNCLEDE
E10	100-114	VGGKCTNCLEDESAK
E11	103-117	KCTNCLEDESAKAKT
E12	106-120	NCLEDESAKAKTMIV
E13	109-123	EDESAKAKTMIVAGV
E14	112-126	SAKAKTMIVAGVVFL
E15	115-129	AKTMIVAGVVFLLAG
E16	118-132	MIVAGVVFLLAGLMV
E17	121-135	AGVVFLLAGLMVIVP
E18	124-138	VFLLAGLMVIVPVSW
E19	127-141	LAGLMVIVPVSWTAH
E20	130-144	LMVIVPVSWTAHNII
E21	133-147	IVPVSWTAHNIIQDF
E22	136-150	VSWTAHNIIQDFYNP
E23	139-153	TAHNIIQDFYNPLVA
E24	142-156	NIIQDFYNPLVASGQ

Table 3
To be continued

Table 3. Amino acid sequence of peptide spot array of human claudin-4.

Spot number	Position of human claudin-4	Sequence
F1	145-159	QDFYNPLVASGQKRE
F2	148-162	YNPLVASGQKREMGA
F3	151-165	LVASGQKREMGASLY
F4	154-168	SGQKREMGASLYVGW
F5	157-171	KREMGASLYVGWAAS
F6	160-174	MGASLYVGWAASGLL
F7	163-177	SLYVGWAASGLLLLG
F8	166-180	VGWAASGLLLLGGGL
F9	169-183	AASGLLLLGGGLLCC
F10	172-186	GLLLLGGGLLCCNCP
F11	175-189	LLGGGLLCCNCPPRT
F12	178-192	GGLLCCNCPPRTDKP
F13	181-195	LCCNCPPRTDKPYSA
F14	184-198	NCPPRTDKPYSAKYS
F15	187-201	PRTDKPYSAKYSAAR
F16	190-204	DKPYSAKYSAARSAA
F17	193-207	YSAKYSAARSAAASN
F18	195-209	AKYSAARSAAASNYV