

		spontaneous resolvers		acutely infected		chronic patients	
number of patients		16	%	16	%	108	%
sex	female	5	31.3	7	43.8	31	28.7
	male	11	68.8	9	56.3	77	71.3
Age	>40	7	43.8	5	31.3	35	32.4
	<40	9	56.3	11	68.8	73	67.6
Ethnicity	Caucasian	16	100.0	15	93.8	87	80.6
	Asian					16	14.8
	Hispanic			1	6.3	1	0.9
	not known					4	3.7
Risk factor	IVDU	11	68.8	9	56.3	70	64.8
	unkown	1	6.3			14	13.0
	BP			1	6.3	12	11.1
	cocaine					6	5.6
	tattoo	2	12.5	1	6.3	3	2.8
	needlestick	1	6.3			2	1.9
	sexual	1	6.3	5	31.3	0	0.0
	vertical					1	0.9
HCV subtype	Genotype 3		N/A	8	50.0	14	13.0
	Genotype 3a		N/A	8	50.0	94	87.0
Viral load	IU/ml (mean)		N/A	1827490		872828	
	IU/ml (range)		N/A	6290-8930000		3614-17880410	
Outcome	cleared spontaneously	16	100.0	4	25.0	0	0.0
	chronic	0	0.0	10	62.5	108	100.0
	not treated	N/A		6	60.0	30	27.8
	treated	N/A		4	40.0	78	72.2
Treatment	unkown			2	12.5		
	SVR			2	50.0	42	53.8
	REL			1	25.0	25	32.1
	NR			1	25.0	6	7.7
	incomplete					3	3.8
	not known					2	2.6

Table S1: Patient details of patients with spontaneously resolved, acute and chronic HCV genotype-3 infection.

ID: patient identifier; IVDU: intravenous drug use; BP: blood products; SVR: sustained virological response; REL: viral relapse; NR: treatment non-response.

ID	Age	Sex	Race	Risk factor	GT	HCV VL IU/ml	ALT	Treatment	outcome
A1	23	F	W	Sexual	3	>700000	68	naïve	cleared spontaneously
A2	31	M	W	IVDU	3a	6290	not known	naïve	cleared spontaneously
A3	23	M	W	IVDU	3a	8930000	46	naïve	cleared spontaneously
A4	21	M	W	IVDU	3	15900	72	naïve	cleared spontaneously
A5	19	M	W	IVDU	3	168000	229	naïve	chronic
A6	20	F	W	IVDU	3	16100	38	naïve	chronic
A7	46	M	W	blood exposure	3	204000	592	naïve	chronic
A8	28	M	W	Sexual	3	136000	698	naïve	chronic
A9	45	F	W	Tattoo	3a	>700000	201	naïve	chronic
A10	21	F	W	IVDU	3a	600000	not known	naïve	chronic
A11	18	F	W	IVDU	3	102000	88	naïve	lost in <6 months
A12	25	F	W	IVDU	3a	not known	21	naïve	lost in <6 months
A13	40	F	W	Sexual	3a	>700000	464	treated	cleared on Tx
A14	41	M	H	Sexual	3a	67535	250	treated	cleared on Tx
A15	49	M	W	Sexual	3a	46603	1441	treated	cleared on Tx
A16	20	M	W	IVDU/Tattoo	3	>700000	383	treated	chronic (non-responder)

Table S2: Patient details acute HCV genotype-3 cohort.

ID: patient identifier; M: male; F: female; W: white; H: Hispanic; IVDU: intravenous drug use; GT: genotype; ALT: alanine aminotransferase; Tx: treatment.

Protein	HLA	Wildtype	Variant	Amino Acids	Peptide ID	Predicted Epitope
NS2	A0101	V	A	879-887	001-A0101	V ILLT S LLY
	A02	L	P	862-871	049-A02	ALQ V WVP P LL
		L	P	870-879	048-A02	LL A RGS R DGV
		Y	H	881-890	052-A02	LL T SLL Y PSL
		Y	H	885-894	047-A02	LL Y PSL I FDI
	A0301	V	A	926-935	006-A0301	RLC M L V RS V M
		V	A	929-938	004-A0301	ML V RS V M G GK
		V	A	930-939	005-A0301	L V RS V M G GK Y
	A2402	Y	H	886-894	007-A2402	L Y PSL I FDI
	B1501	T	A	878-887	008-B1501	GVILL T SLLY
	B15	I	V	946-954	055-B15	S I GR W FN T Y
	B2705	R	K	940-949	010-B2705	FQ M IL S IG R
		R	K	948-956	009-B2705	GR W FN T Y L Y
		H	Y	962-971	011-B2705	MQ H WAA A GLK
	B4402	I	V	822-831	012-B4402	ATL G AG L VL
	B44	I	V	826-835	057-B44	AG I L V LF G FF
	B5101	S	G	871-880	013-B5101	L A RGS R D G VI
C03	L	M	829-838	050-C03	IL V LF G FF T L	
C04	V	I	981-990	051-C04	IF S PM E IK V I	
NS3	A0101	Y	F	1442-1450	014-A0101	ATD A L G MT Y
	A0201	A	D	1389-1398	015-A0201	A LL K GG R H L I
	A0301	V	I	1138-1146	016-A0301	L V TRD A D V I
	B1501	K	R	1296-1305	017-B1501	K L T Y S T Y G K F
	B2705	L	I	1379-1388	018-B2705	I P F Y G K A I P L
		V	I	1632-1641	019-B2705	Y R L G P V Q N E I
	B4402	G	S	1407-1416	020-B4402	DE I AS K L R G M
	B4403	L	S	1639-1647	021-B4403	NE I C L TH P I
B5101	A	D	1388-1397	023-B5101	I ALL K GG R H L	
NS4B	A02	A	T	1873-1882	062-A02	K I M G G E L P T A
		A	T	1880-1889	065-A02	P T A E D M V N L L
	A0301	I	V	1901-1910	026-A0301	G V IC A IL R R
	A2601	R	K	1852-1861	027-A2601	R VLLD I L A G Y
	A68	A	I	1736-1744	067-A68	E K AL G LL Q R
	B27	R	K	1948-1957	061-B27	A R V T A LL S S L
	B4001	I	V	1847-1855	028-B4001	G I GL R V L L
	B51	A	T	1879-1888	066-B51	L P T A E D M V N L
		A	T	1881-1889	064-B51	T A E D M V N L L
C0401	V	I	1733-1742	025-C0401	Q F RE K V L G L L	
NS5A	A0201	G	S	2321-2330	029-A0201	AL P PR G AP P V
	A0301	V, P	A, S	2382-2391	030-A0301	K V P PS P GG E S
	A2601	D	G	2268-2276	033-A2601	ET D A E LS V A
	A68	T	S	1989-1998	070-A68	W V C T V L S D FK
	B0702	V	E	2309-2317	036-B0702	AP D Y V PP T V
		V	E	2313-2322	035-B0702	V PP T V H GC A L
		T	A	2332-2341	034-B0702	PP R R K R T I Q L
	B0801	I	V	2251-2259	037-B0801	E S E T K V V L L
B44	R	Q	2097-2105	071-B44	VE V R R V G D F	
NS5B	A0201	T	I	2489-2498	038-A0201	V L DD H Y K T A L
		N	D	2540-2549	040-A0201	SL S S K A I N Q I
		N	D	2544-2552	039-A0201	K A I N Q I RS V
	A1101	R	K	2500-2509	041-A1101	E V K E R A S R V K
	A2601	K	R	2537-2545	042-A2601	D V R S L S S K A
	B0801	I	M	2507-2515	043-A2601	R V K A R M L T I
	B1501	Q	L	2476-2484	044-B1501	S Q R Q K V T F
	B5101	K	R	2474-2482	045-B5101	S A S Q R Q K R V

Table S3: HLA-associated polymorphisms – sequence polymorphisms located *within* the predicted epitope sites.

HCV viral region, associated HLA type, epitope position, peptide identifier and sequence, as well as wild-type and variant sequences are depicted. Polymorphic sites associated with patient HLA are marked in in bold.

Protein	HLA	Wildtype	Variant Residue	Amino Acids	Polymorphic Site	Peptide ID	Predicted Epitope and Wild-type Residue ()
NS2	A0201	I	V	934-942	943	002-A0201	VMGGKYFQM (I)
		I	V	944-953	943	003-A0201	(I) ILSIGRWENT
	A02	L	P	861-870	871	054-A02	AALQVWVPPL (L)
		Y	H	878-886	887	053-A02	GVILLTSL (Y)
NS3	A68	L	F	1047-1056	1046	058-A68	(L) GTIVTSLTGR
	B5101	A	D	1379-1388	1389	022-B5101	IPFYGKAIP (A)
		A	D	1390-1398	1389	024-B5101	(A) QLKGGRHLI
NSSA	A1101	V	A	2374-2382	2383	031-A1101	DTQSSTTSK (V)
	A2402	D	E	2138-2147	2148	032-A2402	RYAPPCKPLL (D)
NSSB	B5101	M	L	2846-2854	2855	046-B5101	APTIVWRMV (M)

Table S4: HLA-associated polymorphisms – sequence polymorphisms located flanking the predicted epitope sites.

HCV viral region, associated HLA type, epitope position, peptide identifier and sequence as well as wild-type and variant sequences are depicted. Polymorphic sites flanking epitope are indicated in bold within brackets ().

Protein core	Position	3a peptide sequence	Patient	Patient viral sequence	CD8/CD4	
core	27-51	GGQIVGGVYVLPRRGPRL VYVLPRRGPRLGVRATRK	C58 *	GGQIVGGVYVLPRRGPRL	N/A	
			C106	VYVLPRRGPRLGVRATRK		
	66-90	PKARRSEGRSWAQPYPW GRSWAQPYPWPPLYGNEG	S11	SR	ND	CD4
			C5		ND	
			C12		PKARRSEGRSWAQPYPW	
			C37		PKARRSEGRSWAQPYPW	
			450		PKARRSEGRSWAQPYPW	
	130-147	FADLMGYIPLVGAPVGGV	C68 *	GRSWAQPYPWPPLYGNEG		
			C15	FADLMGYIPLVGAPVGGV		N/A
	137-154	IPLVGAPVGGVARALAH	C5	ND		
S15			SR		N/A	
143-158	PVGGVARALAHGVRAL	A16		PVGGVARALAHGVRAL	CD4	
		C6 *		PAGGVARALAHGVRAL		
		C12		PVGGVARALAHGVRAL		
		C13		PVGGVARALAHGVRAL		
		C18		PVGGVARALAHGVRAL		
		C19		PVGGVARALAHGVRAL		
		C22		PVGGVARALAHGVRAL		
		C23		PVGGVARALAHGVRAL		
		C27		PVGGVARALAHGVRAL		
		331 *		PVGGVARALAHGVRAL		
C70		PVGGVARALAHGVRAL				
C77 *		PVGGVARALAHGVRAL				
148-165	ARALAHGVRALDGINFA	C103	ARALAHGVRALDGINFA	CD8		
E2	460-476	CKPITFRQGWGSLTDA FRQGWGSLTDANITGSPD	A15	CKPITFRQGWGSLTDA	CD4	
			C106	FNQGWGSLTDANI SGPSD		
	610-625	LTPRCMVDYPYRLWHY	C106	LTPRCLVDPYRYRLWHY	N/A	
			S1	SR		
	635-650	KVRMFVGGFEHRFTAA	S12	SR		
			A11	KVRMFVGGFEHRFTAA	N/A	
696-719	LIHLHQNIVDVQYLYGV NIVDVQYLYGVGSGMVGW	C13	KVRMFVAGFEHRFTAA			
		S4 *	SR	CD8		
		A16	NIVDVQYLYGVGSGMVGW			
S9 *	SR					

Table S5: HCV genotype-3-specific T-cell targets in HCV structural regions.

HCV genotype-3-specific T-cell targets identified using overlapping peptide pools in HCV structural regions are depicted. All patients targeting specific individual peptides are specified (colour coding: orange-SR, blue-acute, yellow chronic). Circulating viral sequence is depicted when obtained, as well as CD4+ /CD8+ subset analysis. Sequence polymorphisms differing from overlapping peptide set sequence are marked in red. * Epitopes previously described in (Humphreys et al. 2012). N/A not available.

Protein	Position	3a peptide sequence	Patient	Patient viral sequence	CD8/CD4
NS2	886-896	(L)LYPSLIFDI	# A8	LLYPSLIFDI	CD8
			# A8	LYPSLIFDI	
			# C1	LYPSLIFDI	
			# C19	LYPSLIFDI	
			# C47	LYPSLIFDI	
# C58	LYPSLIFDI				
NS2	931-940	LVRVSMGGKY	# S15	SR	CD8
	941-951	FQMILSIIGR	# A16	FQMILSIIGR	CD8
			# C38	FQMILSIIGR	
# C73	FQMVILSIIGK				
NS3	1040-1062	AQQTRGLLGTIVTSLTGR LGTIVTSLTGRDKNVV	# S15	SR	N/A
	1139-1147	LVTRDADVI	# S15	SR	CD8
	1198-1213	KALQFIPVETLSTQAR	S5 *	SR	N/A
	1246-1261	KVPAAYVAQGYNVLVL	S8 *	SR	N/A
	1264-1281	SVAATLGFGSFMGRAYGI	A11	SVAATLGFGSFMGRAYGI	N/A
			S6	SR	
	1282-1305	DPNIRTGNRTVTGAKL GNRTVTGAKLTYSTYK	S1 *	SR	N/A
			S12	SR	
	1370-1387	BEVALGSEGEIPFYGKAI	A13	ND	CD8
	1379-1387	IPFYGKAIFI	A13	ND	
	A2	ND			
	1423-1440	AYYRGLDVSVIPTAGDVV	C22	AYYRGLDVSVIPTAGDVV	N/A
			S11 *	SR	CD4
	1436-1447	GDVVVCATDALMTGF	A15	GDVVVCATDALMTGY	CD8
	1442-1447	ATDALMTGY	A15	ATDALMTGY	
			C68	ATDALMTGF	
			C106	ATDALMTGF	
# C108	ATDALMTGF				
NS3	1520-1537	RPSGMFDSVVLCECYDA DSVVLCECYDAGCSWYDL	C5	ND	CD8
			C7	RPSGMFDSVVLCECYDAGCSWYDL	
			C15 *	RPSGMFDSVVLCECYDAGCSWYDL	
			C17	RPSGMFDSVVLCECYDAGCSWYDL	
			C19	RPSGMFDSVVLCECYDAGCSWYDL	
			C27 *	RPSGMFDSVVLCECYDAGCAWYDL	
			C35	ND	
			C42	ND	
			C44	RPSGMFDSVVLCECYDAGCSWYDL	
			C58 *	RPSGMFDSVVLCECYDAGCSWYDL	
			C96	ND	
C98	ND				
S13	SR				
S14	SR				
1547-1569	RAYLSTPGLPVCQDHLDF GLPVCQDHLDFWESVF	C19	RAYLSTPGLPVCQDHLDFWESVF	N/A	
NS4B	1792-1808	PAVASLMFPTASVTSPL	A8	ND	CD8
			C68 *	PAVASLMFPTASVTSPL	
			C77 *	ND	
	1805-1822	TSPLTTNQTMMFFNILGGW	S8 *	SR	N/A
			S13	SR	
	1825-1842	THLAGPQSSSAFVVSGLA	C77 *	ND	N/A
1853-1862	RVLLDILAGY	A16	KVLLDILAGY	CD8	
		A4	ND		
		C5	KVLLDILAGY		
		C52	ND		
# C77	ND				
1917-1932	EGAVQWMNRLIAFASR	C67 *	EGAVQWMNRLIAFASR	CD8	
NS5A	2030-2047	GVMSTRCPGASIAGHVK	C13 *	GVMSTRCPGASITGHVK	CD8
	2119-2136	CPCQVPAEFTTEVDGVR	A8	CPCQVPAEFTTEVDGVR	
	2126-2141	AEFFTEVDGVRLHRYA	A13	ND	CD8
			A15	ND	
S10	SR				
S15	SR				
2145-2162	KFLLRDEITFMVGLNSYA	A5	ND	N/A	
NS5B	2484-2499	TFDRLQVLDHYKTAL	A13	TFDRLQVLDHYKTAL	CD8
	2490-2499	VLDHYKTAL	# C68	ND	
	2508-2516	RVKARMLTI	A13	RVKARMLTI	CD8
	# S10	SR			
	2548-2565	NQIRSVWEDLLEDTTPI	S2 *	SR	CD4
	2603-2618	KRALYDVIQKLSIETM	S11 *	SR	CD4
	2844-2861	IMYAPTIVWRMVMTHFF	A4	ND	N/A
	2893-2908	IIERLHGLSAFTLHSY	S2 *	SR	CD4
	2947-2964	GKAKICGLYLFNVAVRTK	C37 *	GKAKITGLYLFNVAVRTK	CD8
	2967-2976	KLTPLPAGQL	450 *	KLTPLPAGQLL	CD8

Table S6: HCV genotype-3-specific T-cell targets in HCV non-structural regions.

HCV genotype-3-specific T-cell targets identified using overlapping peptide pools and HLA predicted peptides (#) in HCV structural regions are depicted. All patients targeting specific individual peptides are specified (colour coding: orange-SR, blue-acute, yellow chronic). Circulating viral sequence is depicted when obtained, as well as CD4+ /CD8+ subset analysis. Sequence polymorphisms differing from overlapping peptide set sequence are marked in red. *Epitopes previously described in (Humphreys et al. 2012). N/A not available.

GT3 CD8+ epitopes (Oxford Study Cohort)						GT1 CD8+ epitopes in corresponding regions		
Position	3a peptide sequence	Viral protein	HLA	Pt ID	HLA type	Peptide (Literature)	HLA	First author
931-940	LVRVVMGGKY	NS3	A03	S15	A*3201 A*0301 B*1401 B*0702 C*0802 C*0702 #	no CD8 epitopes described		
941-951	FQMILLSIGR	NS2	B27	A16 C38 C73	A*0201 A*2601 B*2702 B*3801 C*1203 # A*0201 A*1101 B*1801 B*2705 C*0102 C*1203 # A*0201 B*5701 B*2702 C*0701 C*1501 #	no CD8 epitopes described		
1917-1932	EGAVQVMNRLIAFASR	NS4B		C67	A*0101 A*3001 B*1302 B*4402 C*0602 C*0501 # *	no CD8 epitopes described		
2030-2047	GVMSTRCPGASIAGHVK	NS5A		C13	A*1101 A*7401 B*4403 B*38 C*04 C*0702 # *	no CD8 epitopes described		
2484-2499	TFDRLQVLDHDKYKAL	NS5B		A13	A*0101 B*0801 B*5101 C*0102 C*0701 #	no CD8 epitopes described		
2490-2499	VLDHDKYKAL	A02		C68	A*0101 A*0201 B*0801 B*5701 C*0602 C*0701 #			
2508-2516	RVKARMLTI	NS5B	B08	A13 S10	A*0101 B*0801 B*5101 C*0102 C*0701 # A*3201 A*0101 B*0801 B*4402 C*0501 C*0701	no CD8 epitopes described		
2967-2976	KLTLPLPAAGQL	NS5B		450	# *	no CD8 epitopes described		
1853-1862	RVLLDLILAGY	NS4B	A26	A16 A4 C6 C52 C77	A*0201 A*2601 B*2702 B*3801 C*120301 # A*0201 A*2601 B*3801 B*4402 C*0501 C*1203 # A*2301 A*2601 B*3801 B*4901 C*0701 C*1203 # A*0101 A*2601 B*4501 B*0801 C*0602 C*0701 # A*0201 A*2601 B*3801 B*2702 C*1203 C*0102 #	<u>ILAGYAGV</u> <u>ILAGYAGV</u> <u>ILAGYAGV</u>	A2 A2 A2	M Battegay N H Gruener T Kuntzen
886-896	LLYSLIFDI/LVPSLIFDI	NS2	A02/A24	A8 C1 C19 C47 C1 C58	A*2402 A*0201 B*3502 B*4403 C*0401 C*1601 # A*1101 A*2404 B*18 B*3501 C*0401 C*1203 # A*24 B*3501 B*4403 C*0401 C*0409 # A*0205 A*2402 B*1302 B*4901 C*0602 C*0701 # A*0205 A*2402 B*1302 B*4901 C*0602 C*0701 # A*0101 A*0201 B*0702 B*4001 C*0304 C*0702 #	<u>HTLVFDI</u> <u>HTLVFDI</u>	Class I Class I	A L Cox T Kuntzen
2947-2964	GKAKICGLYLFNVAVRTK	NS5B		C37	A*1101 B*0702 B*4402 C*0501 C*0702 # *	<u>GRRAICGRK</u> <u>GRRAICGRK</u> <u>GRRAICGRK</u> <u>KVLFNVAVK</u>	Class I B27 Class I A2	C Neumann-Haefelin C Neumann-Haefelin P T F Kennedy Z Guo
148-165	ARALAHGVRALEDGINFA	core		C103	#	<u>GVRLVDGV</u> <u>VLEDGVN</u> <u>VLEDGVN</u>	A2 Class I Class I	H F Lohr D D Anthony K Sugimoto
1139-1147	LVTRDADVI	NS3	A03	975	A*3201 A*0301 B*1401 B*0702 C*0802 C*0702 #	<u>VTRDADIV</u>	Class I	T Kuntzen
1520-1537	RFSGMDSVVLCRCYDAGCSWYDL	NS3	B35/C04	C5 C7 C15 C17 C19 C27 C35 C42 C44 C58 C96 C98 S13 S14	A*1101 A*1101 B*3501 B*5101 C*0401 C*1402 A*0101 A*0201 B*35 B*14 C*0401 C*0802 A*2402 A*3002 B*0702 B*3501 # * A*1101 A*1101 B*1517 B*3501 C*0401 C*0701 A*1101 A*2402 B*18 B*3501 C*0401 C*1203 A*0301 A*3002 B*0702 B*3501 C*0401 C*0702 # * A*0101 A*2901 B*3501 B*4403 C*0401 C*1601 A*0301 A*2402 B*3501 B*5501 C*0301 C*0401 A*2402 B*3501 B*4403 C*0401 C*0409 # * A*0201 A*1101 B*3501 B*4001 C*0401 C*0301 A*0301 A*2601 B*3501 B*2701 C*0401 C*0202 A*0201 A*1101 B*4402 B*3501 C*0401 C*0501	<u>MFDSVLCRCYDAGC</u>	Class I	D Ciuffreda
2126-2141	AEFFTEVDGVRLLHRYA	NS5A		A13 A15 S10 S15	A*0101 B*0801 B*5101 C*0102 C*0701 # A*0101 B*5201 B*5701 C*0602 C*1202 A*3201 A*0101 B*0801 B*4402 C*0501 C*0701 A*3201 A*0301 B*1401 B*0702 C*0802 C*0702	<u>FFTELDGVRLLHREFAP</u>	Class I	D Ciuffreda
1792-1808	PAVASLMAFTASVTSPL	NS4B		6-23 C68 C77	A*2402 A*0201 B*3502 B*4403 C*0401 C*1601 A*0101 A*0201 B*0801 B*5701 C*0602 C*0701 # * A*0201 A*2601 B*3801 B*2702 C*1203 C*0102 # *	<u>SIMAFTAAV</u> <u>SIMAFTAAV</u> <u>SIMAFTAAV</u>	A2 A2 A2	B Reherrmann K M Chang N H Gruener
696-719	LIHLHQNIVDQVYLYGVSGMVGW	E2		S4 A16 S9	A*02 A*3201 B*2707 B*1501 C*0602 C*0304 # * A*0201 A*2601 B*2702 B*3801 C*1203 # *	<u>ALSTGLHLHQNIVD</u> <u>LHQNIVDQVYLYGVG</u>	Class I Class I	D Ciuffreda D Ciuffreda
1379-1387	IPFYKAIPI	NS3	B51	A13 A2	A*0101 B*0801 B*5101 C*0102 C*0701 # B*4403 B*5101 C*1402 C*1601 #	<u>IPFYKAI</u> <u>IPFYKAIPL</u>	& B51 B51	S Giugliano D Yerly
1436-1447	GDVVCATDALMTGF	NS3		A15 A15	A*0101 B*5201 B*5701 C*0602 C*1202 A*0101 B*5201 B*5701 C*0602 C*1202 #	<u>ATDALMTGY</u> <u>VATDALMTGY</u>	A1 Class I	G M Lauer, 2002 A M Werthheimer
1442-1447	ATDALMTGY	A01		C108 C106 C68	A*0101 A*2402 B*0801 B*3906 C*0701 C*0702 # A*0101 A*0301 B*0702 B*0702 C*0702 C*0702 # A*0101 A*0201 B*0801 B*5701 C*0602 C*0701 #	<u>ATDALMTGF</u> <u>ATDALMTGF</u> <u>ATDALMTGY</u> <u>ATDALMTGY</u> <u>ATDALMTGY</u>	& A1 A1 A1 A1	S Giugliano T Kuntzen A L Cox E Barnes G M Lauer, 2004

Table S7: Overlap between genotype-3-specific CD8+ T-cell targets defined in this study and previously described HCV genotype-1-specific CD8+ targets.

HCV genotype-3-specific CD8+ epitopes and described genotype-1-specific CD8+ epitopes were classified by overlap (left coloured bar): ‘overlapping’ (blue), ‘likely overlapping’ (light blue, <20% sequence differences within targeted area), ‘unlikely overlapping’ (light red, >20% differences within targeted area, or less than 7 amino acids overlap), and ‘not overlapping’ (red). **Left:** For each epitope, viral region, position and genotype-3 sequence is given, as well as patients targeting the epitopes (colour coding: orange-SR, blue-acute, yellow chronic), and patient’s HLA class-I types. **Right:** Published overlapping genotype-1 CD8+ epitopes are specified, including sequence, restricting HLA type and first author of the publication; overlap between epitopes is underlined and genotype-3/genotype-1 sequence differences marked bold. # CD8 restriction for marked patient experimentally defined; * Epitopes

previously described in (Humphreys et al. 2012); & Epitope previously described for genotype-1 and 3.

(M Battegay et al. 1995; Rehermann et al. 1996; K. M. Chang et al. 1997; Löhr et al. 1999; Grüner et al. 2000; Wertheimer et al. 2003; Sugimoto et al. 2003; Lauer et al. 2004; Cox et al. 2005; Kennedy et al. 2006; Kuntzen et al. 2007; Yerly et al. 2008; Ciuffreda et al. 2008; Giugliano et al. 2009; Neumann-Haefelin et al. 2008; Barnes et al. 2012; Guo et al. 2012)

GT3 CD4+ epitopes (Oxford Cohort)						GT1 epitopes described in corresponding regions			
Position	3a peptide sequence	Viral protein	Patient ID			Peptide (Literature)	HLA	First author	
453-476	CPQRLSSCKPITFFRQGWGSLTDANITGFS	E2	A15 C106	DRB1*0701	DRB1*1502	#	no CD4 epitopes described		
2603-2618	KRALYDVIQRKLSIETM	NS5B	S11	DRB1*0404	DRB1*0701	# *	KLPLAVMGSYGFQYSPGQR KLPLAVMGSYGFQYSPGQR	Class-II Class-II J Schulze zur Wiesch, 2007 C L Day	
66-90	PKARRSEGRSNAQPGYFWPLYGNEG	core	C5 C12 C37 450 C68	DRB1*0101 DRB1*0301 DRB1*0401 DRB1*0101	DRB1*0701 DRB1*0401 DRB1*0407 DRB1*0701	# # # *	RRQPTPKARRPEGR T WAQPG RQPIPKVRRPEGR T KVRREPEGR T WAQPG PEGR T WAQPGYFWPLYGNEG PEGR T WAQPGYFWPLYGNEGCGW PEGR T WAQPGYFWP PEGR T WAQPGYFWPL	Class-II HLA-DR HLA-DR Class-II Class-II HLA-DR Class-II	J Schulze zur Wiesch, 2012 J J Lasarte J J Lasarte J Schulze zur Wiesch, 2007 H F Löhr J J Lasarte P T F Kennedy
143-158	PVGGVARALAHGVRL	core	A16 C6 C12 C13 C18 C19 C22 C23 C27 331 C70 C77	DRB1*0801 DRB1*0301 DRB1*1504 DRB1*0301 DRB1*0403 DRB1*0404 DRB1*0701 DRB1*0101 DRB1*0101 DRB1*0101 DRB1*0101	DRB1*1101 DRB1*0401 DRB1*0701 DRB1*1602 DRB1*0701 DRB1*1104 DRB1*0801 DRB1*1501 DRB1*1501 DRB1*1501	# # # # # # # # # *	ADLMGYIPLVGA <u>PLGCAARA</u> ADLMGYIPLVGA <u>PLGCAAR</u> ADLMGYIPLVGA <u>PLGCAARA</u> LMSGYIPLVGA <u>PLGCAARA</u> LVGAP <u>PLGCAARA</u> LVGAP <u>PLGCAARALAH</u> GAP <u>PLGCAARALAHGVRL</u> ED GAP <u>PLGCAARALAHGVRL</u> ED GAP <u>PLGCAARALAHGVRL</u> ED GAP <u>PLGCAARALAHGVRL</u> ED GG <u>ARALAHGVRL</u> ALAHGVRL LAHGVRVLEDGNNYATGNLP	Class-II HLA-DR Class-II Class-II Class-II Class-II Class-II Class-II Class-II Class-II Class-II Class-II	J Schulze zur Wiesch, 2007 F A Castelli J Schulze zur Wiesch, 2012 D Ciuffreda H F Löhr K Sugimoto J Schulze zur Wiesch, 2007 A J MacDonald C L Day J Schulze zur Wiesch, 2012 D Ciuffreda H F Löhr J Schulze zur Wiesch, 2012
1423-1440	AYYRGLDVSVIPTAGDVV	NS3	C22 S11	DRB1*0404 DRB1*0404	DRB1*1104 DRB1*0701	# *	GINAVAYYRGLDVSVIPT <u>SG</u> GINAVAYYRGLDVS GINAVAYYRGLDVSVIPT <u>SG</u> VAYYRGLDVSVIPT <u>S</u> LDVSVIPTSGDVVVVATDAL IPTSGDVVVVSTDALMTG	Class-II Class-II Class-II Class-II Class-II Class-II	J Schulze zur Wiesch, 2007 A M Wertheimer C L Day A M Wertheimer J Schulze zur Wiesch, 2007 N M Tabatabai
2548-2565	NQIRSVWEDLLEDTTPI	NS5B	S2	DRB1*0701	DRB1*1454	# *	HARKAVT <u>HS</u> INSVKDLLEDN SVWKDLLEDNVPFLDTTMA	Class-II Class-II	J Schulze zur Wiesch, 2012 C L Day
2893-2908	IIERLHGLSAFTLHSY	NS5B	S2	DRB1*0701	DRB1*1454	# *	PII <u>QRLHGLSAF</u> SLHSYSPG PII <u>QRLHGLSAF</u> SLHSYSPG	Class-II Class-II	J Schulze zur Wiesch, 2007 J Schulze zur Wiesch, 2012

Table S8: Overlap between genotype-3-specific CD4+ T-cell targets defined in this study and previously described HCV genotype-1-specific CD4+ targets.

HCV genotype-3-specific CD4+ T-cell targets and described genotype-1-specific CD4+ targets and those without defined CD4/CD8 restricted were classified by overlap (left coloured bar): ‘likely overlapping’ (light blue, <20% sequence differences within targeted area), ‘unlikely overlapping’ (light pink, >20% differences within targeted area, or less than 7 amino acids overlap), and ‘not overlapping’ (pink).

Left: For each epitope, viral region, position and genotype-3 sequence is given, as well as patients targeting the epitopes (colour coding: orange-SR, blue-acute, yellow chronic), and patient’s HLA class-II types. **Right:** Published overlapping genotype-1 CD8+ epitopes are specified, including sequence, restricting HLA type and first author of the publication; overlap between epitopes is underlined and genotype-3/genotype-1 sequence differences marked bold. # CD4 restriction for marked patient experimentally defined; * Targets previously described in (Humphreys et al. 2012).

(Lasarte et al. 1998; Lamonaca et al. 1999; Löhr et al. 1999; Tabatabai et al. 1999; Day et al. 2002; MacDonald et al. 2002; Sugimoto et al. 2003; Wertheimer et al. 2003; Kennedy et al. 2006; Castelli et al. 2007; Schulze zur Wiesch et al. 2007; Ciuffreda et al. 2008; Schulze Zur Wiesch et al. 2012).

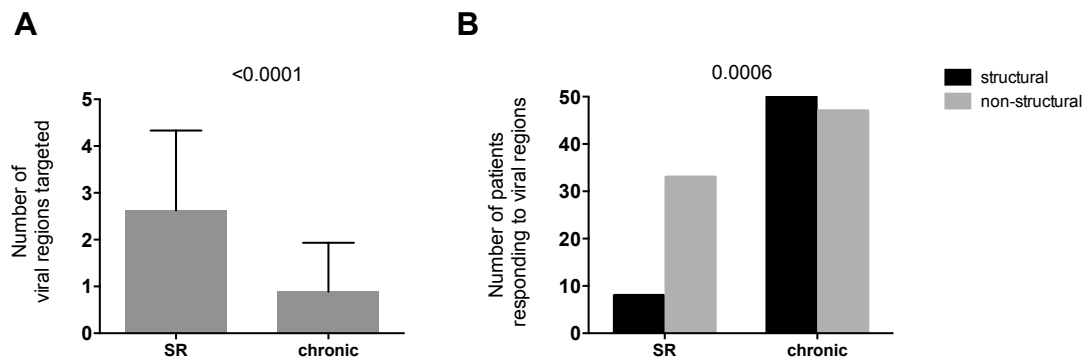


Figure S1: Comparison of breadth and targeted viral regions in spontaneously resolved individuals and patients with chronic HCV genotype-3 infection.

(A) Numbers of viral regions targeted in spontaneously resolved HCV infection and chronic HCV genotype-3 infection (unpaired t-test). (B) Comparison of numbers of patients targeting structural and non-structural regions in patient with chronic HCV genotype-3 infection and patients with spontaneously resolved HCV infection (Fisher's exact).

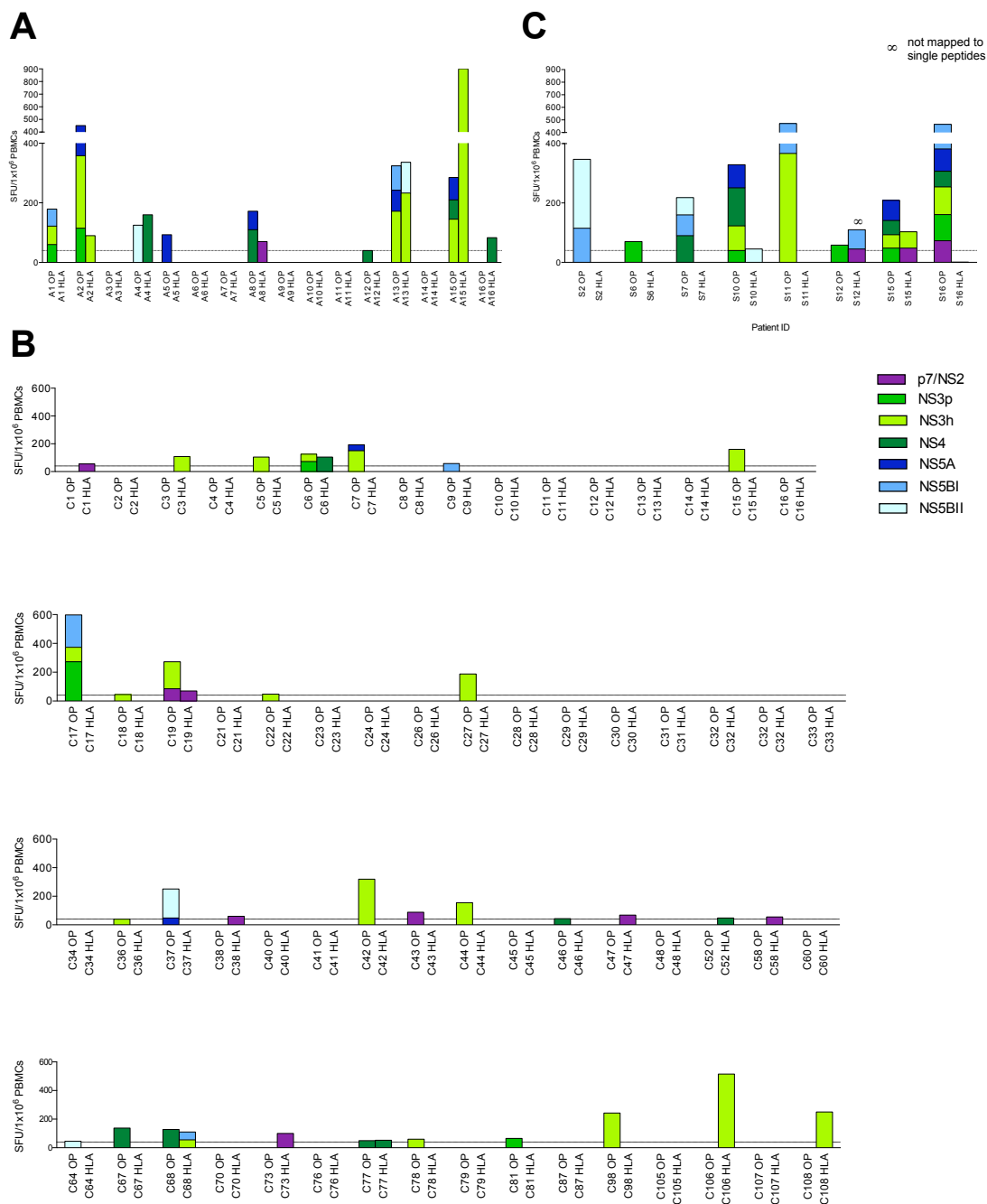
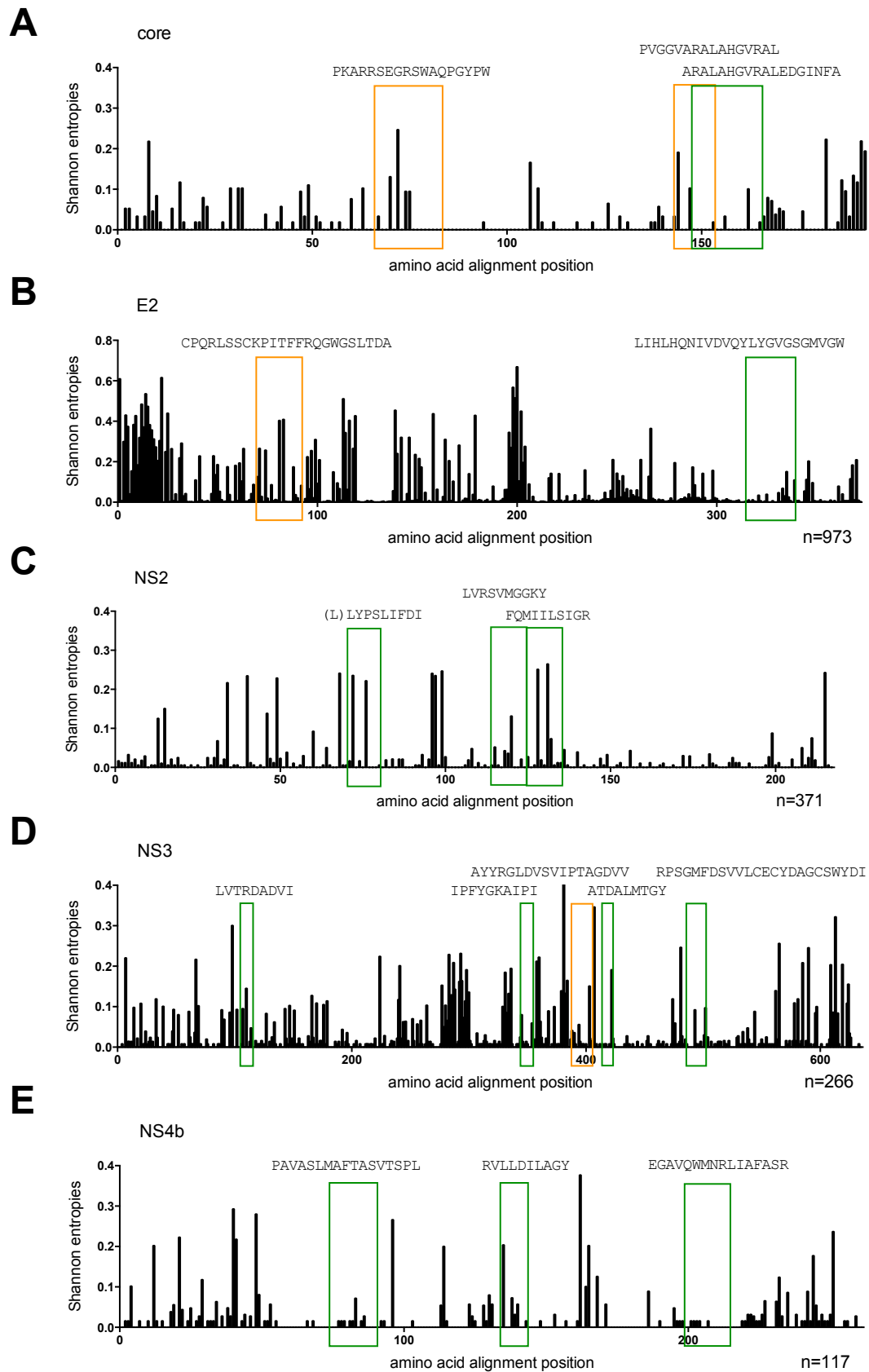


Figure S2: Comparison of responses to overlapping pools and HLA predicted peptides in acutely and chronically HCV genotype-3 infected patients, and patients with spontaneously resolved infection.

T-cell responses detected by IFN γ ELISpot assay to HLA predicted peptides and overlapping peptide pools (OP) were measured in patients with (A) acutely (n=16) and (B) chronically HCV genotype-3 infected (n=64) patients, and (C) spontaneously resolved infection (n=8). Responses to overlapping pools are compared to the HCV viral region in which the HLA predicted peptide falls (NS3p, NS3h, NS4, NS5a, NS5b1, NS5b2). SFU: Spot forming units.



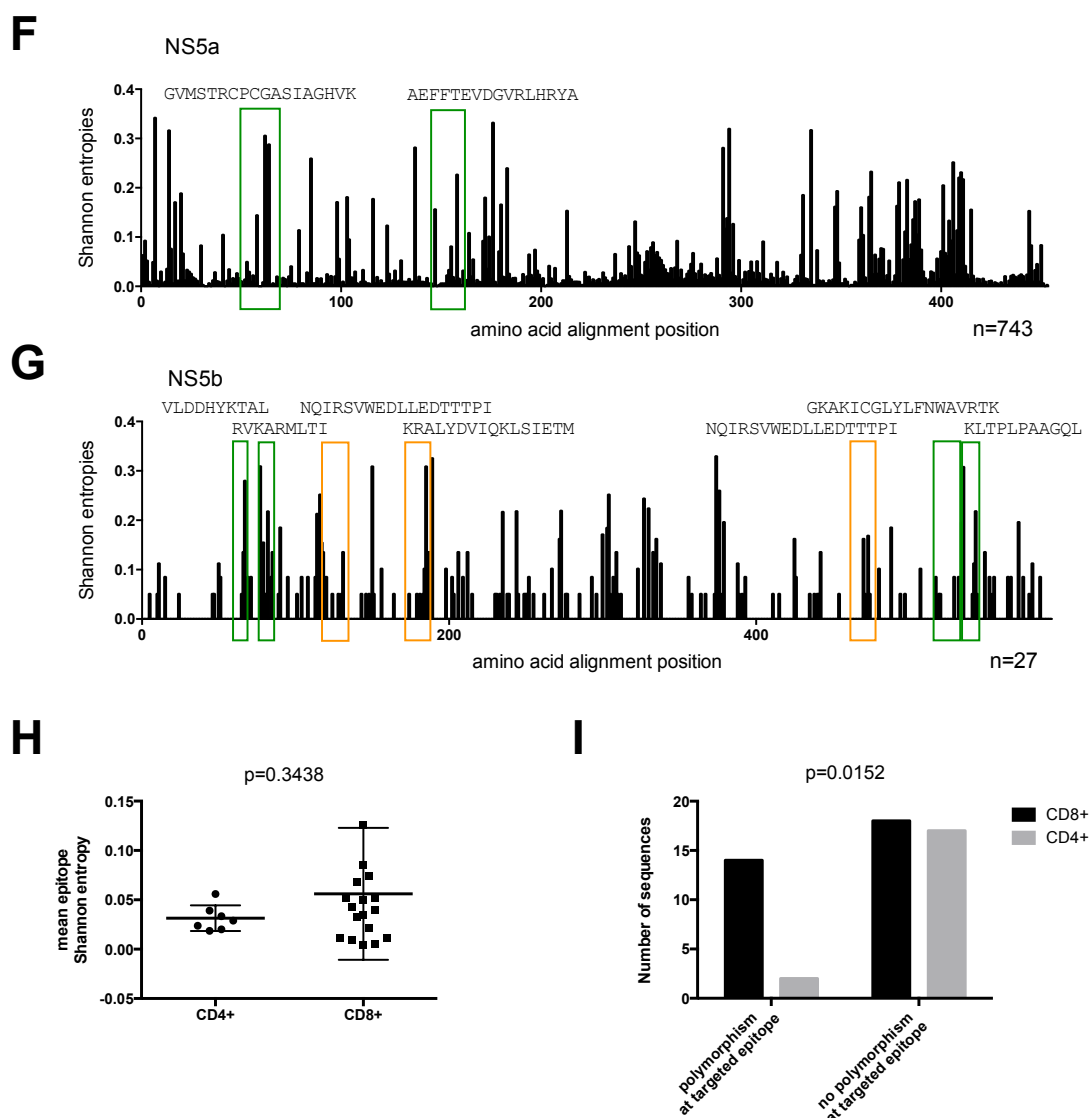


Figure S3: Evaluation of sequence heterogeneity at targeted HCV genotype-3-specific epitopes.

Sequence heterogeneity was evaluated by Shannon entropy scores in HCV viral regions (A) core, (B) E2, (C) NS2, (D) NS3, (E) NS4b, (F) NS5a and (G) NS5b on population level using Los Alamos genotype-3 sequences. The number of sequences used for each calculation is given. CD4+ (orange) and CD8+ (green) T-cell targets detected within the Oxford genotype-3 cohort are marked. (H) Mean Shannon entropy scores for CD4+ and CD8+ epitopes were calculated ($p=0.3438$, unpaired t-test). (I) Sequence polymorphisms within the Oxford cohort were evaluated at 16 CD8+ and 4 CD4+ T-cell targets in patients with detected T-cell responses. Sequence polymorphisms at CD4+ epitopes were detected in 2/19 sequenced patients, whereas polymorphisms at CD8+ epitopes were detected in 14/32 sequenced patients, respectively ($p=0.0152$, Fisher's exact).

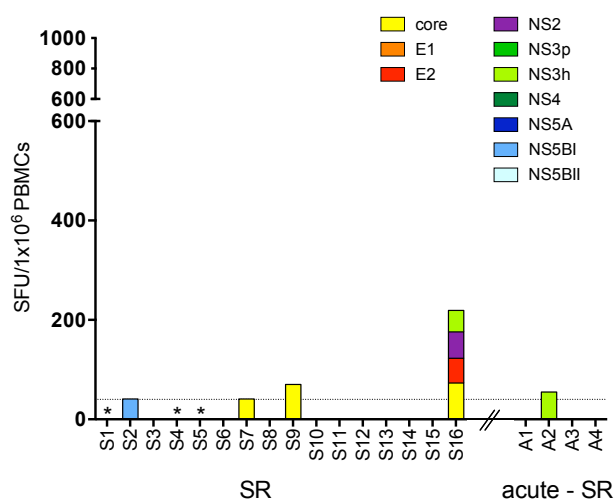


Figure S4: T-cell responses detected against HCV genotype-1 peptide sets in spontaneously resolved patients and acutely infected patients with subsequent resolution of infection.

Hepatitis C virus (HCV) genotype-1 specific T-cell responses were measured by IFN γ -ELISpot assays (spot-forming units (SFU)/10⁶ peripheral blood mononuclear cells (PBMC)) using an HCV genotype-1b-specific peptide set spanning the entire HCV genome in 16 patients with spontaneously resolved infection (SR), and 4 patients acutely infected with HCV genotype-3 who subsequently spontaneously resolved infection (A-SR).

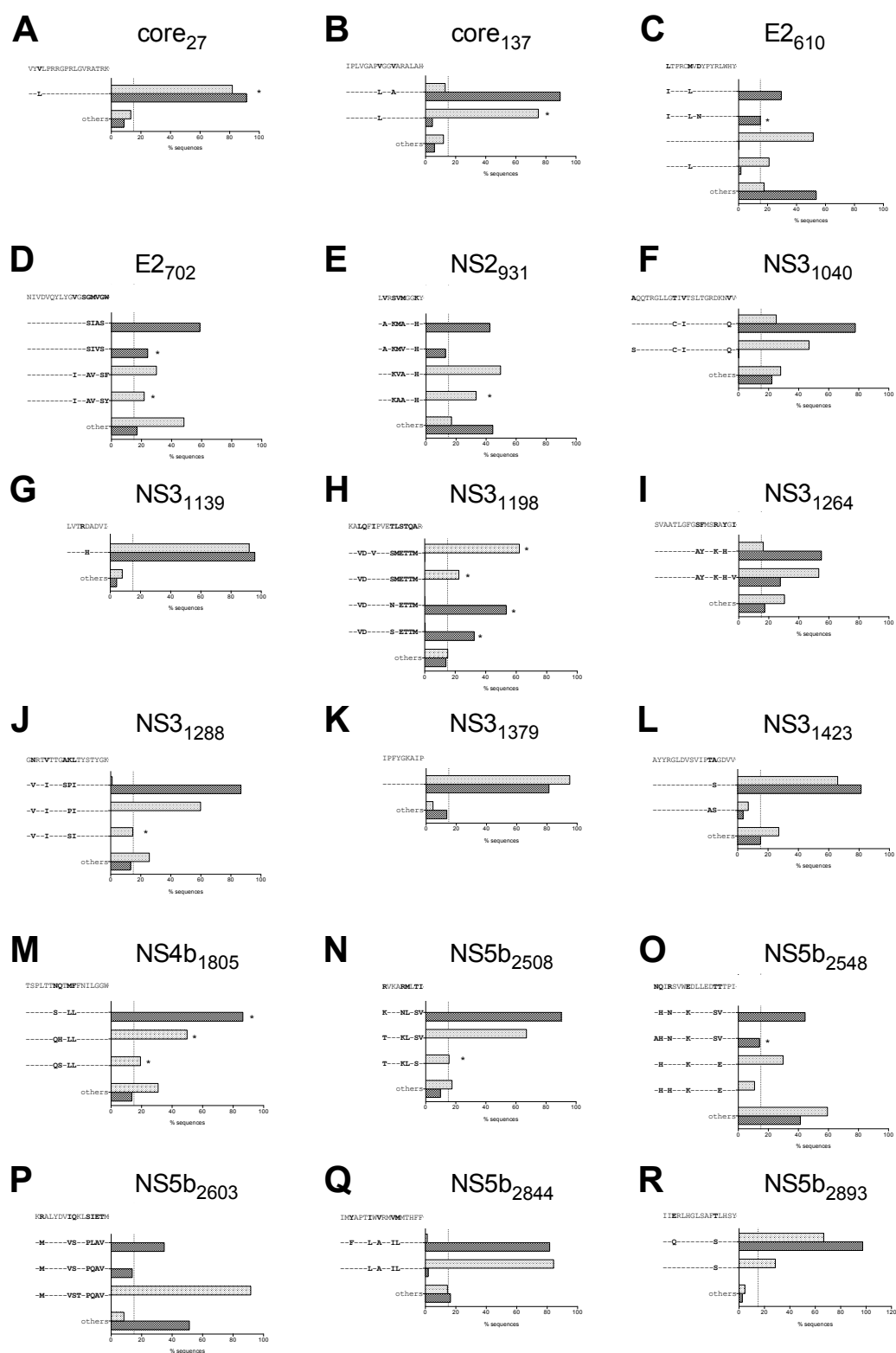


Figure S5: Hepatitis C virus (HCV) genotype-1 sequences variants at T-cell targets identified in spontaneously resolved patients.

HCV genotype-1 sequences were obtained from the Los Alamos database at T-cell targets identified in spontaneously resolved patients, to define common HCV genotype-1 sequence variants (frequencies >15%). For each T-cell target (A-R), HCV genotype-3 consensus sequence (top of each graph) and common HCV genotype-1 variants are specified. Where T-cell cross-reactivity to common variants has not been assessed experimentally due to lack of PBMC, sequences are marked with a star.

A

	Epitope sequence	Viral region	start AA	end AA	HLA type	number of publications	T cell targets in Oxford gt3 cohort	Restriction of HCV gt3 epitopes	
CD8	YLLPRRGPRLL	core	35	44	A2	10			
	GPRLGFRAT	core	41	49	B7	5			
	NEGLGWGTGW	core	87	95	B44	5			
	ADLMGYIPLV	core	131	140	A2	11			
	LLALLSCLTV	core	178	187	A2	5			
	SMVGNWAKV	E1	363	371	A2	5			
	SLLAPGAKQNV	E2	401	411	A2	5			
	NTRPPLGNW	E2	541	549	B57	5			
	RDWAHNL	NS2	957	964	B37	7			
	CINGVCWTV	NS3	1073	1081	A2	31			
	LLCPAGHAV	NS3	1169	1177	A2	5			
	HPNIEEVAL	NS3	1359	1367	B35	7			
	HSKKKDEL	NS3	1395	1403	B8	8			
	KLVALGINAV	NS3	1406	1415	A2	32			
	ATDALMTGY	NS3	1436	1444	A1	7	+	CD8+	
	LLFNILGGWV	NS4b	1807	1816	A2	7			
	VLSDFKFWL	NS4a	1987	1995	A2	7			
	ALYDVVTKL	NS5b	2594	2602	A2	11			
	CD4+	NKRNTNRRPQDKVFFGGGQIVGGVYLLPRRGPRLL	core	11	44	ND	9		
		KVRRPEGRRTWAQPGYPWPPLYGNEGLGWAGWLLSPRGS	core	67	103	ND	10	+	CD4+
ADLMGYIPLVGAPLGGAAARALAHGVRVLED		core	131	160	ND	12	+	CD4+	
ATRDGKLPATQLRRHIDLL		E1	247	265	ND	7			
YFSMVGNWAKVLVVL		E1	361	375	ND	6			
SSDLYLVTRHADVIP		NS3	1127	1141	ND	6			
LETTMRSPVFTDNSSPFVVP		NS3	1201	1220	ND	6			
PAAYAQQGYKVLVNLNSVAA		NS3	1241	1260	ND	8			
LADAGCSGGAYDIIICDECHSTDAT		NS3	1300	1324	ND	8			
EVIKGRHLIFCHSKKCCD		NS3	1383	1401	ND	9			
CNTCVTQTVDFSLDPTFT		NS3	1454	1471	ND	7			
FWAPCRRPSSMFD		NS3	1501	1513	ND	6			
YELTPAETTVRLRAYMNTFGLPVAQD		NS3	1528	1553	ND	11			
THIDAHFLSQT		NS3	1566	1577	ND	6			
ENLPYLVAQATVCAQAQPPPSW		NS3	1581	1604	ND	10			
PLLYRLGAVQNEITLTHP		NS3	1623	1640	ND	8			
VIVGRVLSGKPAIIPDREV		NS4a	1681	1700	ND	6			
GLSTLPGNPAIASL		NS4a	1777	1790	ND	6			
ALVVGVCALILRRHVGPE		NS4a	1891	1910	ND	6			
QCCLDPOARVAIKSLTERL		NS5b	2661	2680	ND	6			

B

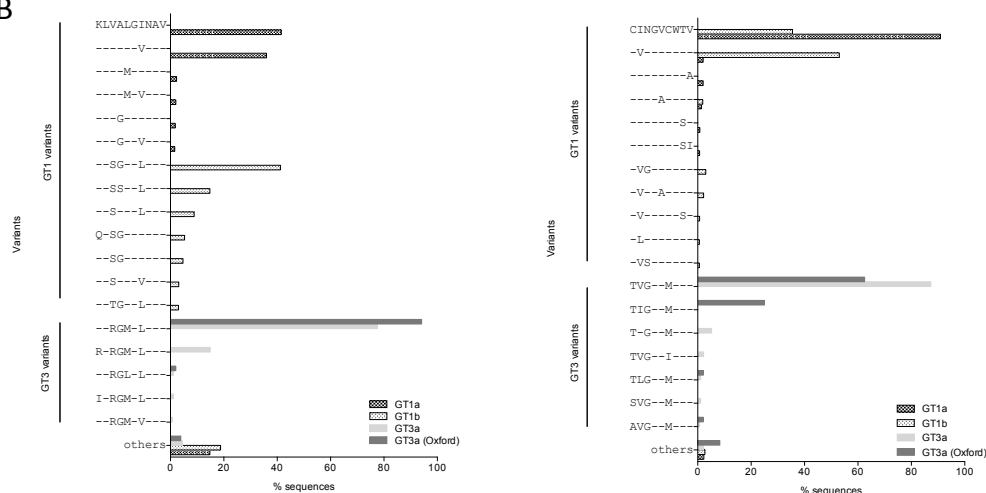


Figure S6: T-cell targets frequently detected in HCV genotype-1 infection are not targeted in HCV genotype-3 infection, likely due to sequence differences between genotypes.

(A) Comparison of CD8+ restricted epitopes dominant in HCV genotype-1 infection (defined as described in 5 or more publications on the IEDB). If the epitope was targeted in the Oxford HCV genotype-3 cohort, it is marked with +. Epitopes described as CD4+ restricted epitopes in the Oxford genotype-3 cohort falling into regions of CD8+ restricted epitopes described as HCV genotype-1 epitopes in the literature are marked in grey. (B) Comparison of circulating viral genotype-1 and genotype-3 sequences at dominant HCV genotype-1 epitopes. Sequences were obtained from the Los Alamos database, with additional in-house genotype-3 sequences.

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