

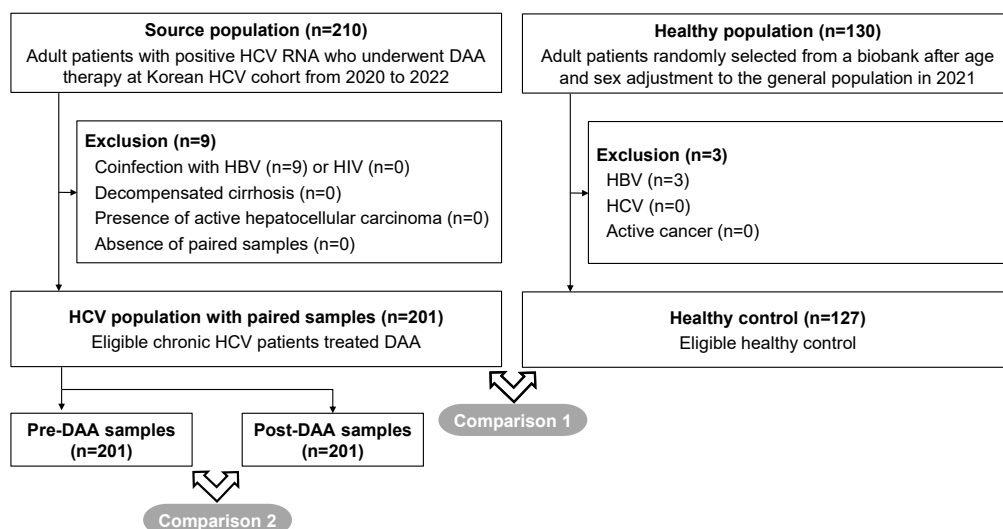


Figure S1 Patient flow gram. DAA, direct-acting antiviral; HBV, hepatitis B virus; HCV, hepatitis C virus.

Table S1 Comparison between ANA-positive and negative groups in healthy controls

Variables	ANA-positive (n=27)	ANA-negative (n=100)	P value
Age (years)	53 [39–70]	56 [46–63]	0.97
Age group (years)			0.77
20–29	4 (14.8)	11 (11.0)	
30–39	3 (11.1)	12 (12.0)	
40–49	4 (14.8)	13 (13.0)	
50–59	5 (18.5)	25 (25.0)	
60–69	4 (14.8)	23 (23.0)	
70+	7 (25.9)	16 (16.0)	
Female	18 (66.7)	45 (45.0)	0.046
Hypertension	7 (25.9)	21 (21.0)	0.58
Thyroid disease	5 (18.5)	3 (3.0)	0.51
Lab findings			
Hemoglobin (g/dL)	13.86±1.46	14.23±1.33	0.21
WBC ($\times 10^3/\mu\text{L}$)	5,229.26±1,847.49	5,499.6±1,386.49	0.41
Platelet ($\times 10^3/\mu\text{L}$)	248.11±44	261.2±50.23	0.22
AST (IU/L)	30.81±14.36	30.77±16.1	0.99
ALT (IU/L)	31.74±24.73	29.58±22.04	0.66
Globulin (g/dL)	2.79±2.90	2.72±0.33	0.31
Protein (g/dL)	7.28±0.29	7.22±0.36	0.45
Albumin (g/dL)	4.49±0.21	4.5±0.24	0.76
Bilirubin (mg/dL)	0.79±0.26	0.88±0.34	0.23
Creatinine (mg/dL)	0.71±0.18	0.74±0.16	0.34
Cholesterol (mg/dL)	187.85±36.86	188.59±35.3	0.92
Fasting blood glucose (mg/dL)	89.81±12.15	93.96±13.79	0.16

Continuous variables are expressed as mean $\pm$ SD or median [IQR], and the categorical variable are indicated as n (%). ALT, alanine aminotransferase; ANA, antinuclear antibody; AST, aspartate aminotransferase; IQR, interquartile range; SD, standard deviation; WBC, white blood cell count.

Table S2 Comparison between ANA-positive and negative groups in HCV patients at PreTx

Variables	ANA-positive (n=65)	ANA-negative (n=136)	P value
Age (years)	65 [55–70]	60 [50–70]	0.19
Females	29 (44.6)	71 (52.2)	0.31
BMI (kg/m ²)	24.08±3.46	23.7±5.1	0.59
History of alcohol use			0.79
Moderate	31 (47.7)	58 (42.6)	
Social	20 (30.8)	45 (33.1)	
None	14 (21.5)	33 (24.3)	
History of tobacco use			0.30
Current	34 (52.3)	59 (43.4)	
History of	12 (18.5)	38 (27.9)	
None	19 (29.2)	36 (26.5)	
Liver disease status			0.16
Hepatitis	50 (76.9)	104 (76.5)	
Cirrhosis	7 (10.8)	24 (17.6)	
HCC	8 (12.3)	8 (5.9)	
Child-Pugh class			0.94
A	50 (76.9)	104 (76.5)	
B	15 (23.1)	32 (23.5)	
C	0 (0.0)	0 (0.0)	
Log HCV RNA	5.70 [5.01–6.57]	6.08 [5.42–6.55]	0.06
HCV genotype			0.43
1	26 (40.0)	67 (49.3)	
2	38 (58.5)	67 (49.3)	
Other	1 (1.5)	2 (1.5)	
DAA regimen			0.93
Glecaprevir/pibrentasvir	57 (87.7)	116 (85.3)	
Elbasvir/grazoprevir	4 (6.2)	7 (5.1)	
Ledipasvir/sofosbuvir	3 (4.6)	10 (7.4)	
Ledipasvir/sofosbuvir + ribavirin	1 (1.5)	2 (1.5)	
Sofosbuvir + ribavirin	0 (0.0)	1 (0.7)	
Lab findings			
Hemoglobin (g/dL)	13.58±1.59	13.89±1.71	0.22
WBC (×10 ³ /μL)	5,544.46±1,360.3	5,993.85±1,962.82	0.06
Platelet (×10 ³ /μL)	188.86±63.81	187.59±61.78	0.89
AST (IU/L)	64.55±51.26	59.86±41.97	0.56
ALT (IU/L)	70.81±64.34	58.37±60.9	0.21
Bilirubin (mg/dL)	0.77±0.42	0.81±0.5	0.58
Protein (g/dL)	7.52±0.67	7.38±0.58	0.12
Albumin (g/dL)	4.09±0.47	4.16±0.44	0.93
Globulin (g/dL)	3.43±0.55	3.21±0.46	0.007
Prothrombin time (INR)	1.03±0.08	1.03±0.1	0.93
Creatinine (mg/dL)	0.91±0.56	0.85±0.46	0.46
Cholesterol (mg/dL)	163.77±38.31	171.6±39.16	0.20
Glucose (mg/dL)	112.51±25.25	118.07±44.69	0.36
AFP (ng/mL)	5.76±6.31	9.07±30.09	0.40
FIB-4	3.36±2.84	3.48±3.19	0.79
APRI	0.79±0.68	0.93±0.86	0.21
TE (kPa) (n=30/72)	8.70 [5.18–11.18]	6.80 [4.90–12.40]	0.30

Continuous variables are expressed as mean ± SD or median [IQR], and the categorical variable are indicated as n (%). PreTx, pretreatment; AFP, alpha fetoprotein; ALT, alanine aminotransferase; APRI, AST to Platelet Ratio Index; AST, aspartate aminotransferase; BMI, body mass index; DAA, direct acting antiviral; FIB-4, Fibrosis-4 index; MELD, Model for End-Stage Liver Disease; HCC, hepatocellular carcinoma; HCV, hepatitis C virus; INR, international normalized ratio; IQR, interquartile range; SD, standard deviation; TE, transient elastography; WBC, white blood cell count.

Table S3 Changes of ANA positivity between pre-DAA and post-DAA

Variables	ANA (+) → (+) (n=37)			ANA (+) → (-) (n=28)			P value [‡]
	Pre-DAA	Post-DAA	P value [†]	Pre-DAA	Post-DAA	P value [†]	
Age (years)	67 [58–72]	NA	NA	59 [53–63]	NA	NA	0.01
Female	16 (43.2)	NA	NA	13 (46.4)	NA	NA	0.80
BMI (kg/m ²)	24.27±3.82	NA	NA	23.82±2.96	NA	NA	0.60
History of alcohol use			NA			NA	0.50
Moderate	20 (54.1)	NA		11 (39.3)	NA		
Social	10 (27)	NA		10 (35.7)	NA		
None	7 (18.9)	NA		7 (25.0)	NA		
History of tobacco use			NA		NA	NA	0.80
Current	21 (56.8)	NA		15 (53.6)	NA		
History of	0 (0.0)	NA		0 (0.0)	NA		
None	16 (43.2)	NA		13 (46.4)	NA		
Liver disease status			NA			NA	0.03
Hepatitis	27 (73.0)	NA		23 (82.1)	NA		
Cirrhosis	7 (18.9)	NA		0 (0.0)	NA		
HCC	3 (8.1)	NA		5 (17.9)	NA		
Child-Pugh class			NA			NA	0.39
A	27 (73.0)	NA		23 (82.1)	NA		
B	10 (27.0)	NA		5 (17.9)	NA		
C	0 (0.0)	NA		0 (0.0)	NA		
Log HCV RNA	5.57±1.25	NA	NA	5.66±0.87	NA	NA	0.77
HCV genotype			NA			NA	0.25
1	13 (35.1)	NA		13 (46.4)	NA		
2	24 (64.9)	NA		14 (50.0)	NA		
Others	0 (0.0)	NA		1 (3.6)	NA		
Lab findings							
Hemoglobin (g/dL)	13.75±1.63	13.69±1.97	0.80	13.27±1.53	13.57±1.62	0.71	0.23
WBC (×10 ³ /μL)	5,575.53±1,414.46	6,402.16±3,012.92	0.30	5,416.43±1,356.55	6,042.86±1,678.65	0.052	0.65
Platelet (×10 ³ /μL)	196.32±63.12	208.32±73.73	0.13	179.39±63.51	191.57±62.33	0.07	0.29
AST (IU/L)	61.11±35.09	24.77±9.69	<0.001	70.39±66.98	36.19±39.42	0.001	0.47
ALT (IU/L)	65.65±52.59	18.37±9.14	<0.001	78.89±76.9	27.92±19.4	0.001	0.41
Globulin (g/dL)	3.36±0.59	3.06±0.49	<0.001	3.56±0.5	3.07±0.41	<0.001	0.14
Protein (g/dL)	7.47±0.67	7.37±0.55	0.20	7.61±0.66	7.34±0.44	0.006	0.40
Albumin (g/dL)	4.22±0.82	4.31±0.34	0.003	4.05±0.52	4.26±0.45	0.009	0.35
Bilirubin (mg/dL)	0.76±0.37	0.76±0.5	0.54	0.79±0.5	0.9±0.63	0.15	0.80
Prothrombin time (INR)	1.03±0.08	1.04±0.09	0.58	1.04±0.08	1.03±0.09	0.99	0.89
Creatinine (mg/dL)	0.96±0.67	1.02±0.61	0.04	0.83±0.36	0.86±0.41	0.17	0.36
Cholesterol (mg/dL)	110.27±24.39	174.05±38.52	0.01	116.41±26.32	167.79±32.78	0.16	0.34
FBS (mg/dL)	164.85±38.51	106.03±19.39	0.14	161.44±38.29	120.25±51.56	0.75	0.73
AFP (ng/mL)	4.93±5.76	4.69±9.63	0.004	6.72±6.9	3.9±4.8	<0.001	0.28
FIB-4	3.32±2.89	2.3±1.92	0.003	3.4±2.78	2.96±4.51	0.007	0.92
APRI	0.74±0.62	0.37±0.3	<0.001	0.83±0.75	0.84±1.85	0.01	0.60
MELD score	3.7±5.57	4.04±4.22	0.23	2.81±3.71	3.47±3.35	0.01	0.47

Continuous variables are expressed as mean ± SD or median [IQR], and the categorical variables are indicated as n (%). [†], the P value between pre-DAA and post-DAA. [‡], the P value between ANA positive maintained group and negative converted group. +, positive; –, negative. AFP, alpha fetoprotein; ALT, alanine aminotransferase; APRI, AST to Platelet Ratio Index; AST, aspartate aminotransferase; BMI, body mass index; DAA, direct acting antiviral; FIB-4, Fibrosis-4 index; MELD, Model for End-Stage Liver Disease; HCC, hepatocellular carcinoma; HCV, hepatitis C virus; INR, international normalized ratio; IQR, interquartile range; SD, standard deviation; TE, transient elastography; WBC, white blood cell count.

Table S4 Changes of ANA negativity between pre-DAA and post-DAA

Variables	ANA (–) → (+) (n=11)			ANA (–) → (–) (n=125)			P value [‡]
	Pre-DAA	Post-DAA	P value [†]	Pre-DAA	Post-DAA	P value [†]	
Age (years)	70 [62–80]	NA	NA	58 [50–69]	NA	NA	0.01
Female	7 (63.6)	NA	NA	64 (51.2)	NA	NA	0.43
BMI (kg/m ²)	25.03±3.58	NA	NA	23.58±5.22	NA	NA	0.37
History of alcohol use			NA			NA	>0.99
Moderate	5 (45.5)	NA		53 (42.4)	NA		
Social	4 (36.4)	NA		41 (32.8)	NA		
None	2 (18.2)	NA		31 (24.8)	NA		
History of tobacco use			NA			NA	0.14
Current	8 (72.7)	NA		61 (48.8)	NA		
History of	0 (0.0)	NA		0 (0.0)	NA		
None	3 (27.3)	NA		62 (49.6)	NA		
Liver disease status			NA			NA	0.08
Hepatitis	6 (54.5)	NA		98 (78.4)	NA		
Cirrhosis	3 (27.3)	NA		21 (16.8)	NA		
HCC	2 (18.2)	NA		6 (4.8)	NA		
Child-Pugh class			NA			NA	0.13
A	6 (54.5)	NA		98 (78.4)	NA		
B	5 (45.5)	NA		27 (21.6)	NA		
C	0 (0.0)	NA		0 (0.0)	NA		
Log HCV RNA	5.79±0.79	NA	NA	5.92±0.86	NA	NA	0.63
HCV genotype			NA			NA	0.60
1	4 (36.4)	NA		63 (50.4)	NA		
2	7 (63.6)	NA		60 (48.0)	NA		
Others	0 (0.0)	NA		2 (1.6)	NA		
Lab findings							
Hemoglobin (g/dL)	12.17±1.51	12.25±1.3	0.82	14.04±1.64	14.01±1.69	0.99	<0.001
WBC (×10 ³ /μL)	5,136.36±1,268.19	5,756.36±1,464.09	0.09	6,069.92±1,998.72	6,437.12±1,939.98	0.01	0.13
Platelet (×10 ³ /μL)	162.91±56.38	170.45±55.77	0.53	194.61±80.02	198.84±60.77	0.02	0.20
AST (IU/L)	53.73±28.39	27.18±9.81	0.004	60.4±43	24.2±7.33	<0.001	0.62
ALT (IU/L)	52.55±37.65	20.91±10.84	0.008	58.88±62.61	18.71±9.03	<0.001	0.74
Globulin (g/dL)	3.45±0.45	3.09±0.44	0.03	3.19±0.46	2.9±0.42	<0.001	0.08
Protein (g/dL)	7.53±0.52	7.33±0.39	0.33	7.36±0.58	7.25±0.44	0.007	0.37
Albumin (g/dL)	4.08±0.44	4.24±0.27	0.17	4.17±0.44	4.35±0.43	<0.001	0.53
Bilirubin (mg/dL)	0.67±0.33	0.73±0.46	0.33	0.82±0.51	0.8±0.64	0.31	0.35
PT (INR)	1.01±0.06	1.01±0.06	0.78	1.03±0.1	1.04±0.11	0.59	0.44
Creatinine (mg/dL)	1.03±0.98	1.67±2.92	0.06	0.84±0.38	0.91±0.7	0.03	0.54
Cholesterol (mg/dL)	151.55±28.22	168.18±49.08	0.29	173.55±39.61	178.66±43.68	0.009	0.08
FBS (mg/dL)	108.33±15.51	101.73±13.54	0.51	118.87±46.23	111.82±31.32	0.07	0.50
AFP (ng/mL)	5.79±5.05	5.61±7.38	0.11	9.39±31.48	154.87±1,619.77	<0.001	0.71
FIB-4	4.33±3.49	3.07±2.24	0.03	3.41±3.17	1.99±1.12	<0.001	0.36
APRI	1.04±1.02	0.44±0.22	0.004	0.92±0.85	0.34±0.17	<0.001	0.65
MELD score	2.92±4.99	4.24±7.4	0.18	3.32±3.81	3.37±4.32	0.65	0.74

Continuous variables are expressed as mean ± SD or median [IQR], and the categorical variables are indicated as n (%). +, positive; –, negative. AFP, alpha fetoprotein; ALT, alanine aminotransferase; APRI, AST to Platelet Ratio Index; AST, aspartate aminotransferase; BMI, body mass index; DAA, direct acting antiviral; FIB-4, Fibrosis-4 index; MELD, Model for End-Stage Liver Disease; HCC, hepatocellular carcinoma; HCV, hepatitis C virus; INR, international normalized ratio; IQR, interquartile range; SD, standard deviation; TE, transient elastography; WBC, white blood cell count.

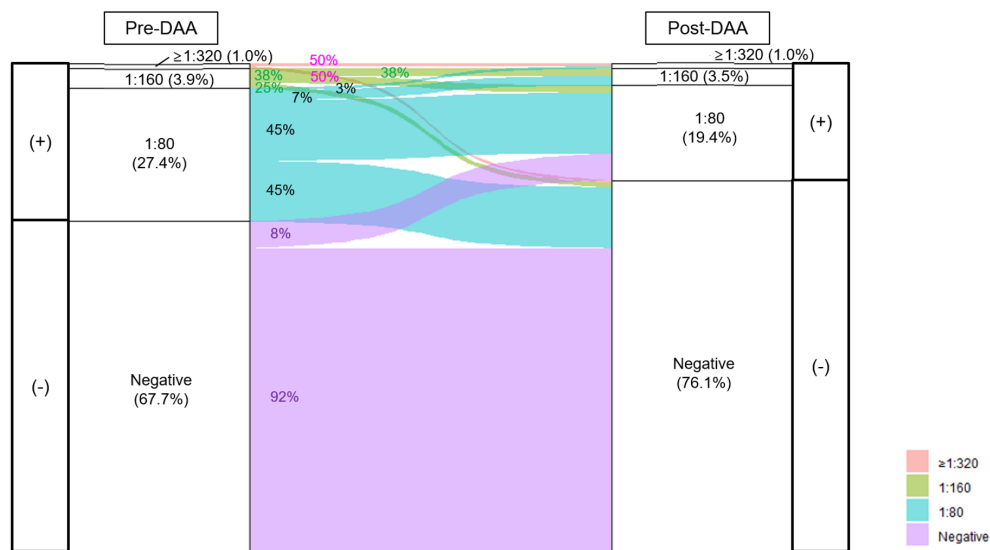


Figure S2 Changes in the ANA titers before and after DAA treatment. This alluvial plot shows the changes in ANA titers before and after DAA treatment. ANA titers declined significantly after SVR ($P=0.006$). At PreTx, 27.4%, 8%, and 1% of patients showed ANA titers of 1:80, 1:160, and $\geq 1:320$, respectively. After achieving SVR, these proportions changed to 19.4% at 1:80, 3.5% at 1:160, and 1% at $\geq 1:320$. Among patients with initial titers of 1:80, 25% transitioned to ANA negativity, while 25% maintained the status quo at 1:80 titer. For those with initial titers of 1:160, 25% underwent conversion to ANA negativity, 38% transitioned to 1:80, and 38% maintained the 1:160 titer. Among the patients who initially presented with 1:320 titer, 50% transitioned to ANA negativity, and 50% maintained the 1:320 titer. Notably, among patients who were initially ANA-negative at PreTx and transitioned to an ANA-positive status, all exhibited ANA titers of 1:80. ANA, anti-nuclear antibody; DAA, direct-acting antiviral; SVR, sustained virological response; PreTx, pretreatment.

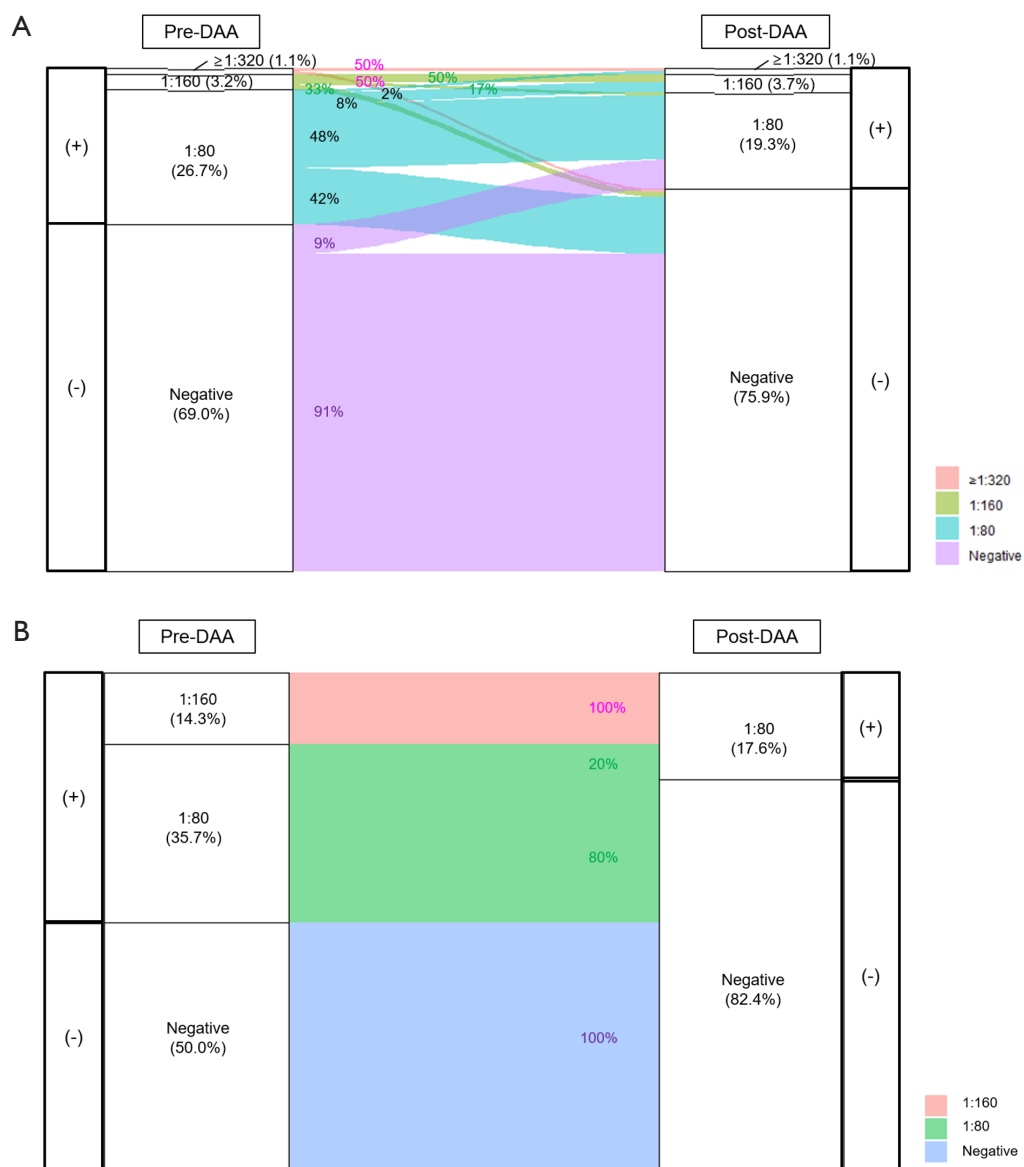


Figure S3 Changes in the ANA titers before and after DAA treatment depending on autoimmune disease. (A) No autoimmune disease. (B) Autoimmune disease. DAA, direct-acting antiviral; ANA, anti-nuclear antibody.

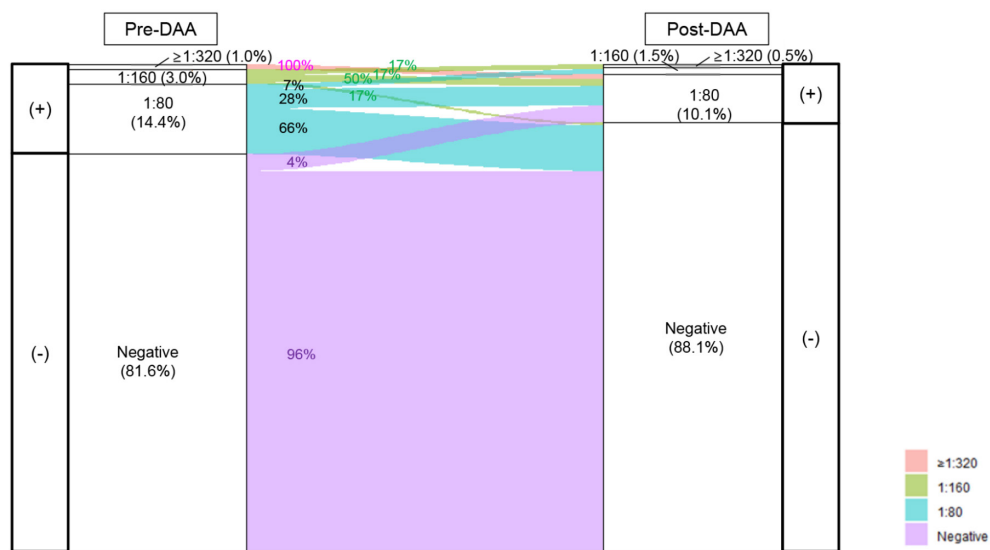


Figure S4 Changes in cytoplasmic antibody before and after DAA treatment. This alluvial plot shows changes in cytoplasmic antibody levels before and after DAA treatment with SVR. The proportion of cytoplasmic ANA positivity decreased significantly from 18.4% at PreTx to 10.9% at SVR ($P=0.006$). DAA, direct-acting antiviral; SVR, sustained virological response; PreTx, pretreatment.

Table S5 Changes of cytoplasmic positivity and patterns in HCV patients receiving DAA therapy

Variables	Pre-DAA (n=201)	Post-DAA (n=201)	P value
Cytoplasmic	37 (18.4)	22 (10.9)	0.006
Speckled	35 (17.4)	18 (9.0)	0.07
Rod and ring	2 (1.0)	4 (2.0)	–

Data are presented as n (%). HCV, hepatitis C virus; DAA, direct acting antiviral.

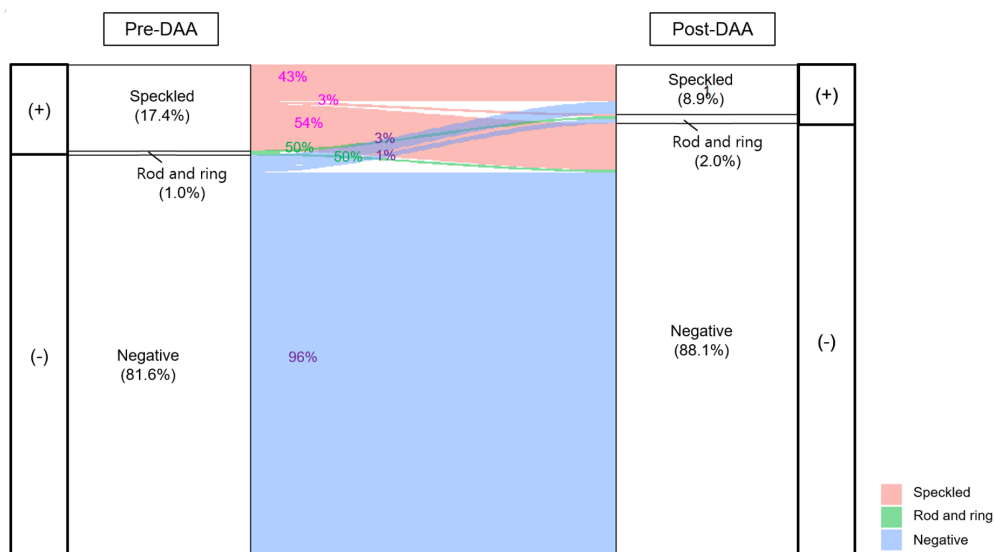


Figure S5 Changes in cytoplasmic antibody patterns before and after DAA treatment. DAA, direct-acting antiviral; SVR, sustained virological response; PreTx, pretreatment.

Table S6 Changes of ANA pattern in ANA positive HCV patients receiving DAA therapy

Variables	ANA (+) → (+) (n=37)	ANA (+) → (-) (n=28)	P value
Nuclear			0.09
Homogenous	20 (54.1)	8 (28.6)	
Speckled	14 (37.8)	13 (46.4)	
Nucleolar	0 (0.0)	4 (14.3)	
Centromere	0 (0.0)	0 (0.0)	
Mid body	1 (2.7)	1 (3.6)	
Spindle fiber	0 (0.0)	1 (3.6)	
Mixed	2 (5.4)	1 (3.6)	
Cytoplasmic	19 (51.4)	7 (25.0)	0.04

Data are presented as n (%). +, positive; -, negative. ANA, antinuclear antibody; DAA, direct acting antiviral; HCV, hepatitis C virus.