Keywords Chronic hepatitis C · Peginterferon and ribavirin · Core amino acid · Interferon sensitivity determining region

Abbreviations

CHC	Chronic hepatitis C
PEG-IFN	Peginterferon
RBV	Ribavirin
RVR	Rapid viral response
cEVR	Complete early viral response
LVR	Late viral response
ETR	End of treatment response
NR	Non response
SVR	Sustained viral response
ISDR	Interferon sensitivity determining region
Aa	Amino acid
ALT	Alanine aminotransferase
PLT	Platelet
HCC	Hepatocellular carcinoma

Introduction

A combination of pegylated interferon (PEG-IFN) and ribavirin (RBV) therapy for 48 weeks achieves a sustained viral response (SVR) rate of 40–50% in chronic hepatitis C (CHC) patients with a high viral load of genotype 1 [1–4]. The dose-reduction rate and the frequency of discontinuation of this treatment are high in aged patients [5]. The SVR rate of the therapy is lower in females than males, especially in older patients in Japan [6].

Around 30% of HCV carriers have serum alanine aminotransferase (ALT) levels within the upper limit of normal ranges [7, 8] and HCV carriers with persistently normal serum ALT (PNALT) and serum platelet (PLT) counts of over $15 \times 10^4/mm^3$ show low grade hepatic fibrosis and good prognosis [9]. Before treating HCV carriers, it is very important to predict non-response to PEG-IFN plus RBV therapy because of its medical cost, adverse effects, and its impact on the long term quality of life.

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There are many factors affecting response to IFN monotherapy and PEG-IFN/RBV therapy, including body mass index (BMI) [10, 11], steatosis [12, 13], insulin resistance [14], stage of liver fibrosis [15, 16], total cholesterol (T. Chol), triglyceride (TG), adherence to both PEG-IFN and RBV [17], race [18, 19], age [1, 2, 20], and viral factors including serum quantity of HCV RNA, HCV genotype and substitution of amino acids (aa) in the interferon sensitivity determining region (ISDR, 2209–2248) of the nonstructural protein 5A (NS5A) [21] and in the core protein [22, 23]. Early viral response is an important predictive factor in PEG-IFN/RBV therapy for CHC patients with genotype 1 and high viral loads [24–27].

The aim of this study was to elucidate the valuable predictive factors of SVR in Japanese patients with HCV genotype 1b high viral loads following 48 weeks of PEG-IFN/RBV therapy, focusing on the relationship between aa substitutions in the ISDR and at core aa 70 and 91 and early viral kinetics.

Patients and methods

Selection of patients

This retrospective study was conducted at 15 clinical sites in Japan which are part of the Study Group of Optimal Treatment of Viral Hepatitis supported by the Ministry of Health, Labor and Welfare, Japan. Eligible subjects were CHC patients, who (1) had received liver biopsy; (2) were genotype 1b with high viral load (≥100 KIU/ml by Cobas Amplicor Hepatitis C Virus Test, version 2.0) at the start of PEG-IFN/RBV therapy; (3) received weekly injections of PEG-IFN-α-2b (PEG-INTRON; Shering-Plough, Kenilworth, NJ) of 1.5 μg/kg bw and oral administration of RBV (Rebetol; Shering-Plough) for 48 weeks. The amount of RBV was adjusted based on the subject's body weight; (600 mg for ≤60 kg bw, 800 mg for 60–80 kg bw, 1,000 mg for > 80 kg bw); (4) were examined serially for quantitative and qualitative HCV RNA; and (5) the aa sequences at positions 70 and 91 in the core region and of the ISDR in the NS5A had been determined in pretreatment sera.

Hepatitis B virus (HBV) infection, human immunodeficiency virus (HIV) infection, autoimmune hepatitis, primary biliary cirrhosis, hemochromatosis, and Wilson's disease were excluded. Histopathological diagnosis was based on the scoring system of Desmet et al. [28]. The definition of alcohol abuse included patients having a history of more than 100 kg of total ethanol intake. Complete blood counts, liver function tests, serum lipids, serum ferritin, serum fibrosis markers, fasting plasma glucose (FPG), and immune reactive insulin (IRI) were examined in most cases. Written informed consent was obtained from all

patients before treatment, and the protocol was approved by the ethics committees in each site.

Study design

Four hundred and nine patients who completed 48 weeks of treatment and were followed for more than 24 weeks after treatment were enrolled in the first study (*Study design 1*).

To elucidate the effect of aa substitution of HCV core and in the ISDR on HCV dynamics, including a rapid viral response (RVR), complete early viral response (cEVR), a late viral response (LVR) and SVR, according to gender and age (<60 years $\geq$ 60 years), 201 of the 409 patients maintaining over 80% adherences to both PEG-IFN and RBV were enrolled in the second study (*Study design 2*).

Nucleotide sequencing of the core and NA5A gene

The nucleotide sequences encoding aa 1–191 (HCV core) and aa 2209–2248 (ISDR) were analyzed by direct sequencing as described by Akuta et al. [22, 27] and Enomoto et al. [21]. In brief, RNA was extracted from the sera and converted to cDNA and two nested rounds of polymerase chain reaction (PCR) were performed. Primers used in the PCR were as follows; (a) Nucleotide sequences of the core region: the first-round PCR was performed with CC11 (sense) and e14 (antisense) primers [22, 27], and the second-round PCR with CC9 (sense) and e14 (antisense) primers [22, 27]. (b) Nucleotide sequences of the ISDR in NS5A: the first-round PCR was performed with ISDR1 (sense) and ISDR2 (antisense) primers [21], and the second-round PCR with ISDR3 (sense) and ISDR4 (antisense) primers [21]. These sequences were compared with the consensus sequence of genotype 1b (HCV-J) [29]. Wild types virus encoded arginine and leucine at aa 70 and 91, respectively, and the aa substitutions were glutamine or histidine at aa 70 and methionine at aa 91.

Viral kinetic study

Serum HCV RNA levels were measured by PCR (Amplicor HCV RNA kit, version 2.0, Roche Diagnostics) using samples taken before treatment and at 4, 12, 24, and 48 weeks after the therapy. SVR was defined as HCV RNA negativity by qualitative analysis by PCR at 24 weeks after the treatment. RVR was defined as HCV RNA negativity at 4 weeks, cEVR as HCV RNA negativity at 12 weeks, LVR as HCV RNA negativity during 13–24 weeks and an end of treatment response (ETR) as HCV RNA negativity at the end of treatment. Patients who remained positive for HCV RNA at the end of the treatment and at 24 weeks after the therapy were defined as non-responders (NR).

Adherences to PEG-IFN and RBV

Adherences to PEG-IFN and RBV were assessed by separately calculating the actual doses of PEG-IFN and RBV received as percentages of the intended dosages. Adherences to PEG-IFN and RBV were divided into two groups; 80%$\leq$ and <80%.

Statistical analysis

All data analyses were conducted using the SAS version 9.1.3 statistical analysis packages (SAS Institute, Cary, NC, USA). Individual characteristics between groups were evaluated by Mann–Whitney *U* test for numerical variables or Fisher's exact test for categorical variables. Variables exhibiting values of $p < 0.1$ in the univariate analysis were subjected to stepwise multivariate logistic regression analysis. The grade of steatosis and iron deposition in liver tissue, BMI, albumin (Alb), low density lipoprotein-cholesterol (LDL-C), homeostasis model assesment-insulin resistance (HOMA-IR), ferritin, and hyaluronic acid were excluded from multivariate logistic regression analysis because of the absence of those data in more than 10% of the patients. All *p* values of $p < 0.05$ by the two-tailed test were considered statistically significant.

Results

Study design 1

Baseline backgrounds, characteristics and adherences of peginterferon and ribavirin in males and females

The treatment outcome of PEG-IFN and RBV combination therapy depends on gender in Japanese patients, so in addition to aa substitutions in the ISDR in NS5A [21] or at HCV core 70 and 91 [22, 27], we compared the baseline characteristics according to gender (Table 1). Males were younger and the grade of hepatic inflammation was milder in males. The serum levels of LDL-C, PLT count, and aa substitutions of ISDR and at core 70 and 91 did not differ significantly different between males and females. The frequency of no alcohol abuse was significantly ($p < 0.0001$) higher in females than males (Some of them are not described in Table 1).

The rates of over 80% adherences to PEG-IFN and RBV were significantly lower ($p = 0.0066$, $p < 0.00001$, respectively) in females than males. Only in those above 60 years did the rate of over 80% adherence to PEG-IFN not differ significantly between males and females, but the rate of over 80% adherence to RBV was significantly lower ($p = 0.035$) in females than males (Table 1).

Table 1 Backgrounds and characteristics of male and female patients

Factors	Gender		*p* value
	Male	Female	
No. of patients	256 (62.6%)	153 (37.4%)	
Age			
Median (range)	53 (18–73)	59 (23–75)	0.00001
F stage			
F0–2	206 (80.5%)	119 (77.8%)	0.592
F3–4	50 (19.5%)	34 (22.2%)	
Grade (A factor)			
A0–1	163 (63. 7%)	79 (51.6%)	0.026
A2–3	93 (36.3%)	74 (48.4%)	
HCV RNA load 0 week (KIU/mL)			
Median (range)	1500 (100–5000 <)	1280 (100–5000<)	0.384
ALT 0 week (IU/L)			
Median (range)	74.5 (16–504)	59 (19–391)	0.001
BMI			
Median (range)	23.6 (17.5–31.2)	22.1 (16.1–33.9)	0.00033
Alb (g/dL)			
Median (range)	4.0 (3.0–5.2)	3.8 (3.0–4.8)	0.011
LDL-C (mg/dL)			
Median (range)	97 (30–185)	90 (34–174)	0.612
T-Chol (mg/dL)			
Median (range)	167 (85–273)	176 (114–261)	0.0016
PLT count ($\times 10^4$/mm^3)			
Median (range)	17.0 (8.0–31.9)	16.4 (8.1–39.9)	0.350
Amino acid mutation of ISDR			
0–1	200 (78.1%)	121 (79.1%)	0.608
2≤	56 (21.9%)	32 (20.9%)	
Amino acid substitution of core 70			
Wild	177 (69.1%)	114 (74.5%)	0.261
Mutant	79 (30.9%)	39 (25.5%)	
Amino acid substitution of core 91			
Wild	153 (59.8%)	98 (64.1%)	0.403
Mutant	103 (40.2%)	55 (35.9%)	
PEG-IFN adherence			
<80%	41 (17.7%)	42 (30.4%)	0.0066
80%≤	190 (82.3%)	96 (69.6%)	
Ribavirin adherence			
<80%	54 (23.6%)	73 (52.1%)	<0.00001
80%≤	175 (76.4%)	67 (47.9%)	
Age: <60 years			
PEG adherence			
<80%	30 (17.8%)	23 (31.5%)	0.027
80%≤	139 (82.2%)	50 (68.5%)	
Ribavirin adherence			
<80%	27 (16.2%)	31 (42.5%)	0.000029
80%≤	140 (83.8%)	42 (57.5%)	
Age: 60 years≤			
PEG adherence			
<80%	11 (17.7%)	19 (29.2%)	0.147
80%≤	51 (82.3%)	46 (70.8%)	
Ribavirin adherence			
<80%	27 (43.5%)	42 (62.7%)	0.035
80%≤	35 (56.5%)	25 (37.3%)	

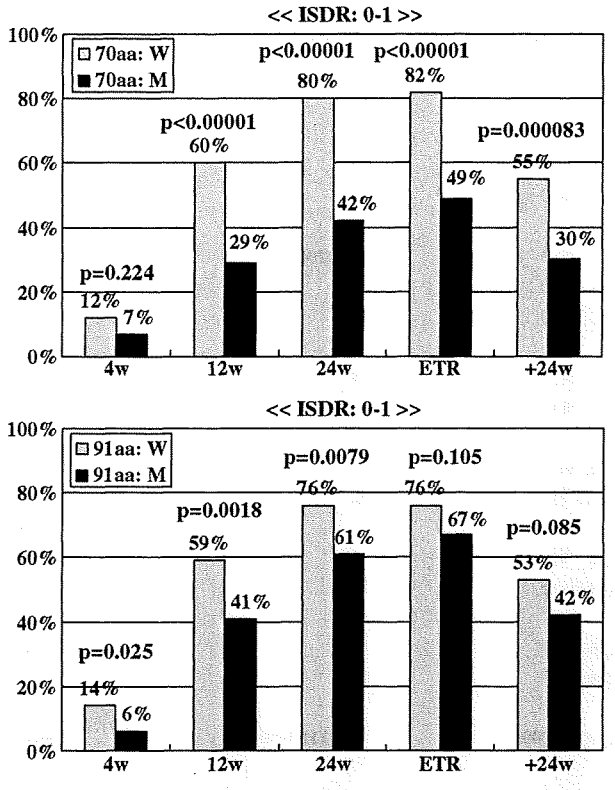

Fig. 1 Relationship between time course of serum HCV RNA negativity and amino acid substitutions in the ISDR and core amino acids 70 and 91. For cases with no or only one amino acid (aa) change in the ISDR, the rates of cEVR, LVR, ETR and SVR were significantly higher in patients with wild type core aa 70 but only the rates of RVR, cEVR, and LVR were significantly higher in patients with wild type core aa 91

Amino acid substitutions

There were no significant differences in the frequency of aa substitutions in the ISDR between males and females. Core aa substitutions at positions 70 and 91 were as follows; 291 (71.1%) were wild type and 118 (28.9%) were mutant at core aa 70, and 251 (61.4%) were wild type and 158 (38.6%) were mutant at core aa 91. There were no significant differences between males and females and between patients below and above 60 years of age.

Virological responses and aa substitutions

The rate of RVR did not differ significantly between males and females. However, more male patients showed HCV RNA negativity at 12 weeks (males vs. females; 60.7 vs. 48.4%, $p = 0.018$), 24 weeks (76.8 vs. 64.2%, $p = 0.0078$) and 48 weeks (78.2 vs. 68.6%, $p = 0.049$), and the proportion of male patients in SVR was significantly higher than that of females (61.3 vs. 37.3%, $p < 0.00001$).

RVR, cEVR and SVR rates were significantly higher in patients with two or more aa mutations in the ISDR compared to patients having no or one aa substitution in that region (20 vs. 11%, $p = 0.044$; 71 vs. 52%, $p = 0.0021$; 66 vs. 49%, $p = 0.0054$, respectively). AA substitution at core position 70 resulted in significantly lower rate of cEVR, LVR, ETR, and SVR (40 vs. 63%, $p = 0.000037$; 51 vs. 81%, $p < 0.00001$; 56 vs. 83%, 41 vs. 57%; $p < 0.00001$, $p = 0.0031$, respectively). Although the patients with the wild type aa at core 91 showed significantly higher rates of RVR and cEVR, the rate of SVR was not significantly higher in those patients (p = 0.054).

SVR rates were 30% for patients with no or one aa substitution in the ISDR and the core aa 70 substitution, and were significantly lower compared to those with the wild type aa core 70 (Fig. 1). These findings were not confirmed in patients with no or one aa substitution in the ISDR and the core aa 91 substitution (Fig. 1).

Factors affecting SVR by univariate analysis

Univariate analysis identified nine parameters that influenced non-SVR significantly: female gender, older age, advanced staged liver fibrosis, high viral load, low serum Alb level, low PLT count, no or one aa substitution in the ISDR, aa substitution at core aa 70, and low adherence to RBV (Table 2). The frequency of steatosis and HOMA-IR were significantly ($p = 0.0057$, $p < 0.00001$, respectively) lower in patients with SVR compared with non-SVR (data not shown). However, these factors were not entered in the multivariate analysis because of the absence of the data in many cases.

Factors affecting RVR, cEVR, and SVR by multivariate logistic regression analysis

Multivariate analysis identified four parameters that influenced RVR independently: low HCV RNA load, low serum ALT level, two or more aa mutations in the ISDR and the wild type aa at core position 91 (Table 3).

Concerning cEVR, male gender, mild fibrosis stage, low HCV RNA load, two or more aa mutations in the ISDR, and the wild type aa at core positions 70 and 91 were independent predictors (Table 3).

Concerning SVR, male gender ($p < 0.0001$), low HCV RNA load ($p = 0.013$), high PLT count ($p = 0.0019$), two or more aa mutations in the ISDR ($p = 0.024$), and wild type core aa 70 ($p = 0.0045$) were found to be independent predictors (Table 3).

The predictive values of the combination of gender, PLT count, ISDR and core aa 70 are shown in Fig. 2a. In male patients having PLT of $<15 \times 10^4/mm^3$, and, no or one aa substitution in the ISDR, the SVR rate was 68% when core 70

Table 2 Univariate analysis to identify the factors of SVR

Factors	Negative of HCV RNA after 24 weeks		*p* value
	(−)	(+)	
No. of patients	214 (52.3%)	195	
Gender			
Male	157 (61.3%)	99	<0.00001
Female	57 (37.3%)	96	
Age			
Median (range)	52.5 (18–75)	58 (20–74)	<0.00001
<60 years	155 (58.1%)	112	0.0018
60 years≤	59 (41.5%)	83	
Age: <60 years			
Male	118 (63.4%)	68	0.010
Female	37 (45.7%)	44	
Age: 60 years≤			
Male	39 (55.7%)	31	0.0011
Female	20 (27.8%)	52	
F stage			
F0–2	190 (58.5%)	135	0.000013
F3–4	25 (29.8%)	59	
Grade (A factor)			
A0–1	138 (56.8%)	104	0.130
A2–3	81 (48.5%)	86	
HCV RNA load 0 week (KIU/mL)			
Median (range)	1300 (100–5000<)	1700 (130–5000<)	0.016
ALT 0 week (IU/L)			
Median (range)	66 (16–391)	67 (19–504)	0.892
BMI			
Median (range)	23.0 (17.3–32.4)	23.25 (16.1–33.9)	0.714
Alb (g/dL)			
Median (range)	4.0 (3.2–5.2)	3.8 (3.0–4.8)	0.0088
LDL-C (mg/dL)			
Median (range)	94.5 (31–185)	97.5 (30–182)	0.611
T-Chol (mg/dL)			
Median (range)	169.5 (85–257)	170 (103–273)	0.511
PLT count ($\times 10^4/mm^3$)			
Median (range)	18.2 (8.7–39.9)	15.1 (8.0–31.9)	<0.00001
<15	54 (36.5%)	94	<0.00001
15≤	160 (61.3%)	101	
Amino acid mutation of ISDR			
0–1	156 (48.6%)	165	0.0054
2≤	58 (65.9%)	30	
Amino acid substitution of core 70			
Wild	166 (57.0%)	125	0.0031
Mutant	48 (40.7%)	70	
Amino acid substitution of core 91			
Wild	141 (56.2%)	110	0.054
Mutant	73 (46.2%)	85	
PEG-IFN adherence			
<80%	35 (42.2%)	48	0.063
80%≤	154 (53.8%)	132	
Ribavirin adherence			
<80%	55 (43.3%)	72	0.048
80%≤	132 (54.5%)	110	

Table 3 Multivariate logistic regression analysis to identify independent predictive factors of RVR, cEVR, and SVR

	Odds ratio	95% CI	*p* value
RVR factors selected by stepwise method			
F stage			
F0–2/F3–4	2.924	0.988–8.696	0.053
HCV RNA load 0 week (KIU/mL)			
<1000/1000≤	2.151	1.130–4.082	0.020
ALT 0 week (IU/L)			
<60/60≤	2.165	1.127–4.149	0.020
Amino acid mutation of ISDR			
2≤/0–1	2.371	1.187–4.735	0.014
Amino acid substitution of core 91			
W/M	2.137	1.021–4.464	0.044
cEVR factors selected by stepwise method			
Gender			
Male/female	1.912	1.209–3.021	0.0055
F stage			
F0–2/F3–4	2.079	1.133–3.817	0.018
HCV RNA load 0 week (KIU/mL)			
<1000/1000≤	1.608	1.002–2.577	0.049
PLT count ($\times 10^4$/mm^3)			
15≤/<15	1.427	0.882–2.309	0.148
Amino acid mutation of ISDR			
2≤/0–1	2.512	1.407–4.485	0.0018
Amino acid substitution of core 70			
W/M	2.513	1.508–4.184	0.0004
Amino acid substitution of core 91			
W/M	1.965	1.241–3.115	0.004
SVR factors selected by stepwise method			
Gender			
Male/female	3.704	2.132–6.410	<0.0001
F stage			
F0–2/F3–4	1.812	0.888–3.690	0.103
HCV RNA load0 week (KIU/mL)			
<1000/1000≤	2.024	1.163–3.534	0.013
PLT count ($\times 10^4$/mm^3)			
15≤/<15	2.469	1.394–4.372	0.0019
Amino acid mutation of ISDR			
2≤/0–1	2.148	1.107–4.170	0.024
Amino acid substitution of core 70			
W/M	2.415	1.316–4.444	0.0045
Amino acid substitution of core 91			
W/M	1.433	0.828–2.481	0.199
PEG adherence (%)			
80≤/<80	1.562	0.834–2.926	0.164

W Wild, *M* Mutant

was a wild type but only 16% in patients with mutant at core 70. In female patients, no or one aa substitution in ISDR and $<15 \times 10^4$/mm^3 of PLT count, the SVR rates were as low as 10 or 8%, irrespective of aa substitution at core 70. SVR was only 24% in patients with substitution of core aa 70 even when the PLT count was $\geq 15 \times 10^4$/mm^3. In this study, the combination analysis of PLT count, ISDR, and core aa substitution was useful for predicting non-SVR.

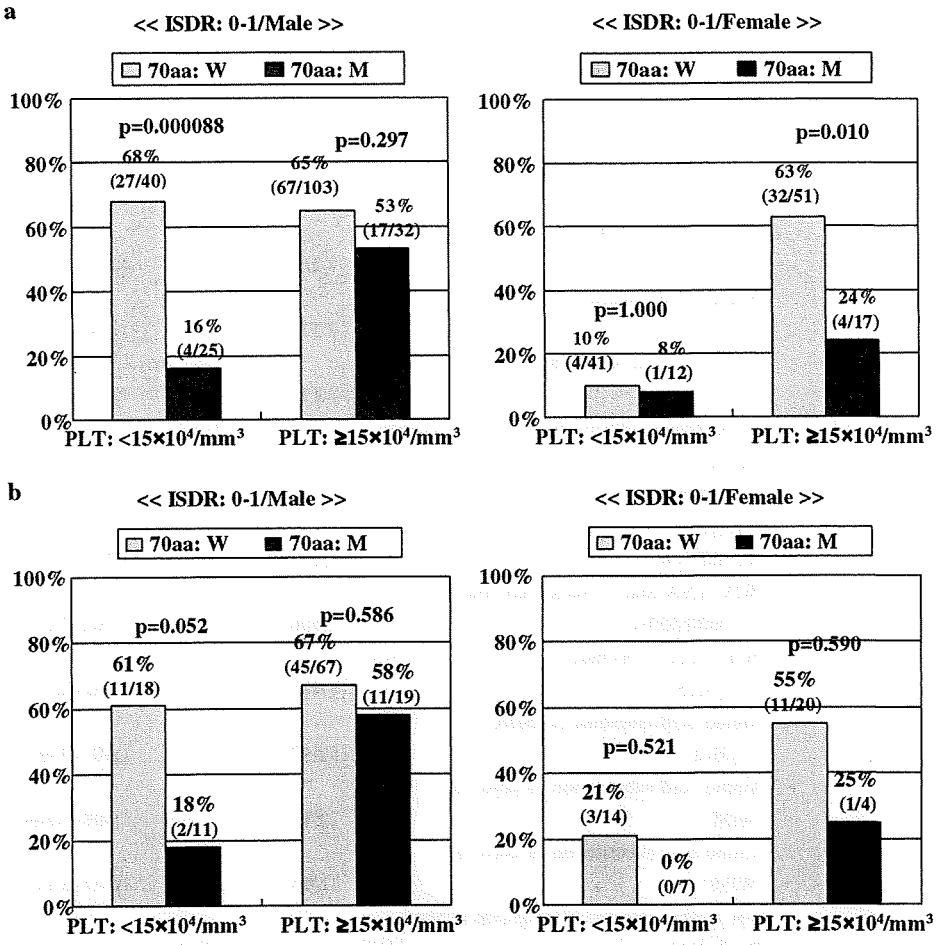

Fig. 2 Relationship between SVR rate and amino acid substitutions in the ISDR and core amino acids 70 and 91, PLT counts and gender difference. The two figures of **a** show the results of *Study 1* and the two figures of **b** show the results of *Study 2*. In male patients with no or only one amino acid (aa) substitution in the ISDR and PLT count of less than $15 \times 10^4/mm^3$, the SVR rate was 68% in those with wild type core aa 70, but only 16% in patients with mutant type of core aa 70, which is significantly different ($p = 0.000088$). There were no significant differences between wild type and mutant type of core aa 70 in the patients with no or one aa substitution in the ISDR and PLT count of over $15 \times 10^4/mm^3$. By contrast, in female patients with no or one aa substitution in the ISDR, there were no significant differences between wild type and mutant type of core aa 70 with PLT count of less than $15 \times 10^4/mm^3$, but there were significant differences between wild type and mutant type of core aa 70 with PLT counts of less than $15 \times 10^4/mm^3$ (**a**). For the patients maintaining over 80% adherences to both PEG-IFN and RBV, in males having no or one aa substitution in the ISDR and PLT counts of less than $15 \times 10^4/mm^3$, a wild type of core aa 70 could predict SVR with a positive predictive value (PPV) of 61% and negative predictive value (NPV) of 82% ($p = 0.052$). However, in male patients with PLT counts of over $15 \times 10^4/mm^3$, core aa 70 was not a useful marker for predicting SVR and non-SVR. The number of female patients with no or one aa substitution in ISDR was too small to reach a definite conclusion (**b**)

Study design 2

The basic features of 201 patients achieving 80% adherences to both PEG-IFN and RBV are as follows: the females were significantly ($p = 0.00006$) older than the males. Iron deposition in liver tissue, alcohol abuse, BMI, serum albumin level, serum ferritin level, and PLT count were significantly higher in males than females. Inflammatory activity was significantly ($p = 0.046$) higher in females than males (data not shown).

AA substitutions in the ISDR were as follows; in males 33 (22.3%) had two or more aa substitutions, in females 8 (15.1%) had two or more aa substitutions. The analysis of core aa position 70 and 91 sequences showed no significant differences in aa substitutions of either core aa 70 or 91 between males and females (data not shown).

In patients less than 60 years of age, SVR rate was significantly higher ($p = 0.0042$) in males than females, but no significant difference was noted between males and females over 60 years old. However, the number of patients over 60 years was small (Table 4).

Table 4 Univariate analysis to identify the significantly different factors between SVR and non-SVR (201 patients received over 80% adherences of both PEG-IFN and RBV)

Factors	Negative of HCV RNA after 24 weeks		*p* value
	(−)	(+)	
No. of patients	111 (55.2%)	90	
Gender			
Male	93 (62.8%)	55	0.00037
Female	18 (34.0%)	35	
Age			
Median (range)	51 (18–70)	56 (23–74)	0.00025
<60 years	91 (60.3%)	60	0.014
60 years≤	20 (40.0%)	30	
Age: <60 years			
Male	79 (66.4%)	40	0.0042
Female	12 (37.5%)	20	
Age: 60 years≤			
Male	14 (48.3%)	15	0.243
Female	6 (28.6%)	15	
F stage			
F0–2	103 (60.9%)	67	0.0012
F3–4	8 (25.8%)	23	
Grade (A factor)			
A0–1	80 (59.3%)	55	0.189
A2–3	31 (47.0%)	35	
HCV RNA load 0 week (KIU/mL)			
Median (range)	1300 (110–5000<)	1280 (130–5000<)	0.351
ALT 0 week (IU/L)			
Median (range)	74 (16–268)	67.5 (19–504)	0.752
BMI			
Median (range)	23.1 (17.3–31.0)	23.6 (16.1–33.9)	0.626
Alb (g/dL)			
Median (range)	3.95 (3.3–5.2)	3.9 (3.0–4.8)	0.079
LDL-C (mg/dL)			
Median (range)	96 (31–185)	97.5 (30–182)	0.865
T-Chol (mg/dL)			
Median (range)	170 (85–248)	170 (105–273)	0.624
PLT count ($\times 10^4/mm^3$)			
Median (range)	18.9 (8.7–30.9)	15.55 (7.2–28.4)	0.00003
<15	23 (35.9%)	41	0.00024
15≤	88 (64.2%)	49	
Amino acid mutation of ISDR			
0–1	84 (52.5%)	76	0.159
2≤	27 (65.9%)	14	
Amino acid substitution of core 70			
Wild	91 (61.5%)	57	0.0037
Mutant	20 (37.7%)	33	
Amino acid substitution of core 91			
Wild	73 (60.3%)	48	0.083
Mutant	38 (47.5%)	42	

Virological responses and aa substitution

The rates of RVR, cEVR, LVR, ETR and SVR in males and females were 12.5 versus 11.3% ($p = 1.000$), 59.6 versus 43.4% ($p = 0.053$), 74.3 versus 50.0% ($p = 0.0018$), 76.2 versus 66.7% ($p = 0.198$), and 62.8 versus 34.0% ($p = 0.00037$), respectively (data not shown). The backgrounds and characteristics of SVR and non SVR patients are shown in Table 4. There were significant differences in gender (male vs. female; $p = 0.00037$), age (<60 years vs. ≥60 years; $p = 0.014$), F stage (F0-2 vs. F3,4; $p = 0.0012$), PLT count ($<15 \times 10^4/mm^3$ vs. $15 \times 10^4/mm^3 \leq$; $p = 0.00024$), and substitution of core aa 70 (wild type vs. mutant, p = 0.0037) between SVR and non-SVR patients. The distribution of fatty change in liver tissue (≤10% vs. 11–33% vs. 34%≤; $p = 0.046$) and the grade of HOMA-IR (1.7 vs. 3.9, $p = 0.0018$) were significantly different between SVR and non-SVR (data not described in Table 4).

Factors affecting SVR by multivariate logistic regression analysis

Male gender ($p = 0.0006$), mild fibrosis stage ($p = 0.027$), and wild type of core aa 70 ($p = 0.043$) were independent predictors of SVR.

Valuable markers for predictions of sustained virological response to peginterferon and ribavirin therapy

Two or more aa mutations in the ISDR, wild type core aa 70, $\geq 15 \times 10^4/mm^3$ of PLT count, and male gender were selected statistically as independent predictors of SVR. We show here SVR rates of the patients having over 80% adherences to both PEG-IFN and RBV (Fig. 2b). In males having no or one aa substitution in the ISRD and PLT count of $<15 \times 10^4/mm^3$, wild type core aa 70 could predict SVR with a positive predictive value (PPV) of 61% and negative predictive value (NPV) of 82% ($p = 0.052$). In females, the SVR rate was very low in those who had substitution of core aa 70, but there was no significant difference between patients with wild type and substitution of core aa 70. The number of female patients was too small to provide a definite conclusion.

Discussion

The present multivariate logistic regression analysis revealed that male gender, low HCV RNA load, high PLT count, and two or more aa mutations in the ISDR and wild type core aa 70 were independent predictors for SVR. PLT count significantly decreased corresponding to the progression to the stage of liver fibrosis in CHC [9, 30, 31].

It has been considered that the low adherence level to PEG-IFN/RBV is a major cause of a significantly lower SVR rate in females and older patients [32]. The percentage of patients having over 80% adherences to both PEG-IFN and RBV was significantly lower in females than males, however, differences in the adherence to PEG-IFN/RBV between males and females were not independent predictive factors of non-SVR.

A recent report from Japan showed six or more mutations in the variable region 3 (V3) of nonstructural protein 5A (NS5A) plus upstream flanking region NS5A (aa 2334–2379), referred to as the IFN/RBV resistance determining region (IRRDR), was a useful marker for predicting SVR, but the ISDR sequence was not valuable for predicting SVR [33]. However, the number of subjects in that study was too small ($n = 45$) to reach an acceptable conclusion.

To elucidate the factors affecting low SVR rate in older female patients, we performed a multivariate logistic regression analysis using patients who achieved ≥80% adherence to both PEG-IFN and RBV. Male gender, stage of mild liver fibrosis, and wild type core aa 70 were independent predictors of SVR. In this study, blood concentration of RBV was determined in fewer than 50% of cases during treatment. Thus we cannot exclude the possibility of the effect of the blood concentration of RBV during treatment on the low SVR rate in females and older patients.

From the present analysis, it was clear that ALT, BMI, Alb, T. Chol, and adherence to RBV differed significantly between males and females, however, these factors were not independent predictors of SVR. There is a report that steatosis is an important cofactor that reduces the SVR rate in genotype 1 infected patients [34], however, such an effect was not seen in this study. Thus we could not identify the factors associated with a significantly lower SVR rate in females than males.

In the present multivariate logistic regression analyses, patients having wild type core aa 91 had significantly higher rates of RVR and cEVR, but not SVR, and patients with wild type core aa 70 had significantly higher rates of cEVR and SVR, but not RVR. Patients having two or more aa substitutions in the ISDR had significantly higher rates of RVR, cEVR, and SVR. Although several possibilities have been considered concerning the effects of aa substitutions of core protein on SVR in PEG-IFN/RBV therapy for CHC patients, the exact mechanisms have not yet been elucidated.

Recent reports have indicated that low serum IP-10 (interferon-γ inducible protein 10 kDa) [35], a higher HCV-specific CD8 cell proliferation potential [36], and a high ratio of Th1/Th2 [37] are good predictors of SVR to

PEG-IFN/RBV therapy. These results indicate the importance of immunological status and immunological response to treatment in patients difficult to treat with PEG-IFN/RBV therapy for CHC.

The present univariate analyses revealed that there were many factors relating to RVR, cEVR, and SVR including LDL-C, HOMA-IR, fatty change in liver tissue, and hyaluronic acid, however some of these factors had not been examined in some participating institutes. We consider that we must perform a prospective mass study using many factors including immunological aspects, viral factors, disease status, and therapeutic aspects to elucidate the reason that older female patients are resistant to a combination of PEG-IFN and RBV therapy in CHC with a high viral load genotype 1b.

In conclusion, our results demonstrated that wild type core aa 70, two or more aa mutations in the ISDR, low viral load, high PLT counts, and male gender are useful markers for predicting SVR.

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References

1. Manns MP, McHutchinson JG, Gordon SC, Rustgi VK, Shiffman M, Reindollar R, et al. Peginterferon alfa-2b plus ribavirin for initial treatment of chronic hepatitis C: a randomized trial. Lancet. 2001;358:958–65.
2. Fried MW, Shiffman ML, Reddy KR, Smith C, Marinos G, Gonzales FL, et al. Peginterferon alfa-2a plus ribavirin for chronic hepatitis C virus infection. N Engl J Med. 2002;347:975–82.
3. Hadziyannis S, Sette H Jr, Morgan TR, Balan V, Diago M, Marcellin P, et al. Peginterferon-alfa-2a plus ribavirin combination therapy in chronic hepatitis C. a randomized study of treatment duration and ribavirin dose. Ann Intern Med. 2004;40:346–55.
4. Hiramatsu N, Kurashige N, Oze T, Takehara T, Tamura S, Kasahara A, et al. Early decline of hemoglobin can predict progression of hemolytic anemia during pegylated interferon and ribavirin combination therapy in patients with chronic hepatitis C. Hepatol Res. 2008;38:52–9.
5. Honda T, Katano Y, Urano F, Murayama M, Hayashi K, Ishigami M, et al. Efficacy of ribavirin plus interferon-α in patients aged 60 years with chronic hepatitis C. J Gastroenterol Hepatol. 2007;22:989–95.
6. Sezaki H, Suzuki F, Kawamura Y, Yatsuji H, Hosaka T, Akuta N, et al. Poor response to pegylated interferon and ribavirin in older women infected with hepatitis C virus of genotype 1b in high viral load. Dig Dis Sci 2009;54:1317–24
7. Puoti C, Castellacci R, Montagness F, Zaltron S, Stornaiuolo G, Bergami N, et al. Histological and virological features and follow-up of HCV carriers with normal aminotransferase levels: the Italian Study of the Asymptomatic C Carriers (ISACC). J Hepatol. 2002;37:117–23.
8. Hui CK, Belaye T, Montegrande K, Wright TL. A comparison in the progression of liver fibrosis in chronic hepatitis C between persistently normal and elevated transaminase. J Hepatol. 2003;38:511–7.
9. Okanoue T, Makiyama A, Nakayama M, Sumida Y, Mitsuyoshi H, Nakajima T, et al. A follow-up study to determine the value of liver biopsy and need for antiviral therapy for hepatitis C virus carriers with persistently normal serum aminotransferase. J Hepatol. 2005;43:599–605.
10. Bressler BL, Guindi M, Tomlinson G, Heathcote J. High body mass index in an independent risk factor for non response to antiviral treatment in chronic hepatitis C. Hepatology. 2003;38:639–44.
11. Walsh MJ, Jonsson JR, Richardson MM, Lipka GM, Purdi DM, Clouston AD, et al. Non-response to antiviral therapy is associated with obesity and increased hepatic expression of suppressor of cytokine signaling 3 in patients with chronic hepatitis C, viral genotype 1. Gut. 2006;55:604–9.
12. Patton HM, Patel K, Behling C, Bylund C, Blatt LM, Vallee M, et al. The impact of steatosis on disease progression and early and sustained treatment response in chronic hepatitis C patients. J Hepatol. 2004;40:484–90.
13. Asselah T, Rubbia-Brandt L, Marcellin M, Negro F. Steatosis in chronic hepatitis C: why does it really matter? Gut. 2006; 55:123–30.
14. Romero-Gomez M, Del Mar Viloria M, Andrade RJ, Salmeron J, Diago M, Fernandez-Rodriguez CM, et al. Insulin resistance impairs sustained response rate to peginterferon plus ribavirin in chronic hepatitis C patients. Gastroenterology. 2005;128: 636–41.
15. Bruno S, Camma C, Di Marco V, Rumi M, Vinci M, Cammozzi M, et al. Peginterferon alfa-2b plus ribavirin for native patients with genotype 1 chronic hepatitis C: a randomized controlled trial. J Hepatol. 2004;41:474–81.
16. Everson GT, Hoefs JC, Seeff LB, Bonkovsky HL, Naishadham D, Shiffman ML, et al. Impact of disease severity on outcome of antiviral therapy for chronic hepatitis C: lessons from the HALT-C trial. Hepatology. 2006;44:1675–84.
17. McHutchison JG, Manns M, Patel K, Poynard T, Lindsay KL, Trepo C, et al. Adherence to combination therapy enhances sustained response in genotype-1-infected patients with chronic hepatitis C. Gastroenterology. 2002;123:1061–9.
18. Jeffers LJ, Cassidy W, Howell CD, Hu S, Reddy R. Peginterferon alfa-2a (40kd) and ribavirin for black American patients with chronic HCV genotype 1. Hepatology. 2004;39:1702–6.
19. Muir AJ, Bornstein JD, Killenberg PG, Atlantic Coast Hepatitis Treatment Group. Peginterferon alfa 2b and ribavirin for the treatment of chronic hepatitis C in blacks and non-Hispanic whites. N Engl J Med. 2004;350:2265–71.
20. Poynard T, Marcellin P, Lee SS, Niederau C, Minuk GS, Ideo G, et al. Randomized trial of interferon alpha 2b plus ribavirin for 48 weeks or 24 weeks versus interferon alpha 2b plus placebo for 48 weeks for treatment for chronic infection with hepatitis C virus. International Hepatitis Interventional Therapy Group (IHIT). Lancet. 1998;352:1426–32.
21. Enomoto N, Sakuma I, Asahina Y, Kurosaki M, Murakami T, Yamamoto C, et al. Mutations in the nonstructural protein 5 A gene and response to interferon in patients with chronic hepatitis C virus 1b infection. N Engl J Med. 1996;334:77–81.
22. Akuta N, Suzuki F, Sezaki H, Suzuki Y, Hosaka T, Someya T, et al. Association of amino acid substitution pattern in core protein of hepatitis C virus genotype 1b high viral load and non-virological response in interferon-ribavirin combination therapy. Intervirology. 2005;48:372–80.
23. Donlin MJ, Cannon NA, Yao E, Li J, Wahed A, Taylor MW, et al. Pretreatment sequence diversity differences in the

full-length hepatitis C virus open reading frame correlate with early response to therapy. J Virol. 2007;81:8211–24.
24. Davis GL, Wong JB, McHutchison JG, Manns MP, Harvey J, Albrecht J. Early virologic response to treatment with peginterferon alfa-2b plus ribavirin in patients with chronic hepatitis C. Hepatology. 2003;38:645–52.
25. Ferenci P, Fried MW, Shiffman ML, Smith CI, Marinos G, Goncales FL, et al. Predicting sustained virological responses in chronic hepatitis C patients treated with peginterferon alfa-2a ribavirin. J Hepatol. 2005;43:425–33.
26. Moucari R, Ripault M-P, Oules V, Martinot-Peignoux M, Asselah T, Boyer N, et al. High predictive value of early viral kinetics in retreatment with peginterferon and ribavirin of chronic hepatitis C patients non-responders to standard combination therapy. J Hepatol. 2007;46:596–604.
27. Akuta N, Suzuki F, Kawamura Y, Yatsuji H, Sezaki H, Suzuki Y, et al. Predictive factors of early and sustained responses to peginterferon plus ribavirin combination therapy in Japanese patients infected with hepatitis C virus genotype 1a: amino acid substitutions in the core region and low-density lipoprotein cholesterol level. J Hepatol. 2007;46:403–10.
28. Desmet VJ, Gerber M, Hoofnagle JH, Manna M, Scheuer PJ. Classification of chronic hepatitis: grading and staging. Hepatology. 1994;19:1513–20.
29. Kato N, Hijikata M, Ootsuyama Y, Nakagawa M, Ohkoshi S, Sugimura T, et al. Molecular cloning of the human hepatitis C virus genome from Japanese patients with non-A, non-B hepatitis. Proc Natl Acad Sci USA. 1990;87:9524–8.
30. Okanoue T, Itoh Y, Minami M, Sakamoto S, Yasui K, Sakamoto M, et al. Interferon therapy lowers the rate of progression to hepatocellualr carcinoma in chronic hepatitis C but not significantly in an advanced stage: a retrospective study in 1148 patients. J Hepatol. 1999;30:653–9.
31. Okanoue T, Itoh Y, Minami M, Hashimoto H, Yasui K, Yotsuyanagi H, et al. Guidelines for the antiviral therapy of hepatitis C virus carriers with normal serum aminotransferase based on platelet count. Hepatol Res. 2008;38:27–36.
32. Iwasaki Y, Ikeda H, Araki Y, Osawa T, Kita K, Ando M, et al. Limitation of combination therapy of interferon and ribavirin for older patients with chronic hepatitis. Hepatology. 2006;43:54–63.
33. El-Shamy A, Nagano-Fujii M, Sasase N, Imoto S, Kim SR, Hotta H. Sequence variation in hepatitis C virus nonstructural protein 5A predicts clinical outcome of pegylated interferon/ribavirin combination therapy. Hepatology. 2008;48:38–47
34. Patton HM, Patel K, Behling C, Bylund D, Blatt LM, Vallee M, et al. The impact of steatosis on disease progression and early and sustained treatment response in chronic hepatitis C patients. J Hepatol. 2004;40:484–90.
35. Lagging M, Romero A, Westin J, Norkrans G, Dhillon AP, Palwlosky JM, et al. IP-10 predicts viral response and therapeutic outcome in difficult-to-treat patients with HCV genotype 1 infection. Hepatology. 2006;44:1617–25.
36. Pilli M, Zerbini A, Penna A, Orlandini A, Lukasiewicz E, Pawlotsky JM, et al. HCV-specific T-cell response in relation to viral kinetics and treatment outcome (DITTO-HCV Project). Gastroenterology. 2007;133:1132–43.
37. Shirakawa H, Matsumoto A, Joshita S, Komatsu M, Tanaka N, Umemura T, et al. Pretreatment prediction of virological response to peginterferon plus ribabirin therapy in chronic hepatitis C patients using viral and host factors. Hepatology. 2008; 48:1753–60.

ウイルス肝炎検診と病診連携のすすめかた

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ウイルス肝炎検診の現状●

平成14年から19年にかけて5年間で老人保健事業に基づく基本検診のなかで40歳から5歳きざみで節目にあたる人に対して，B型・C型ウイルス肝炎検診が施行された．また，節目以外の人で輸血を受けたり，過去に大きな手術を施行されたことがあるハイリスク者では肝炎ウイルスの感染のリスクがあるため，節目外検診が施行された．B型肝炎陽性者は1.2%，C型肝炎陽性者は1.1%にみられ，一定の成果があった．しかし，大都市を中心として肝炎検診の受診率が低く，また肝炎ウイルスに感染していると判明しても専門医を受診して適切な治療が受けられた人が少ないなどの問題が指摘されている．

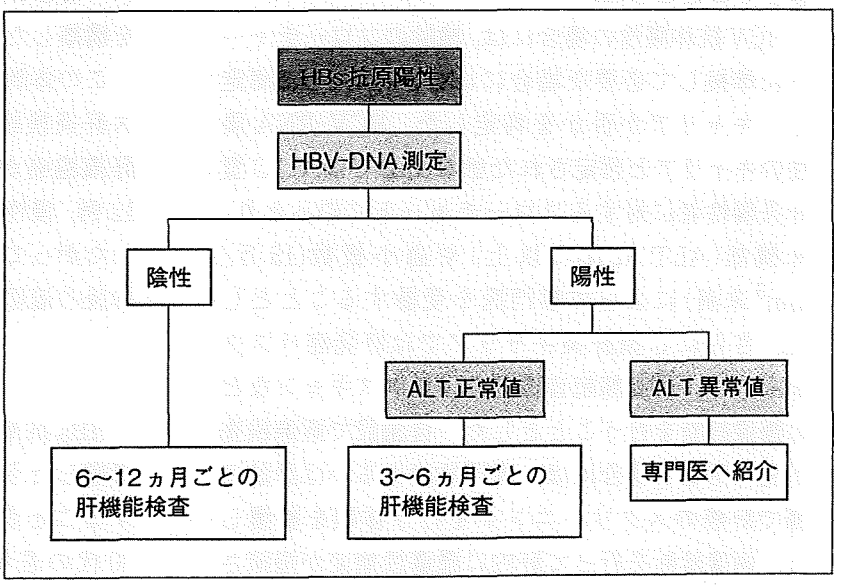
そこで平成19年度から特定感染症検査等事業として20～39歳の受診希望者と40歳以上の未受診者に対して特定感染症等検査事業を継続し，保健所のみならず医療機関で肝炎検診を受けることが可能となっている．さらに老人保健事業として新40歳の節目検診と41歳以上のハイリスク者を対象とした節目外検診が行われている．さらに政府管掌健康保険として生活習慣予防健診を受けることができる人で，検査を希望する35歳以上の人ではウイルス肝炎検診を受けることが可能になっている．まず，肝炎検診を広く行い，肝炎ウイルスキャリアを発見することが重要である．

肝炎ウイルス感染者に対する対応●

肝炎ウイルス感染者(キャリア)を発見したら，適切な対処を行い必要な治療を受けるような体制作りが大切である．平成14年にウイルス肝炎検診が始まったときに，われわれは地元の武蔵野・三鷹医師会と協議し，ウイルス肝炎検診で陽性と判明した患者に対するフローチャートを作成した(図1，2)．

HBs抗原陽性と判明した患者に対しては，腹部超音波などの画像診断を行い，肝内に腫瘍性病変がないかをチェックし，さらにHBV-DNAを測定することとした．HBV-DNA陽性で血清ALT値が異常値であった場合には，専門医を受

図1　HBs抗原陽性と判明したキャリアに対する対応(武蔵野・三鷹方式)



0910-1551/10/￥100/頁/JCOPY

●B型・C型肝炎ウイルスキャリアでは腹部エコーが必要である.
●C型肝炎感染者ではALT 30 IU/l以上または血小板15万/μl以下は専門医を紹介する.

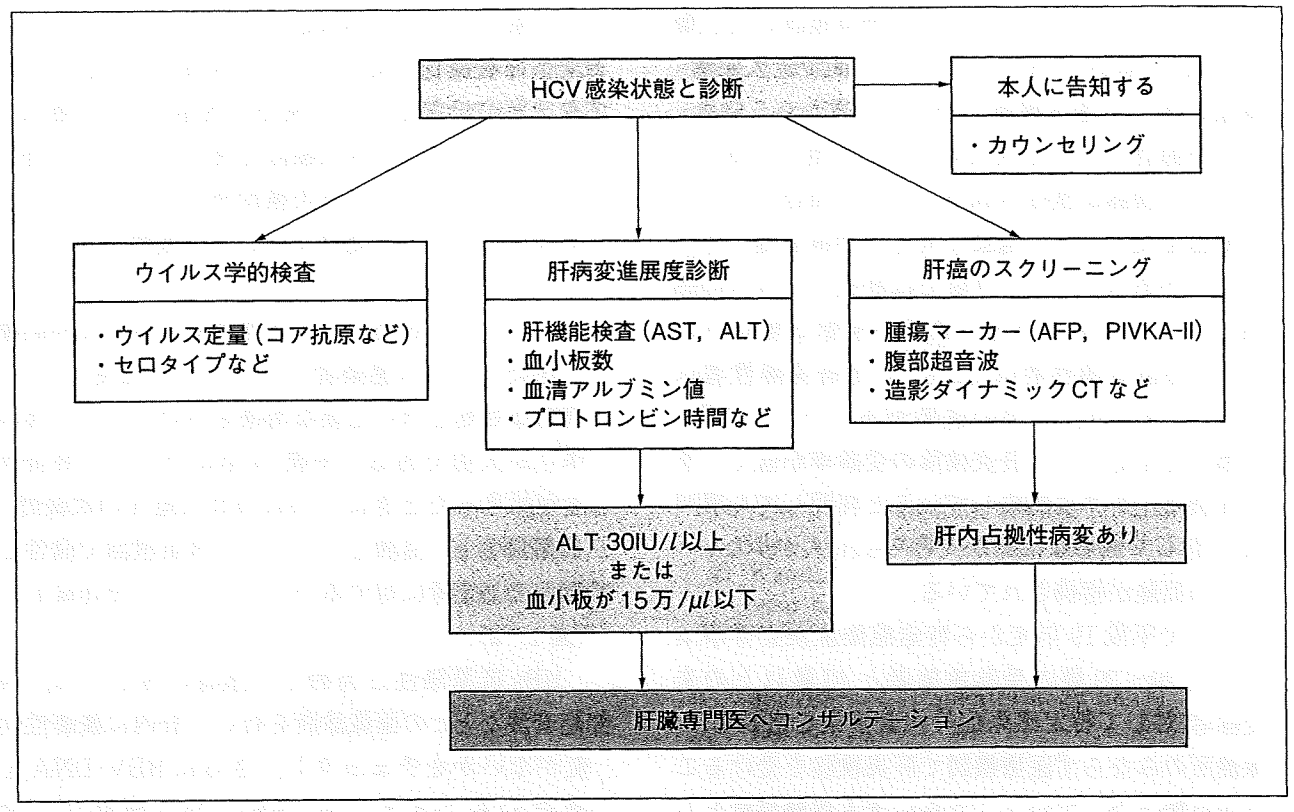


図2 C型肝炎キャリアと判明した場合の対応(武蔵野・三鷹方式)

診することとした.

HCV抗体陽性の場合には,検診のフローチャートに準拠して必要な場合にはHCV-RNAを測定し,キャリアか否かを判定した.HCV-RNA陽性のキャリアと判定された患者に対しては,C型肝炎陽性者に対するフローチャートにのっとり,肝機能(ALT 31 IU/l以上)や血小板数(15万/mm^3未満)によって専門医を受診することとした.さらにC型肝炎キャリアでは肝発癌リスクがあるため,腹部超音波や造影CTスキャンなどの画像診断を行うこととした.自施設で画像検査が施行困難な場合には,専門施設あるいは診診連携で肝癌のスクリーニングを行う体制を整備した.画像診断を行って肝内に腫瘍性病変が指摘された場合には,専門医を紹介するというシステムを構築した.

この体制で運用した成果として5年間にウイルス肝炎検診でC型肝炎陽性者と判明した5例で肝細胞癌が発見され,肝硬変と診断されたのは12例,慢性肝炎と診断されたのは72例であった.したがって肝炎ウイルスキャリアについては,検診後の適切な対応が重要である.

治療が必要なB型・C型肝炎●

HBs抗原陽性者においては,肝発癌リスクを考慮したうえで抗ウイルス療法を行う対象を設定することが重要である.B型慢性肝炎では20～30代の若年者でも肝発癌がみられる.そこで,

●B型肝炎感染者は40歳以上，HBV-DNA高値，血小板低下のいずれかの場合，専門医を受診．

●肝疾患診療では地域医療連携パスが有用である．

表1　ALTが基準値内のC型肝炎における抗ウイルス療法の適応ガイドライン

	血小板≧15万μ/ml	血小板＜15万μ/ml
ALT≦30 IU/l	2～4MごとにALTの測定．ALT異常値の時点で，完治の可能性・発癌リスクを評価し，抗ウイルス療法を考慮	線維化進展例がかなり存在することから，可能なら肝生検を施行し，F2A2以上は抗ウイルス療法を考慮．肝生検未施行例は，2～4MごとにALTを測定し，異常値を呈した時点で抗ウイルス療法を考慮
31≦ALT≦40 IU/l	65歳以下は抗ウイルス療法の適応	慢性肝炎治療に準じる

データマイニング解析を用いてHBs抗原陽性者の肝発癌リスクを検討した．その結果，年齢が40歳以上，HBV-DNAが4.0 log copies/ml以上，あるいは血小板数17万/mm^{3}以下であることが肝発癌関連因子であることが判明した．したがってこれらの3項目のいずれかを満たす場合には専門医を受診して抗ウイルス療法に適応を判定する必要がある．

C型肝炎においては，厚生労働省研究班(熊田博光班長)から血清ALT値が31 IU/l以上あるいは血小板数15万/mm^{3}未満のいずれかの場合には，インターフェロン治療を考慮する必要があるというガイドラインが示されている(表1)．したがって，これを遵守して専門医を受診させてインターフェロン治療を考慮する必要がある．

連携パスを用いた医療連携●

B型・C型肝炎の治療ガイドラインは毎年更新されている．さらに肝発癌を防止するための抗ウイルス療法の適応についても，新たな知見によって変化している．そこでかかりつけ医が的確に判定して治療適応を決定することが困難となっている．また，専門医においては多岐にわたるすべての診療を一手に行うことが困難という状況である．

そこで地域における医療連携をスムーズに行うシステム作りが必要となっている．われわれは武蔵野市と三鷹市において専門医療期間と医師会の合同で肝疾患の地域連携パスを5種類作成して運用している．5種類の連携パスは，

① 慢性肝炎の経過観察のための連携パス

② ALT正常値のC型肝炎の経過観察の連携パス

③ 難治性C型慢性肝炎治療の連携パス

④ 肝硬変治療の連携パス

⑤ 肝癌治療後の再発早期発見および再発抑止のための連携パスである．実際の連携パスの1例を図3に示した．

専門医が診療する部分は，網掛けで示しかかりつけ医が担当する部分は白抜きで表示した．紙を用いて運用し，かかりつけ医と専門医が情報共有しながら1人の患者の診療にあたることが可能である．このパスによって患者の安心感が得られ，一定レベルの医療が地域で提供できるシステムとなっている．これらの連携パスを用いて病診連携をすすめ地域ぐるみで肝癌撲滅を展開することが重要と思われる．

さらに連携パスを効果的に運用するため，専門医とかかりつけ医が定期的に会合を開いて，パスの改良を行い，症例検討を行うことによって実際の運用面での問題点を抽出していくことが大切である．

◎専門医とかかりつけ医が連携して診療にあたるため，連携パスが有用である．

◎連携パスには専門医とかかりつけ医の役割分担をもり込む．

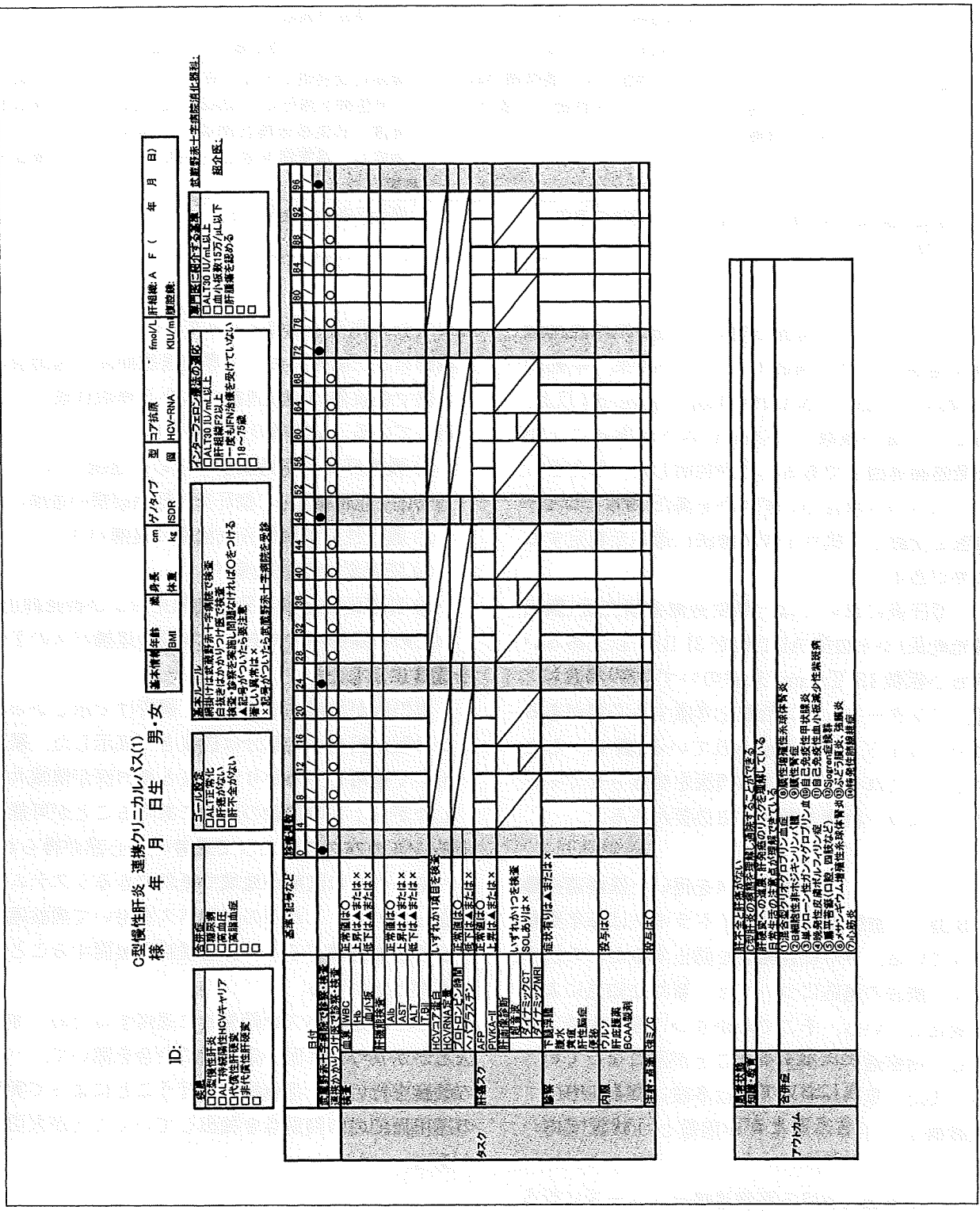


図3 C型慢性肝炎の経過観察のための連携パス（武蔵野・三鷹方式）

肝炎が発症したらどう対応すればよいでしょうか？*

泉　並木**

●回答のポイント●

1）HBs 抗原と HCV 抗体を測定し，陽性の場合には HBV DNA または HCV RNA の測定を行い感染持続の有無を調べる。
2）肝癌を併発するリスクがあるため，腹部超音波検査を 3～6 カ月に 1 回施行し，必要に応じて造影 MRI などで精査する。
3）HBV DNA 高値の B 型肝炎には核酸アナログ内服，HCV RNA 陽性 C 型肝炎に対してはインターフェロン治療の適応を判断する。
4）ディスポの器具を使用し，エリスロポエチンやヘパリンを分割投与しないなど院内感染に十分留意する。

解　説

Ⅰ．透析患者の肝炎ウイルス感染の実態

透析患者では輸血を頻回に受けるなど感染の機会が多かったため，肝炎ウイルスキャリアの感染率が高い。2007 年末における HBs 抗原陽性率は男性 2.12%，女性 1.78%であり非透析患者の HBs 抗原陽性率の 1.3%に比較して高率である。原疾患では妊娠腎や妊娠高血圧症候群が多く，透析期間が長い患者ほど陽性率が上昇している。B 型肝炎ウイルスは感染力が強く少量の血液から感染し，さらに劇症肝炎を発症する可能性があるため注意を要する。1994 年に東京の透析施設で 5 名が感染し 4 名が死亡した院内感染がみられ，さらに 1999 年に兵庫県の透析施設で 7 名の患者に感染がみられ 6 名が死亡する事例が発生している。

C 型肝炎ウイルス（HCV）の感染者は 2007 年の透析医学会の集計によると 9.84%である。統計がとられるようになった 1999 年以降次第に低下している（**図 1**）ものの，非透析患者の感染率の 1.1%に比較してきわめて高い。輸血が行われる頻度が多いことも一因であり，エリスロポエチンシリンジが導入されて HCV 陽性率は低下しているものの，十分な対策が今後も必要であることを意味している。透析患者の HCV 感染は重要な問題であり，厚生労働省の研究班（班長 秋葉隆）が組織され，重要な提言がなされている[1,2]。

* Hepatitis B and C associated with end-stage renal disease patients undergoing hemodialysis
key words：C 型肝炎ウイルス，B 型肝炎ウイルス，肝癌，hepatitis C virus，hepatitis B virus
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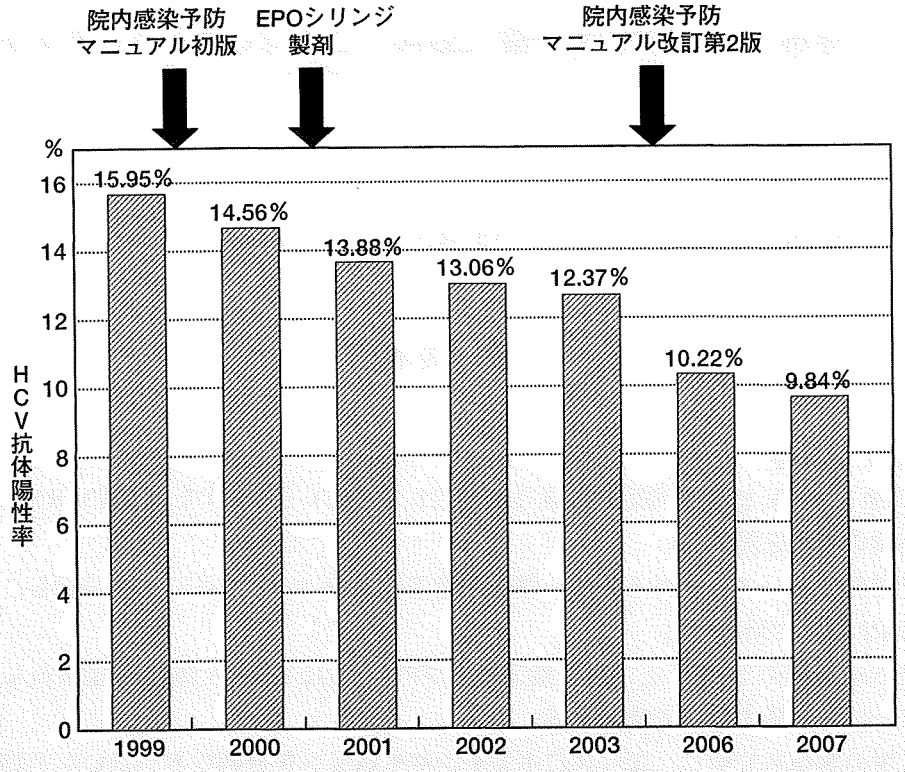

図1　慢性透析患者の HCV 抗体陽性率
（日本透析医学会「わが国の慢性透析療法の現状」より引用）

II．HBs 抗原や HCV 抗体陽性が判明した場合の患者本人の対策

HBs 抗原陽性であった場合には，その患者が現在も感染が持続しているか否かを調べる必要がある。HBV DNA の測定系が改良され，微量のウイルスから高い量まで 1 本の採血で行えるようになった。HBV DNA が陽性の場合には感染が持続していることを意味し，患者血液が感染源になり得る。現在内服抗ウイルス薬できわめて有効な核酸アナログが使用できるため，必要に応じて専門医に相談することが望ましい。

HCV 抗体陽性の場合には HCV RNA を測定する。現在リアルタイム法による測定を行えば感度良く測定できる。HCV RNA が陽性である場合には慢性肝炎である可能性があり，さらに他患者への感染のリスクがあるため，感染防止対策が必要である。C 型肝炎の場合には，感染が持続した場合，自然経過で 20～30 年で肝硬変や肝癌に移行する率が高い。特に透析患者では AST や ALT の肝機能が異常値を示さないことが多いため，慢性肝炎であることを見逃さないことが重要である。

HCV 感染者の場合には，肝癌を合併するリスクが高く[3]，早期に肝癌を発見することが必要である。定期的な腹部超音波検査が必要である。透析患者では AST や ALT などの肝機能検査が異常値とならない場合が多く（表），慢性肝炎の存在を診断することが困難である[4]。ALT 値が 20 IU/L 以上の場合には慢性肝炎の可能性が高く，肝生検を行いインターフェロン（IFN）治療の適応を判断する必要があり，肝臓専門医との連携が重要である。

III．HCV 陽性透析患者に対する IFN 治療

透析患者の HCV 感染者では血清 ALT 値に異常値がみられないまま，慢性肝炎が進行し肝細胞癌を発症する例がしばしば認められる。そこで，HCV

表　透析患者における血清 AST 値と ALT 値

HCV 抗体	AST (IU/L) 陽性	AST (IU/L) 陰性	ALT (IU/L) 陽性	ALT (IU/L) 陰性
輸血歴有	20.6±16.7 (313)	15.9±9.0 (733)	20.8±17.6 (313)	11.8±7.9 (733)
輸血歴無	23.0±15.9 (83)	13.8±6.3 (968)	23.8±24.2 (83)	12.4±7.0 (968)

カッコ内は症例数（平均±標準偏差）　（文献 4）より引用）

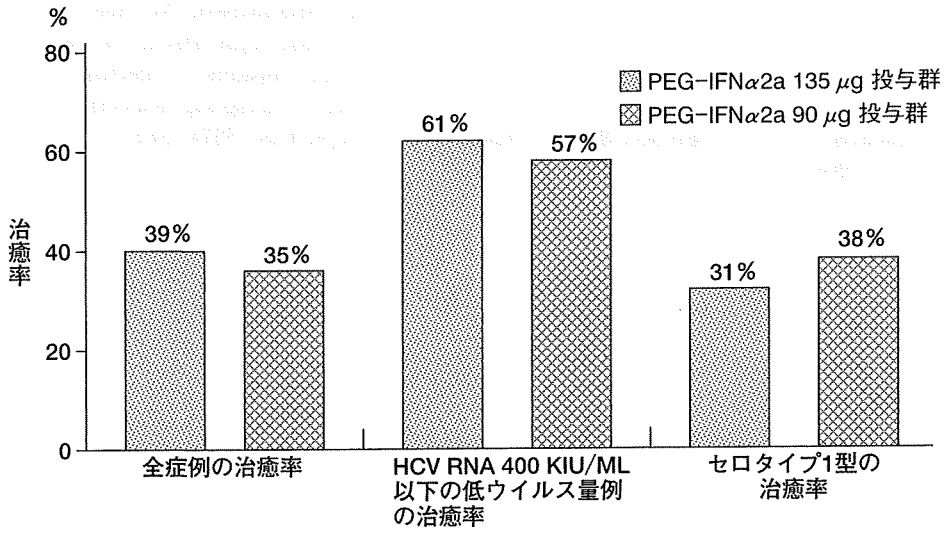

図 2　HCV 陽性透析患者に対する PEG-IFN 単独による抗ウイルス効果（文献 5）より引用）

感染者で慢性肝炎の例では IFN 治療の適応を判断して施行する必要がある。

α型 IFN による治療では透析患者では血中濃度が上昇し，脳症など副作用の頻度が高い。そこで透析患者では投与量や投与間隔を調節して投与する必要がある。さらに非透析患者の C 型慢性肝炎の難治例に用いられているリバビリン内服は，腎機能低下例では禁忌となっているため透析患者では用いることが困難である。

最近，ポリエチレングリコール（PEG）を結合させることによって作用時間を延長し，副作用を軽減した PEG-IFN 製剤が用いられるようになった。透析患者では，PEG-IFN の投与量を通常投与の 2/3 に減少させて投与した場合にウイルス排除効果が高いことが報告されており（**図 2**）[5]，週 1 回の皮下注射で忍容性が高く自覚症状の副作用が軽減されたため，わが国の透析患者での治療成績向上につながることが期待される。

IV. 感染予防対策

わが国では透析施設での肝炎ウイルスの集団感染が問題となった事例が報告されている。1994 年と 1999 年の B 型肝炎の感染により透析患者がそれぞれ 4 人と 6 人が死亡している。さらに HCV の感染事例では 1997 年に広島県の透析施設で 7 例の感染がみられ，正確な感染経路は不明であったものの抜針時の手袋を変えていなかったことなど感染防止マニュアルが遵守されていなかったことが指摘されている。また 2000 年には静岡県の透析施設で 11 名の透析患者の感染がみられ，分子生物学的解析によって同一感染源と思われ，ヘパリン生食の汚染が原因と考えられている。

そこで感染予防の対策を講じることが重要であ

る。静脈圧モニターの共有や局所麻酔薬の分注，ヘパリン生食使用など血液が付着する可能性がある場合には感染を生じさせないための対策が重要である。ディスポ注射器を用いることや処置の際に手袋を使用し，それを使い捨てにすることを徹底して行うことが必要である。普段の手洗いなどプレコーションの教育を励行しなければならない。IFN による HCV の治療は感染源を減らすという意味でも必要な対策となる。

文　献

1) 佐藤千史：透析患者における C 型肝炎の対策．肝胆膵 **57**：1005-1011，2008
2) 秋葉　隆，川口良人，黒田満彦，他：日本の透析施設における HCV 感染に関する実態調査．透析医会誌 **27**：77-82，1997
3) Sakai Y, Izumi N, Tazawa J, et al：Characteristics of anti-HCV antibody-positive patients with hepatocellular carcinoma on chronic hemodialysis：Recommendation of periodic ultrasonography for early detection. Nephron **74**：386-389, 1996
4) 酒井義法，佐藤千史：C 型肝炎の治療．透析患者の合併症とその対策―肝炎関連ウイルス―，日本透析医会・合併症対策委員会編，pp35-44，日本透析医会，2000
5) Peck-Radosavljevic M, Bolietis J, Besisik F, et al：Low-dose peginterferon alfa-2a（40 kD）（PAGASYS）to treat hepatitis C-infected end-stage renal disease patients undergoing hemodialysis：Final study results. J Hepatol 48：S374, 2008

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肝癌再発予防にIFN, PEG-IFN/Ribaはどれだけ有効か？

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