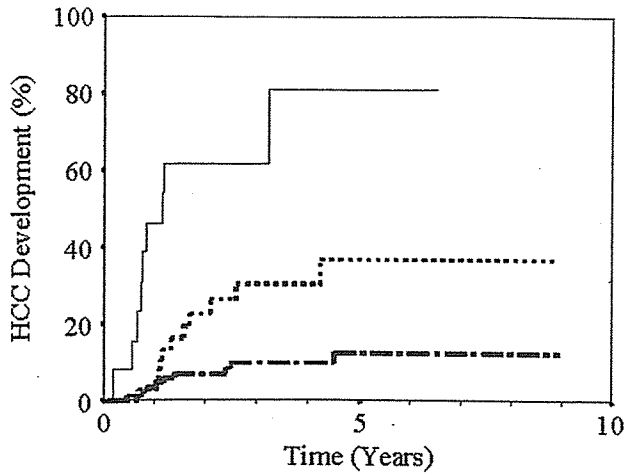

FIGURE 3. Cumulative HCC development rate from high-grade dysplastic nodules (HGDN, continuous line), low-grade DN (LGDN, dotted line), and regenerative nodules (RN, dashed line). HGDN developed into HCC more often compared with LGDN and RN ($P < 0.0001$).

TABLE 4
Factors Associated with HCC Progression from Hepatic Nodular Lesions by Multivariate Analysis (Cox Proportional Hazard Model)

Factors	Category	Hazard ratio (95% CI)	*P* value
Histology			
LRN	1	1	< 0.001
LGDN	2	2.96 (1.20–7.31)	
HGDN	3	16.8 (6.19–45.6)	
CT-AP			
Low attenuation, –	1	1	0.004
Low attenuation, +	2	3.04 (1.42–6.50)	

HCC: hepatocellular carcinoma; CI: confidence interval; LRN: large regenerative nodule; LDGN: low-grade dysplastic nodule; HGDN: high-grade dysplastic nodule; CT-AP: computed tomographic-arterial portography.

nodule on CT-AP, and liver histology on tumor biopsy were significant factors by univariate analysis. Furthermore, the etiology of chronic liver disease, serum albumin, serum bilirubin, prothrombin time, serum AFP level, serum DCP level, platelet count, US pattern, and hyperattenuation on CT-HA were not significant.

Subsequently, multivariate analysis by the Cox proportional hazard model was performed to adjust for the confounding effect on each variable. Histologic diagnosis was the most significant factor for development of HCC ($P < 0.0001$). Compared with RN, the rate of development of HCC in HGDN was as much as 16.8-fold higher (95% confidence interval [CI], 6.19–45.6), and in LGDN was 2.96-fold (95% CI, 1.20–7.31) higher. Decreased portal blood flow in the nodule on CT-AP was also significant (hazard ratio [HR], 3.04; 95% CI, 1.42–6.50; $P = 0.004$) (Table 4).

In addition, HCC developed at other sites of the liver during observation in 49 patients. However, there was no correlation between malignant transformation of the observed nodule and the subsequent development of HCC in other sites. In addition, no HCC developed from smaller nodules that were identified on first examination, during the follow-up period.

Case Reports of Hepatic Nodule Transforming to HCC during Four-Year Follow-Up

Figure 4 shows a case of HCC that progressed from a dysplastic nodule. This patient was a 61-year-old male with hepatitis C virus (HCV)-related cirrhosis. A 6-mm diameter hyperechoic nodule was found on US during the course of cirrhosis (Fig. 4A). Although detailed imaging analysis, including dynamic CT, hepatic angiography, CT-HA, and CT-AP, was carried out, the nodule was not detected with these modalities. FNAB was performed under US guidance, and the histologic diagnosis obtained from the specimen was LGDN (Fig. 5A). Therefore, the nodule was carefully followed up every 3 months on US. Three years later, the nodule was a little enlarged to 9 mm in diameter, and low-echoic foci appeared inside the nodule (Fig. 4B). Another year later, diameter of the tumor rapidly increased to 17 mm, and the nodule showed a "mosaic pattern," which is the typical sign of classical HCC on US (Fig. 4C). On this account, HCC development of the nodule was strongly suspected. Hepatic angiography was then carried out and showed typical hypervascular staining (Fig. 4D). The nodule was surgically resected afterward, and the specimen showed histologic features of moderately differentiated HCC (Fig. 5B).

Figure 6 shows another case of HCC that progressed from HGDN. This patient was a 65-year-old male with HCV-related cirrhosis. A 16-mm diameter hyperechoic nodule was found on US during the course of cirrhosis (Fig. 6A). The nodule was not detected on helical dynamic CT. Hepatic angiography, including CT-AP and CT-HA, was carried out, and a vague slightly low-attenuating area detected on both CT-AP and CT-HA (Fig. 6B–C). FNAB was performed under US guidance, and the histologic diagnosis was HGDN (Fig. 7A). Two years later, the nodule was found slightly enlarged to 20 mm in diameter on US, and the nodule became detectable on dynamic CT. (Fig. 6D). Furthermore, detailed image diagnosis was performed. Although there was no hypervascular staining on hepatic angiography, a relatively well bordered low-attenuating area was detected on CT-AP (Fig. 6E). In addition, a slightly high-attenuating rim was noted in the low-attenuating area on CT-HA (Fig. 6F). The nodule was surgically resected afterward, and the

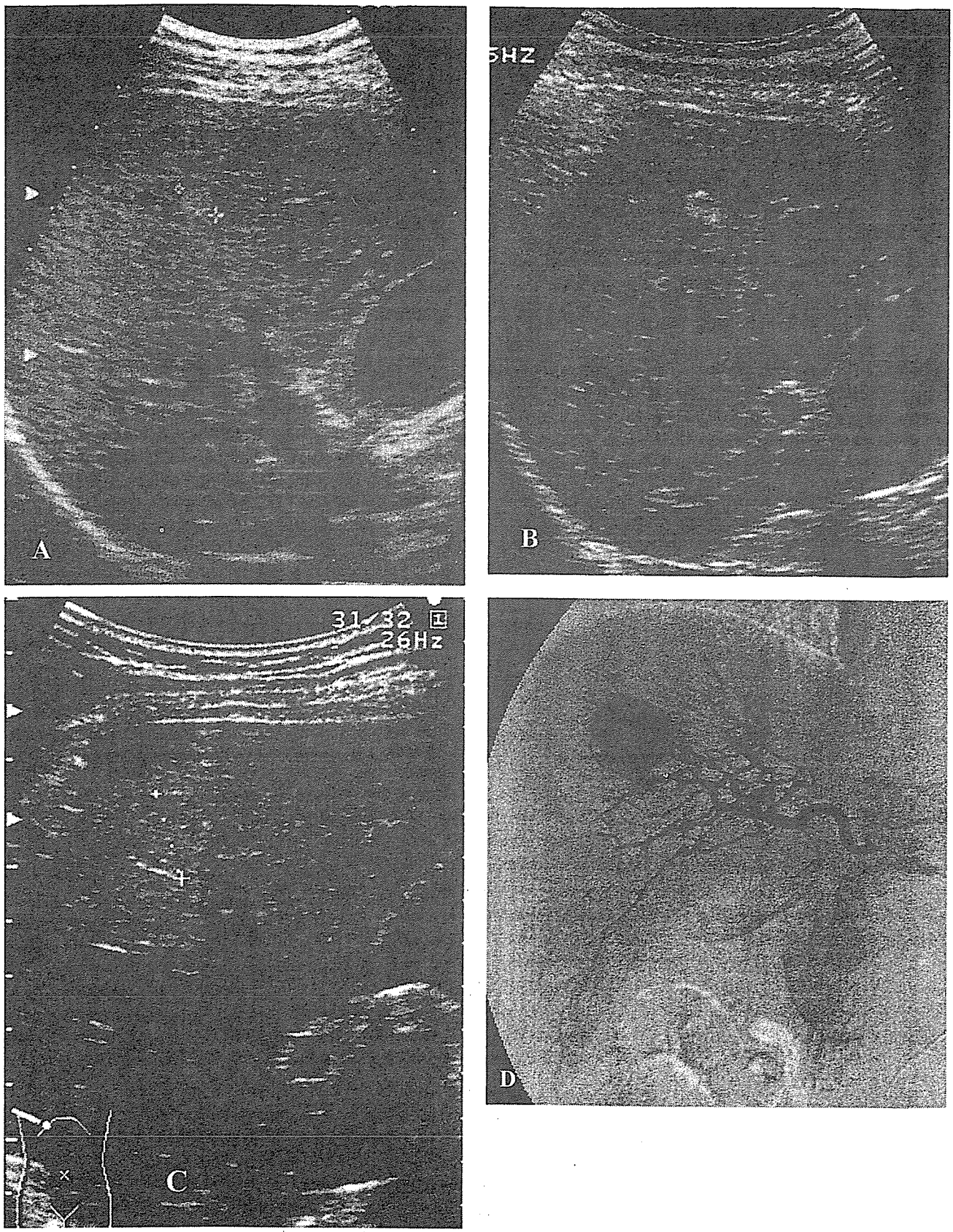

FIGURE 4. **(A)** A 6-mm diameter hyperechoic nodule appeared on ultrasonography (US) during the follow-up of HCV-related cirrhosis. **(B)** Four years later, the nodule increased in size to 9 mm in diameter and low-echoic foci appeared inside the nodule. **(C)** The nodule rapidly grew in size within 1 more year and showed a "mosaic pattern" on US. **(D)** Hepatic angiography was carried out before surgical resection. The angiogram showed typical hypervascular staining of HCC in the right lobe of the liver.

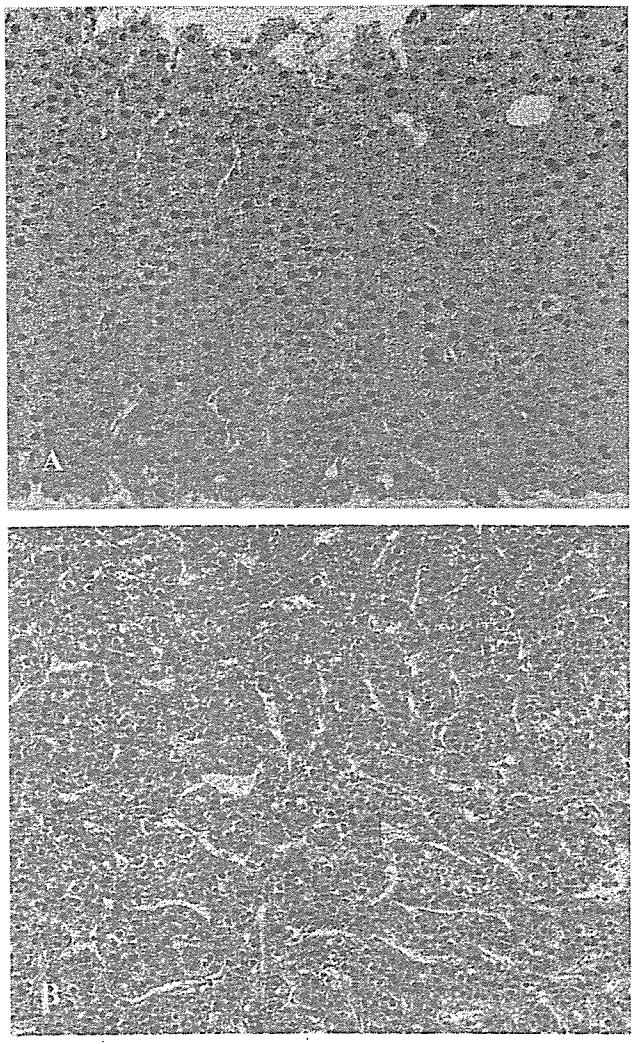

FIGURE 5. (A) Liver tissue sample obtained at first biopsy. The cell density and nuclear-cytoplasmic ratio was slightly increased (left) compared with the surrounding liver tissue (right). Small cell change was also observed and the histologic diagnosis was low-grade dysplastic nodule. (B) Liver tissue obtained by surgical resection. The tissue showed a trabecular pattern. The histologic diagnosis was well to moderately differentiated HCC. H&E staining in both panels; original magnification ×100.

specimen showed histologic features of well differentiated HCC (Fig. 7B).

Comparison of Tumor Diameter at First Biopsy and End of Observation Period

The above-mentioned analysis reviewed predictive factors for development of HCC by liver tissue diagnosis at the first tumor biopsy. When we observed a change in nodule diameter during follow-up of more than 5 mm, HCC developed in 10 of 17 patients, whereas HCC developed in only 12 of 137 nodules in which the diameter enlarged by less than 5 mm. The relation between tumor enlargement and HCC progression was statistically significant ($P < 0.001$).

DISCUSSION

The importance of dysplastic nodules as precancerous lesions of HCC is well established in Japan,[13,20–24] but less emphasized in Western countries.[25] Sakamoto et al. studied 320 resected liver tissues and concluded that multistep carcinogenesis is one pathway to HCC development.[13] Furthermore, several reports from Western countries in explanted whole liver from non-Japanese patients suggested that macroregenerative nodules may also represent precancerous lesions.[26,27]

However, consistent with previous reports, not all hepatic nodular lesions that we found on screening by US progressed to HCC.[23,24,28] Some nodules remained unchanged, and other nodules disappeared during long-term observation. Therefore, identification of true precancerous liver lesions, especially among patients with chronic liver disease, is important. The aims of the current study were to estimate the HCC progression rate of hepatic nodular lesions and to examine factors associated with malignant transformation.

Several reports have examined HCC development from borderline lesions;[23,24,28–31] however, no reports have included as many patients as the current study of 154 patients who were histologically diagnosed with dysplastic nodules and fully examined by imaging procedures before tumor biopsy. Notably, all patients also received imaging diagnosis every 3 months with a median follow-up of 2.8 years.

Twenty-nine of 154 (18.8%) hepatic nodules in our study transformed into HCC. The cumulative HCC development rates for such intrahepatic nodular lesions were 7.0% at 1 year, 19.9% at 3 years, and 27.4% at 5 years. Our findings are similar to those of Borzio et al.,[28] who reported an HCC rate of 31% in 90 large regenerative and dysplastic nodules and Seki et al.,[24] who reported HCC development in 12.1% of 33 dysplastic nodules measuring < 3 cm diameter at diagnosis.

With respect to predictive factors for development of HCC, age >60 years, ICG R15 > 30%, tumor diameter >14 mm, detection of a nodule on dynamic CT, decrease of portal blood flow in the hepatic nodule on CT-AP, and liver histology of tumor biopsy were significant factors by univariate analysis. The severity of background liver disease also has been reported as an important risk factor for development of HCC.[7–10] Therefore, high ICG R15 may affect potential HCC development. With respect to the effect of age, underlying liver disease would be expected to advance with

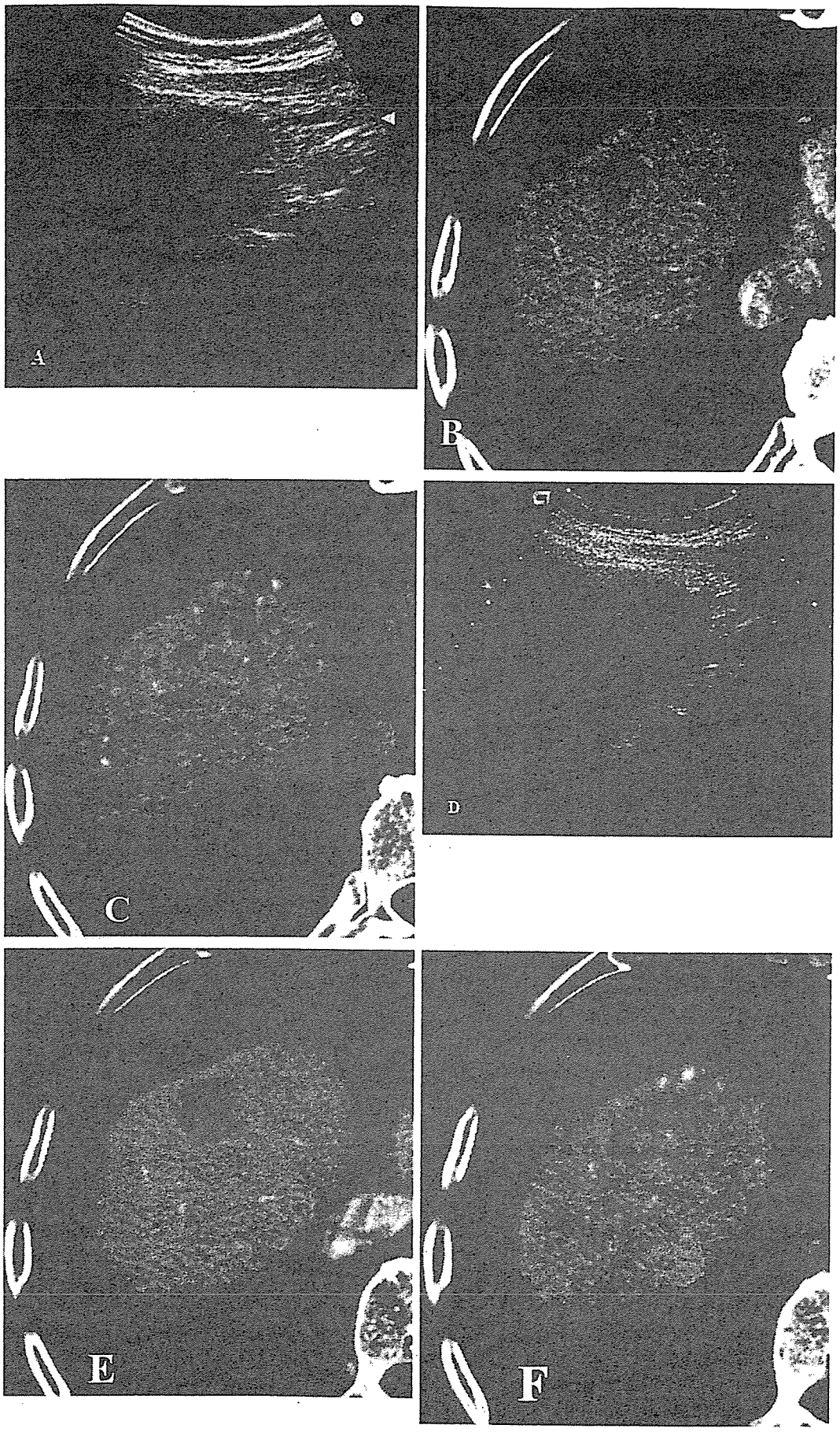

FIGURE 6

aging, and genetic mutation of oncogenes or tumor suppressor genes could occur at higher frequency with aging. We speculate that such alterations may generally increase risk of carcinogenesis with increasing age.

With respect to tumor diameter, it is intuitive that larger nodules could represent a more advanced stage in multistep carcinogenesis, and, thus, would progress to HCC more frequently than smaller ones. There is a problem of lead-time bias at this point. However, in our study, 133 of the 154 (86.4%) patients had undergone US screening before the first detection of hepatic nodules, and, thus, lead-time bias should have been minimized.

In multivariate analysis, histologic diagnosis and decrease of portal flow in CT-AP were independent factors for prediction of malignant transformation. In our study, we classified liver histology into three groups; HGDN, LGDN, and RN. According to this classification, as predicted, the progression rate of HCC from HGDN was significantly high, and the annual HCC development rate exceeded 30% in the first 2 years. The regression coefficient of HGDN in multivariate analysis was as much as 16.8 compared with RN. Therefore, we can conclude that HGDN was a true precancerous lesion of HCC.

Because of radiologic innovations, the relation between tumor progression and vascular supply of hepatic tumors is well documented. Hayashi et al.[29] followed up dysplastic nodules detected on CT-HA and CT-AP and described how portal blood flow in the nodule gradually decreased with acquisition of malignant tumor features. These results are consistent with our results in that in both univariate and multivariate analyses, reduced portal blood flow was a risk factor for HCC development. We realize that CT-AP and CT-HA are rather invasive methods and cannot easily be conducted repeatedly. Dysplastic nodules or RNs are usually isovascular or hypovascular.[29] In fact, in our patients, the arterial phase on dynamic CT was isoattenuating in 26 and low-attenuating in the remaining 9, and all 35 nodules were low-attenuating at portal venous phase and/or the equilibrium phase. Although detection of the nodule on dynamic CT was significant for predicting HCC transformation, as determined by univariate analysis, this was not significant by multivariate analysis. This could mean that CT-AP is superior to dynamic CT in detecting reduced portal blood flow and, hence, a better predictor of progression to HCC.

With respect to evaluation of tumor arterial blood flow, although we reviewed the results of CT-HA in our patients, we could not determine the relation between arterial blood flow and tumor progression. Hepatic arterial flow decreased in dysplastic nodules and in early stage HCC, and then it increased as lesions progressed to classic HCC.[29] Such a two-phase arterial

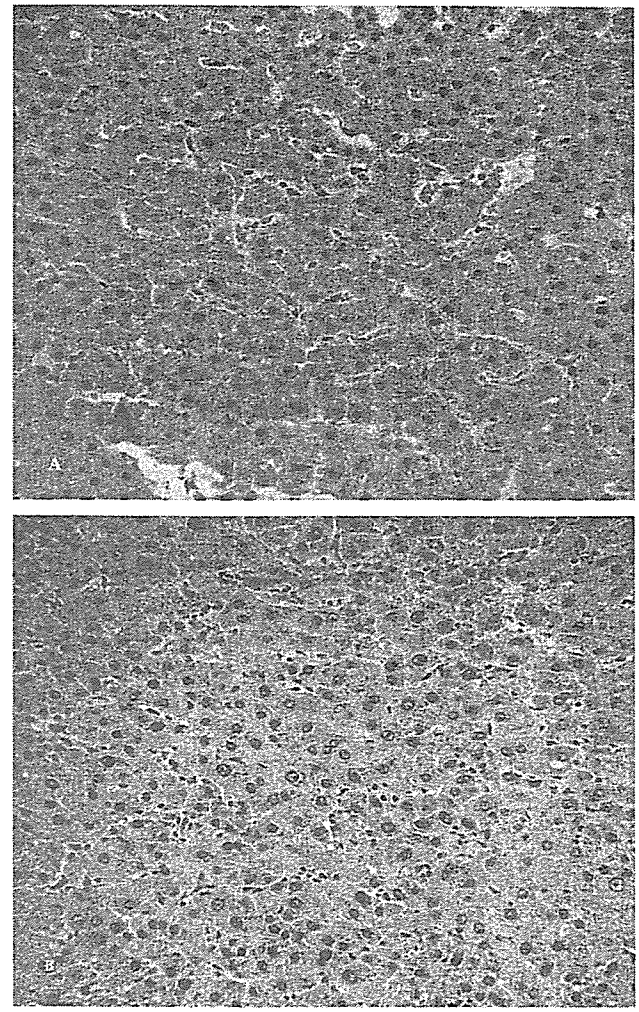

FIGURE 7. **(A)** The first histopathologic diagnosis based on examination of US-guided fine needle aspiration biopsy of the nodule was high-grade dysplastic nodule. **(B)** The nodule was subsequently resected surgically, and the specimen showed histopathologic features of well differentiated HCC. H&E staining in both panels; original magnification ×100.

FIGURE 6. **(A)** A 16-mm diameter hyperechoic nodule was found on US during the course of cirrhosis. **(B)** A vague and slightly low-attenuating area was detected on both CT-AP. **(C)** The nodule showed slightly hypoattenuating area on CT-HA. **(D)** Two years later, the nodule increased in size to 20 mm in diameter on US. **(E)** Further detailed image diagnosis was performed and a relatively well bordered low-attenuating area was detected on CT-AP. **(F)** In addition, a slightly high-attenuating rim appeared in the low-attenuating area on CT-HA.

flow change may have complicated our statistical analysis.

Previous studies showed the usefulness of MRI for characterization of hepatic nodular lesions.[32–35] Earls et al.[32] examined thin-section MRIs of explanted liver and reported that MRI depicted 41 of 42 (98%) hepatic nodular lesions, which included dysplastic nodules as well as HCC. Furthermore, Matsui et al.[33] demonstrated that hyperplastic adenomatous nodules were hyperintense on T1-weighted spin-echo imaging and hypointense on T2-weighted spin-echo imaging, and both features were useful for the differentiation of such borderline lesions from HCC in the cirrhotic liver. Furthermore, other groups also indicated the superiority of dynamic contrast-enhanced MRI and ferumoxides-enhanced MRI relative to CT for diagnosis of small HCC.[34,35] We also examined dynamic MRI in 48 patients and ferumoxides MRI in 25 patients. Because the number of these patients was small, we did not include MRI results in our analysis. Choi et al.[34] indicated that ferumoxides-magnetic resonance imaging can be used instead of CT-AP and CT-HA, because both modalities have almost the same sensitivity and high specificity for diagnosis of HCC. Further studies are needed to confirm the usefulness of ferumoxides-MRI for diagnosis of malignant transformation of hepatic nodular lesions.

There are certain limitations in our study. First, tumor biopsy was usually carried out under US guidance, and, therefore, nodules that were not observed on US were not included in this study. In our clinical practice, we sometimes find hepatic nodules that show low attenuation on CT-HA and/or CT-AP, but they are barely observed on US. Tsuchiyma et al.[31] examined repeated CT-HA and reported that 18.8% of small stained spots progressed to HCC during a mean follow-up of 29 months. Indeed, it is technically difficult to confirm such nodules histologically, and detailed investigation is required in the future.

Second, problems of sampling error and sampling variation are always inherent in this kind of examination. Indeed, nine patients in our study progressed to HCC despite histologic diagnosis of RN at first biopsy. In these cases, we cannot exclude sampling errors or sampling variation. However, to minimize such problems, we routinely recorded the scene of the US-guided biopsy on video recorder to confirm that the sample was actually from the nodule. In addition, it is possible that samples obtained by needle biopsy did not reflect the most malignant part of the nodule, particularly when the nodule was heterogeneous. On this account, we usually obtained samples from two or more parts of a nodule to prevent sampling variation. If a nodule showed a heterogeneous US pattern, we obtained samples from each part of the nodule.

We recognized limitations of biopsy diagnosis for predicting HCC progression, as nine patients whose initial histologic diagnosis was RN later developed HCC. For this reason, it is necessary to include imaging diagnostic techniques such as CT-HA or CT-AP to predict liver cancer development.

Our present results allow us to conclude that dysplastic nodules, in particular HGDN, are true precancerous lesions of HCC. Hemodynamic changes in these nodules predicted progression to HCC; however, angio-computed tomography is an invasive examination, and repeated studies are not feasible. Recently, it was reported that HCC and borderline lesions, like dysplastic or regenerative nodules, can be discriminated by Levovist contrast-enhanced US.[36,37] Because commercial use of Levovist was not possible until 1999 in our country, we have not included this technique in the current study. We are very interested in whether contrast-enhanced harmonic US is useful in predicting HCC development from hepatic nodular lesions, as this examination is less invasive than CT-HA or CT-AP and does not require hospitalization.

On the basis of changes observed during follow-up, our results indicated that enlargement of tumor diameter was the most important factor suggesting malignant transformation. Changes in US pattern also indicated HCC development. Indeed, low-echoic foci appeared in the center of hyperechoic nodules preceding HCC diagnosis in four patients who developed HCC. This finding is consistent with multistep carcinogenesis in HCC.

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Prediction model of hepatocarcinogenesis for patients with hepatitis C virus-related cirrhosis. Validation with internal and external cohorts

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***Background/Aims*: To estimate hepatocarcinogenesis rates in patients with hepatitis C virus (HCV)-related cirrhosis, an accurate prediction table was created.**

***Methods*: A total of 183 patients between 1974 and 1990 were assessed for carcinogenesis rate and risk factors. Predicted carcinogenesis rates were validated using a cohort from the same hospital between 1991 and 2003 ($n=302$) and an external cohort from Tokyo National Hospital between 1975 and 2002 ($n=205$).**

***Results*: The carcinogenesis rates in the primary cohort were 28.9% at the 5th year and 54.0% at the 10th year. A proportional hazard model identified alpha-fetoprotein (≥20 ng/ml, hazard ratio 2.30, 95% confidence interval 1.55–3.42), age (≥55 years, 2.02, 95% CI 1.32–3.08), gender (male, 1.58, 95% CI 1.05–2.38), and platelet count (< 100,000 counts/mm^3, 1.54, 95% CI 1.04–2.28) as independently associated with carcinogenesis. When carcinogenesis rates were simulated in 16 conditions according to four binary variables, the 5th- and 10th-year rates varied from 9 to 64%, and 21–93%, respectively. Actual carcinogenesis rates in the internal and external validation cohorts were similar to those of the simulated curves.**

***Conclusions*: Simulated carcinogenesis rates were applicable to patients with HCV-related cirrhosis. Since, hepatocarcinogenesis rates markedly varied among patients depending on background features, we should consider stratifying them for cancer screening and cancer prevention programs.**

***Keywords*: Cirrhosis; Hepatocellular carcinoma; Carcinogenesis; Hepatitis C virus; Simulation; Proportional hazard model; Validation; Prediction**

1. Introduction

There is increasing evidence that chronic hepatitis C virus (HCV) infection is closely associated with the occurrence of hepatocellular carcinoma (HCC) [1–4]. The incidence of patients with HCV-related HCC has increased recently in several parts of the world [5–9]. In Japan, blood transfusion and parenteral drug use became prevalent in 1960s, and patients with HCV-related cirrhosis gradually increased around 1980s. Since, an effective and truly curative therapy for a large and advanced HCC still remains limited at best, evaluation and assessment of carcinogenesis in chronic liver disease and detection at an early stage of HCC are of great importance. Reports of HCC development rates in HCV-cirrhosis differ [10–13], probably due to

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differences of patient characteristics in varied study populations. The lack of reliable data as to the natural history of cirrhosis makes it difficult to evaluate the exact role and cost-effectiveness of interferon therapy.

Platelet count has been used to predict hepatocarcinogenesis [10,13,14], but its usefulness for distinguishing the HCC appearance rate is based on discrimination between chronic hepatitis and cirrhosis [15–18]. Predicting carcinogenesis solely on the basis of platelet count is less valuable in a cohort of patients with cirrhosis, because the liver disease has already advanced to a certain stage with a uniformly low platelet count. When a cohort of patients with HCV-related cirrhosis is analyzed by platelet count, it is usually not possible to discriminate between a super-high-risk group for carcinogenesis and a relatively low-risk group. The availability of a general model that can accurately predict the HCC development rate in HCV-related disease based on readily available data would be helpful in planning the treatment of these patients. Moreover, such a model could be used for the selection and stratification of patients for clinical trials.

In this study, we tried to develop a prediction model for hepatocarcinogenesis rate, using a large cohort with a long observation period. This model was also validated with two independent patient cohorts for generalization and clinical application.

2. Patients and methods

2.1. Study population

Among 457 consecutive patients diagnosed with liver cirrhosis between 1974 and 1990 at Toranomon Hospital, Tokyo, 258 patients had positive anti-HCV antibody (second-generation anti-HCV, enzyme-linked immunosorbent assay, Dainabot, Japan), positive HCV-RNA, and negative hepatitis B surface antigen (HBsAg, radioimmunoassay, Dainabot, Tokyo, Japan). Among them, 75 patients met either of the following exclusion criteria: (1) possible association with HCC, (2) association of hemochromatosis, autoimmune liver disease, primary biliary cirrhosis, alpha-1-antitrypsin deficiency, or Wilson disease, (3) daily drinking habit of 75 g or more, (4) alpha-fetoprotein (AFP) of 400 ng/ml or higher, (5) advanced and decompensated stage of cirrhosis with encephalopathy and refractory ascites, or (6) a short follow-up period of 6 months or less. We excluded those patients with Child–Pugh [19] stage C, because of substantial difference in carcinogenesis [20,21]. Consequently, 183 patients were retrospectively analyzed for HCC appearance rate.

2.2. Background and laboratory data

Table 1 summarizes the profiles and data of the 183 patients at the time of diagnosis. The group consisted of 92 men and 91 women aged from 28 to 80 (median, 55 years). The diagnosis of cirrhosis was made by peritoneoscopy, biopsy or both in 118 patients, and by clinical symptoms with ultrasonographic findings in 55 patients. When the ultrasonography (US) showed a typical irregular-surfaced liver with coarse internal architecture in addition to overt ascites or esophageal varices demonstrated by fiberscopic examination, we regarded the disease as cirrhosis. Although 12.7% of patients (23/181) showed normal aminotransferases at the time of the diagnosis of cirrhosis, all of those patients had been followed up as having chronic hepatitis with fluctuated aminotransferases.

Table 1
Patient profiles and laboratory data at the time of diagnosis of cirrhosis (primary cohort of Toranomon Hospital between 1974 and 1990, $n = 183$)

Demography and backgrounds		
Total number	183	
Sex (M/F)	92/91	
Age, median (range)	55 (28–80)	
Diagnostic method		
Peritoneoscopy and/or biopsy	118 (64.5%)	
Clinical (ultrasonography plus varices or ascites)	65 (35.5%)	
History of blood transfusion	82 (44.8%)	
Diabetes mellitus	23 (12.6%)	
Previous medical history of chronic hepatitis	34 (18.6%)	
Interferon therapy during observation	24 (12.0%)	
Refractory ascites and/or encephalopathy	0	
Hepatitis B surface antigen, positive	0 (100%)	
Anti-hepatitis C virus, positive	183 (100%)	
Hepatitis C virus RNA, positive	183 (100%)	
Child–Pugh score A	136 (74.3%)	
Child–Pugh score B	47 (25.7%)	
Observation period (year) median (range)	10.5 (0.5–26.0)	
Laboratory data	Median (range)	Valid data
Albumin (normal, 3.9–5.1 g/dl)	3.9 (2.5–5.1)	183
Bilirubin (normal, 0.3–1.1 mg/dl)	1.1 (0.4–4.4)	183
Aspartic transaminase (normal, $\leq$38 IU/L[a])	69 (17–372)	181
Alanine transaminase (normal, $\leq$50 IU/L[a])	56 (9–282)	181
Platelet (normal, 149–315$\times 1000^3$/mm^3)	95 (33–213)	183
ICG R15[b] (normal, $\leq$10%)	27 (6–81)	173
Prothrombin time (normal, $\geq$70%)	79 (54–100)	183
Gamma-globulin (normal, $<$1.5 g/dl)	1.9 (1.0–3.5)	174
Alpha-fetoprotein (normal, $<$5 mg/L)	16.5 (3–256)	166
HCV genotype[c]		
1b	107 (69.9%)	153
2a/2b	39 (25.5%)	
Combined/others	7 (4.6%)	
Not examined	30	

[a] Numbers of normal aspartic and alanine transaminases were 25 (13.8%) and 69 (38.1%), respectively. Both transaminases were normal at the time of the diagnosis of cirrhosis in 23 patients (12.7%).
[b] ICG R15: indocyanine green retention rate at 15 min.
[c] HCV genotyping was classified according to Simmonds et al. [22].

HCV-RNA measurement and HCV genotyping [22] are analyzed with nested polymerase chain reaction using initial sera stored at -80 °C.

2.3. Follow-up of patients and diagnosis of hepatocellular carcinoma

Patients were followed-up monthly following the diagnosis of cirrhosis by monitoring hematological and biochemical data. Diagnostic imaging by US was taken approximately once a year in each patient. After 1987, imaging procedures with US or computerized tomography (CT) were performed twice or more per year in the majority of patients for early detection of HCC. HCC was diagnosed by typical hypervascular characteristics on angiography. When combined use of imagings could not demonstrate a typical image of HCC (13/107, 12.1%), a fine needle biopsy was obtained for microscopic examination.

Twenty-four patients (13.1%) received interferon during the follow-up period. Since the therapy could affect the natural clinical course of viral hepatitis, they were treated as censored at the time of the initiation of interferon in the analysis. Sixteen (8.7%) cases were lost to follow-up, and median observation period was 10.5 years (range, 7.0–14.9). Those patients lost to follow-up were treated as censored data in the following statistics.

Any death unrelated to liver disease and cirrhosis-related liver failure were also classified as withdrawal and regarded as a censored case.

2.4. Statistical analysis and predictive model for carcinogenesis

The HCC development rate was analyzed using Kaplan–Meier technique [23] and differences in curves were tested using the log-rank test. The independent risk factors associated with the rate of HCC development were studied using stepwise method of non-time-dependent Cox regression analysis [24]. Potential risk factors assessed for liver carcinogenesis included the following 16 variables: age, sex, HCV genotype, association of diabetes mellitus, total alcohol intake (cumulative alcohol intake ≥200 kg), family history of liver disease, history of blood transfusion, association of ascites, serum aspartic transaminase (AST), alanine transaminase (ALT), gamma-glutamyl transpeptidase (GGT), globulin, platelet count, AFP, indocyanine green retention rate at 15 min (ICG R15), and Child–Pugh score [19]. Each variable was transformed into categorical data consisting of two simple ordinal numbers (zero or one) for univariate and multivariate analyses. Although, proper transformation of variables were recommended in this kind of study [25], logarithmic transformation was not employed even for variables with non-symmetric distribution, because simple dichotomization also seemed reliable and robust statistically and because the simplicity was considered to bring about eventual clinical usefulness. Although, a cut-off value of 20 ng/ml proved to be an important point in our previous studies about prediction of liver cancer development in cirrhosis [10,26], other threshold values of dichotomizations were chosen from near figures to median values. In running the proportional regression analysis, care was taken to avoid overfitting the model by studying no more than one variable for every 10 events of carcinogenesis. Goodness-of-fit test together with log-minus–log plot was performed to confirm the proportionality assumption in the model. Since, missing data was not replaced, reduced numbers of cases were use in multivariate analysis. A P-value of less than 0.05 was considered to be significant.

The prognostic model was generated using Cox's regression procedure from the database of the 183 cirrhotic patients in Toranomon Hospital from 1974 to 1990. Using a final model for prediction of HCC appearance, carcinogenesis rate was predicted by substituting the corresponding ordinal numbers (zero or one) for every significant covariate in a given condition of the patients. Simulated carcinogenesis rates were computed for each state consisting of all statistically significant variables.

An internal and external cohorts of patients with HCV-positive cirrhosis verified the predicted carcinogenesis rates and curves: a cohort of 302 patients with HCV-cirrhosis diagnosed at Toranomon Hospital between 1991 and 2003 (internal validation group), and a cohort of 205 patients diagnosed at Tokyo National Hospital, Tokyo, Japan, between 1975 and 2002 (external validation group). The actual survival rates were calculated by the Kaplan–Meier technique in each risk group from the two validation cohorts, and evaluated by log-rank test according to the procedures of Christensen et al. [27].

Data analysis was performed with SAS version 9.1.3 software (SAS Institute, Inc., NC, USA).

The Human Ethics Review Committee of Toranomon Hospital approved the study protocol.

3. Results

3.1. Rate of hepatocellular carcinogenesis and risk factors

During the observation period, 107 (58.5%) out of 183 patients with HCV-related cirrhosis developed HCC. The cumulative HCC appearance rates of all patients were 15.0% at the end of the 3rd year, 28.9% at the 5th year, 37.8% at the 7th year, and 54.0% at the 10th year. Crude HCC development curve was drawn together with those of internal and external validation cohorts (Fig. 1).

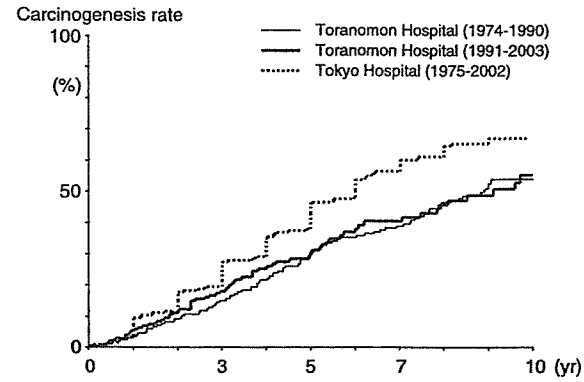

Fig. 1. Cumulative hepatocellular carcinogenesis rates in 183 patients who were diagnosed with HCV-related cirrhosis at Toranomon Hospital between 1974 and 1990. The 5th and 10th year rates were 28.9 and 54.0%, respectively (solid thin line). HCC appearance curves were also drawn in the internal (solid thick line) and external (dotted thick line) validation cohorts. The cancer appearance rate of Tokyo Hospital was significantly higher than those of the other two cohorts from Toranomon Hospital (P=0.0015, log-rank test).

Carcinogenesis rate in Tokyo Hospital was significantly higher than that of Toranomon Hospital (log-rank test P= 0.0015). The risk factors for carcinogenesis were explored using non-time dependent proportional hazard analysis. In the final step of multivariate analysis, AFP (P<0.001), age (P=0.001), sex (P=0.030), and platelet count (P=0.031), were identified as independent significant predictors of future HCC appearance (Table 2). The hazard ratio of patients with AFP value of ≥20 ng/ml was 2.30 compared with those with lower AFP value, and the hazard ratio of patients of ≥55 years of age was 2.02 compared with younger patients. Child–Pugh score did not affect the carcinogenesis rate independently.

As for 23 patients with normal aminotransferases initially, 5- and 10-years carcinogenesis rates were 27.3 and 39.4%, respectively.

3.2. Simulation of carcinogenesis rates in patients with each prognostic factor

Simulated carcinogenesis curves were generated in each patient group with the Cox proportional hazard model by substituting the corresponding value for each parameter. Based on the four significant covariates, a total of 16 carcinogenesis curves were drawn, and simulated carcinogenesis rates were also estimated in the subgroups. To facilitate the practical use of the prediction model for carcinogenesis rate, we tabulated the results of estimated HCC appearance rates at the end of the 5th and 10th year (Table 3), in which calculated rates for a patient could be easily found for a given set of patient parameters (AFP, age, platelet and gender).

The model showed that when a patient is a male younger than 55 years, with a platelet count less than 100,000/mm^3 and an AFP value less than 20 ng/ml, the estimated hepatocarcinogenesis rates are 19% at the end of the 5th

Table 2
Factors associated with hepatocarcinogenesis (compensated cirrhosis, $n=183$, 1974–1990 cohort of Toranomon Hospital)

Factors	Category	No. of primary cohort	*B*	SE	Hazard ratio (95% CI)	*P*
Alpha-fetoprotein	0: <20 (ng/ml)	97			1	
	1: ≥20 (ng/ml)	69	0.83	0.20	2.30 (1.55–3.42)	<0.001
Age	0: <55 (year)	80			1	
	1: ≥55 (year)	103	0.74	0.22	2.02 (1.32–3.08)	0.001
Sex	0: Female	91			1	
	1: Male	92	0.46	0.21	1.58 (1.05–2.38)	0.030
Platelet count	0: ≥100,000/mm^3	87			1	
	1: <100,000/mm^3	96	0.43	0.20	1.54 (1.04–2.28)	0.031

year and 43% at the 10th year. The highest carcinogenesis rates were computed for males 55 years or older with a low platelet count and a high AFP value (64% at the 5th year, 93% at the 10th year), while the lowest estimated rates were found in females younger than 55 years with a high platelet count and a low AFP value (9% at the 5th year, 21% at the 10th year).

3.3. Validation of the prediction values of carcinogenesis rate

The reliability of the estimated HCC development rates was validated using internal (Toranomon Hospital, 1991–2003) and external (Tokyo National Hospital, 1975–2002) cohorts consisting of patients with HCV-related cirrhosis. Table 4 shows brief characteristics of patients in the two cohorts.

Since, HCC development curves were coarse and unreliable when a subgroup consisted of fewer patient number than 15, six figures of carcinogenesis curves were shown in principal subgroups consisting of ≥20 patients in each validation cohort (Fig. 2). When the parameters for all of the four significant covariates were at their worst (male ≥55 years, AFP≥20 ng/ml, and platelet count<100,000/mm^3), the simulated carcinogenesis rates were 64% at the end of the 5th year and 93% at the 10th year. On the other hand, the actual carcinogenesis rates in the internal and external validation cohorts were 54.9 and 61.5% at the 5th year, and 100 and 100% at the 10th year, respectively. The latter curves corresponded significantly with the simulation-generated carcinogenesis rate (Fig. 2a). Similarly, the other five simulated carcinogenesis curves were compared with both internal and external validation cohorts (Fig. 2b–f). Although the remaining 10 curves were not shown because of lack of sufficient patient number in the subgroup, actual carcinogenesis curves for the internal and external cohorts showed very analogous rates to the simulated ones, indicating that the simulation effectively predicted the future carcinogenesis rates. When we compared actual carcinogenesis rates in the validation groups with their calculated simulation values, 74.0% (375/507) and 70.4% (357/507) of the validation values for their 5th and 10th rates were coincident with those of predicted ones and stayed in an interval between +10% and −10% of simulated values. Although those patients in a large cohort consisting of 15 patients or more (e.g. Fig. 2a–f) usually showed a reliable and consistent values with simulated ones, those in a small cohort often revealed a labile and different values from simulated ones.

When a combined patient group of the three cohorts was analyzed, the same factors proved to affect the HCC appearance rate significantly: AFP (hazard ratio 2.19, $P<0.001$), age (1.96, $P<0.001$), sex (1.80, $P<0.001$), and platelet count (1.51, $P=0.009$). Hazard ratios with 95% confidence interval and *P*-values were also calculated in the individual validation groups (Table 5).

In addition, we evaluated the 'group factor' (study group, internal, and external validation groups) as a covariate in ordinary proportional hazard analysis for a combined patient group. Although, the internal and validation groups showed a slightly low (0.90) and high (1.26) hazard ratios for HCC development compared with that of the study group, the other four factors proved to show higher hazard ratios in the model (Table 6).

Table 3
Simulated carcinogenesis rates in stratified patient groups according to gender, age, platelet count, and alpha-fetoprotein value

Gender	Age (years)	Platelet	Alpha-feto-protein (ng/ml)	Simulated carcinogenesis rate (%)	
				5-year	10-year
Men					
	<55	<100,000/mm^3	<20	19	43
			≥20	42	77
		≥100,000/mm^3	<20	13	31
			≥20	32	65
	≥55	<100,000/mm^3	<20	32	65
			≥20	64	93
		≥100,000/mm^3	<20	23	50
			≥20	50	83
Women					
	<55	<100,000/mm^3	<20	13	30
			≥20	30	61
		≥100,000/mm^3	<20	9	21
			≥20	22	47
	≥55	<100,000/mm^3	<20	22	49
			≥20	49	83
		≥100,000/mm^3	<20	16	37
			≥20	37	69

Table 4
Patient profiles and laboratory data of two cohorts for validation: an internal cohort (Toranomon Hospital from 1991 to 2003, $n=302$) and an external cohort (Tokyo National Hospital, $n=205$)

	Internal cohort (Toranomon Hospital, 1991–2003)		External cohort (Tokyo National Hospital, 1975–2002)	
Demography and backgrounds				
Total number	302		205	
Sex (M/F)	166/136		111/94	
Age (year)[a]	59 (28–80)		62 (13–83)	
Diagnostic method				
Peritoneoscopy and/or biopsy	128		115	
Clinical diagnosis	174		90	
Interferon therapy				
Yes	105 (34.8%)		12 (5.9%)	
No	197		193	
Observation period (year)[a]	5.3 (0.5–13.9)		7.5 (0.5–30.8)	
Laboratory examination	Internal cohort (Toranomon Hospital, 1991–2003)	Valid data	External cohort (Tokyo National Hospital, 1975–2002)	Valid data
Platelet ($\times 1000^3/mm^3$)[a]	91.5 (25–223)	302	100 (19–310)	205
Alpha-fetoprotein (ng/ml)[a]	14 (1–380)	296	15 (2–365)	205

[a] Expressed by median (range).

3.4. Estimation of carcinogenesis rates by number of unfavorable risk factors

The prognostic model showed that the HCC development rate was significantly affected by the following four unfavorable factors: high AFP ($\geq$20 ng/ml), older age ($\geq$55 years), low platelet count ($<100{,}000/mm^3$), and male sex. Although, limitation of predictability could not be avoided because of different values of hazard ratios, we attempted to make more convenient HCC prediction curves. Five carcinogenesis curves were generated according to the number of unfavorable risk factors among the four significant covariates: no factors, one, two, three, and four unfavorable factors. When no unfavorable factor was found in a cohort of HCV-cirrhosis, the hepatocarcinogenesis rates were 9% at the end of the 5th year and 21% at the 10th year. Similarly, when one, two, three and four factors were found in a cohort, the carcinogenesis rates were 16, 28, 46, and 64% at the 5th year, and 35, 55, 78, and 93% at the 10th year, respectively (log-rank test, $P=0.0001$).

To validate the reliability of the concise prediction curves, the actual carcinogenesis curves were generated by the product-limit method for the 1991–2003 internal cohort of our hospital (Fig. 3). All actual carcinogenesis curves fitted well with the simulated curves, except for the subgroup with 'no unfavorable factors': none of 11 patients in this subgroup developed HCC during a median observation period of 10.0 years (25 percentile 8.1 years, 75 percentile 10.8 years).

4. Discussion

Ten-year-rate of HCC development has been reported as 50–80% in some cohorts of HCV-positive cirrhosis [10–13,28], and the cohorts in our hospital showed 54–55%, and Tokyo Hospital 68%. However, the reasons for the significant differences found in the rates among various hospitals have not been fully elucidated until recently. Many risk factors have been identified as important for the development of HCC in patients with hepatitis or cirrhosis [10,13,29,30], but of even greater interest is the precise prediction of HCC. In order to establish a reliable method for predicting carcinogenesis risk in a variety of patients with HCV-positive cirrhosis (compensated and decompensated), we investigated a large cohort of patients with few dropout cases, using a multivariate proportional model.

In the final step of multivariate analysis, AFP, age, platelet and gender were independently associated with HCC development in the primary cohort of our hospital. A total of 16 simulated carcinogenesis curves were drawn according to the four binary factors. Surprisingly, the estimated carcinogenesis curves significantly differed from each other among the stratified subgroups in our hospital, depending on demographic and background characteristics. In the case of a patient with HCV-cirrhosis, the combination of age, gender, AFP and platelet count could give important prognostic information about future carcinogenesis risk. When HCC appearance rates were simulated under 16 conditions according to the four binary variables identified by multivariate analysis, the 5th year rate varied from 9 to 64%, and 10th year rates from 21 to 93%. On the other hand, aminotransferase level and Child–Pugh score were poor predictors of carcinogenesis in patients with HCV-cirrhosis.

We recognized that the HCC development rate should be evaluated more specifically for each subgroup than for the entire cohort of HCV-positive cirrhosis patients. Integration of the four predictive factors could provide useful information about HCV-related carcinogenesis in actual clinical practice. The reported diversity of carcinogenesis

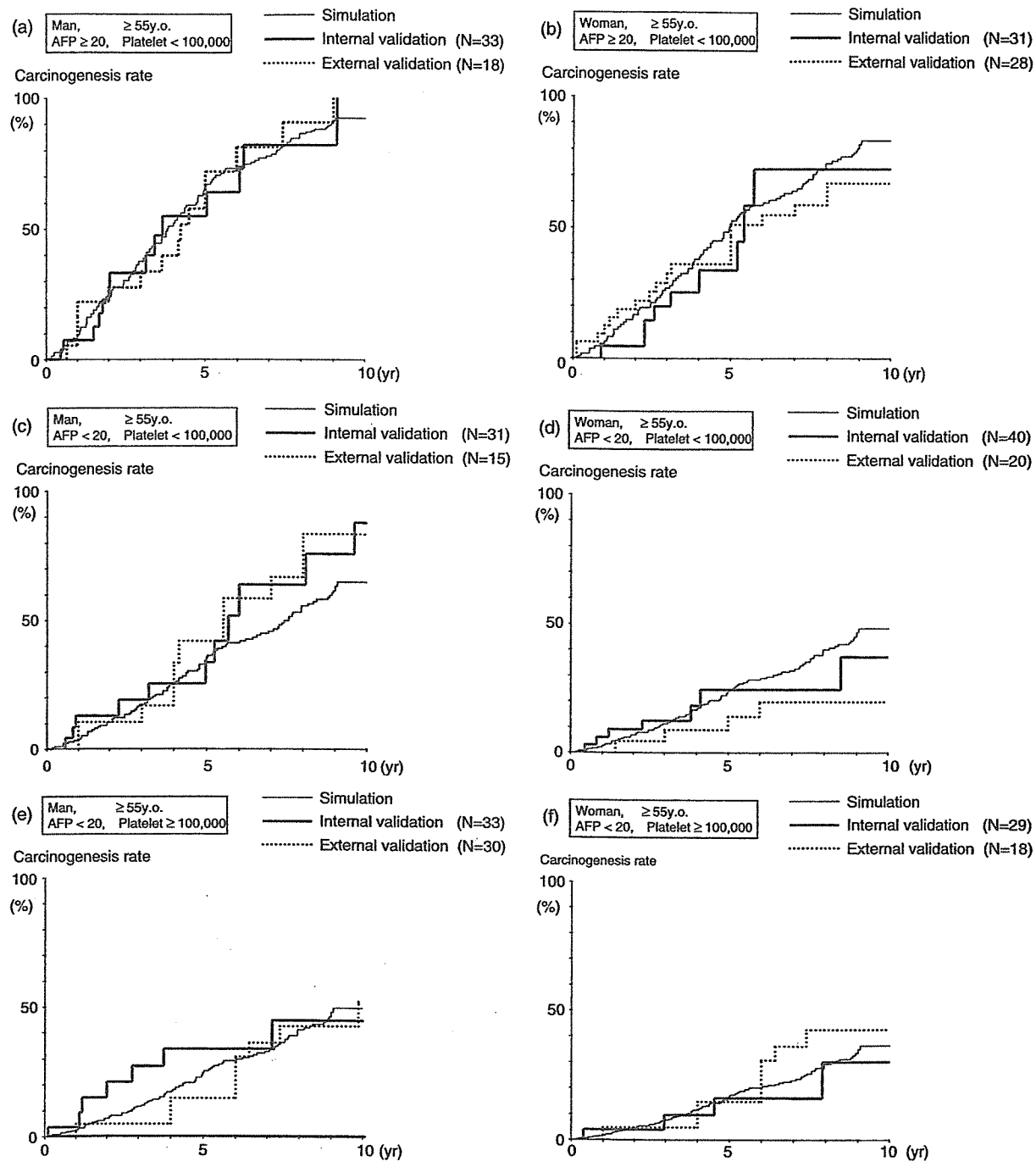

Fig. 2. Simulated carcinogenesis curves with actual carcinogenesis rates of internal and external validation cohorts, according to four significant predictors (gender, age, alpha-fetoprotein [AFP], and platelet count). *Thin solid lines*: simulated carcinogenesis curves, *bald lines*: actual curves of internal cohort (Toranomon Hospital, 1991–2003), *bald dotted lines*: actual curves of external cohort (Tokyo National Hospital, 1975–2002). (a) Carcinogenesis curves for subgroup of man, age≥55 years, AFP≥20 ng/ml, and platelet count<100,000/mm^3. (b) Subgroup of woman, age≥55 years, AFP≥20 ng/ml, and platelet count<100,000/mm^3. (c) Subgroup of man, age≥55 years, AFP<20 ng/ml, and platelet count<100,000/mm^3. (d) Subgroup of woman, age≥55 years, AFP<20 ng/ml, and platelet count<100,000/mm^3. (e) Subgroup of man, age≥55 years, AFP<20 ng/ml, and platelet count≥100,000/mm^3. (f) Subgroup of woman, age≥55 years, AFP<20 ng/ml, and platelet count≥100,000/mm^3.

rates also explains the inconsistency of estimated carcinogenesis rates from untreated cirrhosis caused by HCV. One of the reasons why carcinogenesis rates differed between the two hospitals seemed to originate from the difference of age of the patient populations. Current study did aim at precise prediction of carcinogenesis rate of each cirrhotic patient in different hospital and different period of time.

Validation of such a model is essential before these tools can gain widespread clinical use [31]. The best way to validate these models is to assess their performance in sets

Table 5
Significance of four factors associated with hepatocarcinogenesis in the internal validation group ($n=302$) and external validation group ($n=205$, 1975–2002 cohort of Tokyo National Hospital)

Factors	Internal validation cohort (1991–2003 Toranomon Hospital)		External validation cohort (1975–2002 Tokyo National Hospital)	
	Hazard ratio (95% CI)	*P*	Hazard ratio (95% CI)	*P*
Alpha-fetoprotein	1		1	
	2.13 (1.21–3.78)	0.009	2.23 (1.55–3.23)	<0.001
Age	1		1	
	3.36 (1.56–7.23)	0.002	1.55 (0.96–2.48)	0.071
Sex	1		1	
	1.78 (0.99–3.19)	0.040	2.01 (1.38–2.92)	<0.001
Platelet	1		1	
	1.49 (0.83–2.67)	0.18	1.40 (0.97–2.02)	0.070

of patients who are independent in place and time [32]. This external validity is particularly important when models are used to predict outcomes in daily practice, because it is well known that prognostic models do not perform as well in patients outside the clinical context in which they are developed [33]. This study shows that our prognostic model accurately predicts carcinogenesis rates for patients with HCV-cirrhosis from a chronologically different group and a geographically different referral center, and therefore supports the generalization and reliability of the model. The two validation cohorts (302 and 205 patients) were classified into 16 groups according to their risk factors, and the values for the actual and model-predicted survival of each risk group were compared graphically using actual Kaplan–Meier curves. The model provided a very good fit with the carcinogenesis data of each risk group in the validation cohorts (Fig. 2a–f).

We could not draw meaningful and reliable carcinogenesis curves in the remaining 10 risk groups, because of small patient numbers. The significance of current study might be the prediction of hepatocarcinogenesis in these small patient groups.

We also tried to predict carcinogenesis risk using a simplified process in the same patient group, using few unfavorable risk factors instead of individual items of the risk factors. The clinical characteristics of the 302 patients in the internal validation cohort, for whom complete information was available, are summarized in Table 4, together with the characteristics of the 183 patients used to develop the model. Since, both groups of patients were very similar in terms of their risk variables, the estimated carcinogenesis curves showed good agreement: all actual carcinogenesis curves fitted well with the simulated curves, except for a subgroup with 'no unfavorable factors'. The reason for the inconsistency was that none of the 11 patients in the subgroup developed HCC, and because the 'best' subgroup might include a significant number of patients with far better liver function tests for cirrhosis. Since, the external validation cohort included older patients with low platelet counts, the differences in the proportion of unfavorable risk factors would produce contradictory results in this kind of analysis when only using few risk factors.

For pragmatic purposes, a good prognostic model, in addition to being generalizable, needs to be based on readily accessible variables and can be calculated easily at the bedside [34]. Our model employs four variables that are readily available for every patient with cirrhosis, and includes the responses to four yes/no questions. With the help of a pocket table (Table 3), a calculator is even not needed to determine the carcinogenesis risk of a given patient and their estimated median carcinogenesis rate. Since, there is considerable diversity in carcinogenesis risk among individual patients with HCV-cirrhosis, these results will be useful for stratification of patients in future cancer prevention trials. Even though predictability of carcinogenesis risk in individual patients is limited in this kind of statistics [35], this study will be helpful to realize the diversity of carcinogenesis rate in the same 'HCV-related cirrhosis'.

In conclusion, our four-variable model is a simple and useful tool for predicting carcinogenesis rates in patients with cirrhosis caused by HCV. Prediction models for HCC

Table 6
Multivariate analysis for a combined patient group of study cohort, internal validation cohort, and external validation cohort

Factors	Category	Hazard ratio (95% confidence interval)	*P*
Alpha-fetoprotein	0: <20 (ng/ml)	1	
	1: ≥20 (ng/ml)	2.22 (1.77–2.79)	<0.001
Age	0: <55 (year)	1	
	1: ≥55 (year)	1.90 (1.44–2.51)	<0.001
Sex	0: Female	1	
	1: Male	1.90 (1.50–2.40)	<0.001
Platelet count	0: ≥100,000/mm^3	1	
	1: <100,000/mm^3	1.46 (1.16–1.84)	0.001
Patient groups	0: Study cohort	1	
	1: Internal validation cohort	0.90 (0.66–1.23)	0.52
	2: External validation cohort	1.26 (1.04–1.57)	0.023

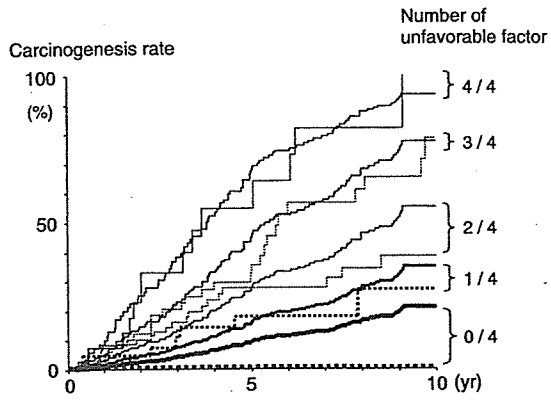

Fig. 3. Simulated HCC appearance curves with actual appearance rates of internal and external validation cohorts, according to the number of unfavorable risk factors. Five solid curves show simulated carcinogenesis rates drawn according to the number of unfavorable risk factors; none (the thickest line), one, two, three, and four (the thinnest line). Five dotted curves indicate actual HCC appearance curves of the validation cohort (Toranomon Hospital, 1991–2003).

development that combine several variables of patient data to indicate the probability of clinical outcome are powerful tools for assisting physicians in the decision-making process. Our model can be used for prediction of HCC in daily clinical practice by hepatologists, for education and information for individual patients, for selection of a candidate for a cancer prevention program, and for a proper stratification of cirrhotic patients in clinical trials for the purpose of cancer prevention. The consistency and reproducibility of the present model should also be confirmed by other institutions outside Japan.

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特集／肝機能検査値 — その上手な読み方

各種肝疾患の肝機能異常

C　型　慢　性　肝　炎

荒　瀬　康　司　　熊　田　博　光

は　じ　め　に

わが国の肝障害の主体は肝炎ウイルス性である。飲酒過剰によるアルコール性肝障害あるいは肥満や糖尿病等に起因する脂肪肝，非アルコール性脂肪性肝炎等もあるが，最も重要なのはウイルス性肝炎である。さらに，肝細胞癌が発癌した状態まで考えると，90%以上がB型かC型の肝炎ウイルスに由来する病態である。特にC型はウイルス性肝炎のなかでも70～90%を占め慢性肝炎の診療にあたっては日常臨床にて最も遭遇し易い疾患である。

C型の慢性肝疾患患者の診療においては，血液生化学的検査がきわめて重要である。その理由は血液生化学的検査により以下のような事項の把握が行いうるからである。

① 肝病変進展度の把握
② 予後の推測
③ 治療法の選択
④ 肝外病変

そこで本稿ではこの項目の順で記していく。

I．肝　病　変　進　展　度

C型慢性肝疾患の進展度の確定診断は現在でも腹腔鏡肝生検等による肝の病理学的所見による。表1にC型慢性肝疾患での腹腔鏡像と肝生検診断との関係を示した。表1で注目されることは腹腔鏡像と肝生検診断とが必ずしも一致しないことである。すなわち腹腔鏡像で結節を呈し，肝硬変と診断された場合でも約1/4の例では組織学的にはF4である肝硬変とは診断されない。従って，腹腔鏡と肝生検を組み合わせた検査が確定診断には重要と考えられる[1]。

しかしながら，このような病理学的診断は血液生化学的検査，画像検査等に比較して侵襲性の大きい検査であり同時に検査に伴う出血，ショック等の合併症がみられうる。従って血液生化学的検査の所見より慢性肝炎と肝硬変とが判別できる計算式が存在すると都合がよい。当院では腹腔鏡肝生検にて病理学的に確定診断されたC型慢性肝疾患205例につき，患者の性，年齢，その他血液生化学的検査値等計20項目を用いた判別関数を算出し多変量解析にて判別式作成を試みた。その結果慢性肝炎・肝硬変の判別には，γ-グロブリン，ヒアルロン酸，性別および血小板数の4因子が重要であり，これらの因子より表2に示した判別式を作成した。この判別式を計算し，値が－ならば慢性肝炎，＋ならば肝硬変と判定すると正診率は

表　1　　C型慢性肝疾患での腹腔鏡像と肝生検診断との関係

腹腔鏡像＼肝生検診断	Stage 1	Stage 2	Stage 3	Stage 4	Total
Smooth	161 (79.3%)	37 (18.2%)	5 (2.5%)	0	203
Irregular	31 (16.9%)	84 (45.9%)	48 (26.2%)	20 (10.9%)	183
Nodular	0	4 (7.1%)	11 (19.6%)	41 (73.2%)	56
Total	192	125	64	61	442

虎の門病院消化器科

表 2　慢性肝炎と肝硬変の判別式（A）

A=0.124×γ-グロブリン（%）＋0.001×ヒアルロン酸（μg/dl）
＋（−0.413）×性別（男=1，女=2）＋（−0.075）
×血小板数（万/μl）−2.005

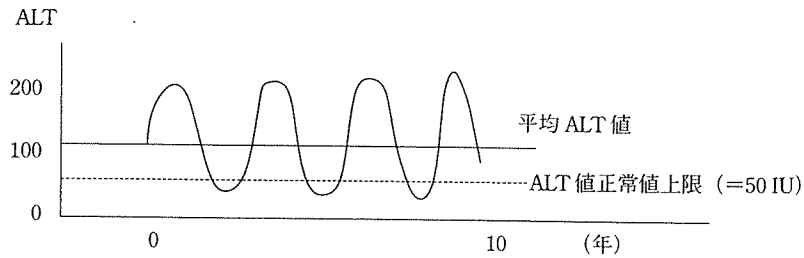
判定　Aが（−）→ 慢性肝炎
　　Aが（＋）→ 肝硬変　} と判定

正診率　91.2%

表 3　ALT 積算値による C 型慢性肝炎進行度の予測

ALT積算値＝平均ALT異常率×観察期間（年）

$$\text{平均ALT異常率}=\frac{\text{平均ALT値}}{\text{ALT値正常値上限（=50 IU）}}$$



$$\text{ALT 積算値}=\frac{\text{平均 ALT 値}}{\text{ALT 値正常値上限（=50 IU）}}\times\text{観察期間（年）}$$

$$=(100/50)\times 10=\underline{20}$$

図 1　ALT 積算値による C 型慢性肝炎進行度の予測

91.2%であった[2]。以上慢性肝炎・肝硬変の判別には性差を除けばγ-グロブリン，ヒアルロン酸，血小板の3因子が重要であった為，この3因子につき以下に述べる。

1. γ(ガンマ)-グロブリン

γ-グロブリンは本来形質細胞が産生するもので，血清中のγ-グロブリン量は，正常では1～1.5 g/dl 程度であり，血清蛋白中の相対的割合は10～20%である。ウイルス性肝炎・自己免疫性肝炎ではしばしば血中濃度が増加し，1.5 g/dl 以上となり，血清蛋白中の分画でも20%をしばしば越える。肝硬変となるとウイルス性の有無にかかわらず，アルコール性などの原因によるものでも高値となる。肝疾患の程度が高度となると一般にγ-グロブリン値は増加するが，個人差が大きい。

2. 血　小　板

慢性肝炎から肝硬変に進行すると徐々に血小板数の減少がみられる。血小板減少の機序は門脈圧亢進に基づく脾腫（脾機能亢進症）による血小板破壊の亢進，骨髄での血小板産生の低下等による。ウイルス性の肝障害では，肝病変の進行に並行して血小板減少が進むため，重症度判定の簡単な指標となる。一般に，慢性肝炎が軽度～中等度であれば20万/mm³で血小板数は正常だが，慢性肝炎高度となると15万/mm³程度に低下する。初期肝硬変では12万/mm³，完成された肝硬変では10万/mm³程度に低下する。

3. ヒアルロン酸

ヒアルロン酸は生体内結合組織に広く分布する酸性ムコ多糖であり，線維芽細胞などの間葉系細胞で生成され，その多くは肝類洞内皮細胞で異化される。肝病変の進展につれ肝類洞内皮細胞が障害され，ヒアルロン酸は増加してくる。基準値は50ng/ml 以下である。200ng/ml 以上に増加すれば肝硬変の可能性が高くなる。

Ⅱ. 予　後　の　推　測

C 型慢性肝疾患においては，肝炎の活動状態が長く続けば肝硬変へ進展しやすく，また肝発癌も増加するとの報告が多い。そこでトランスアミナーゼ異常の程度とその継続期間の尺度として表3に記した ALT 積算値を用いて C 型慢性肝炎進行度を検討予測した。

図2に具体的に2症例を示した。2例とも初回肝生検組織像は F1 であった。症例1は経過中 ALT の平均値が250 IU/L で経過し，初回肝生検より11年後には肝発癌がみられ，非癌部も肝硬変

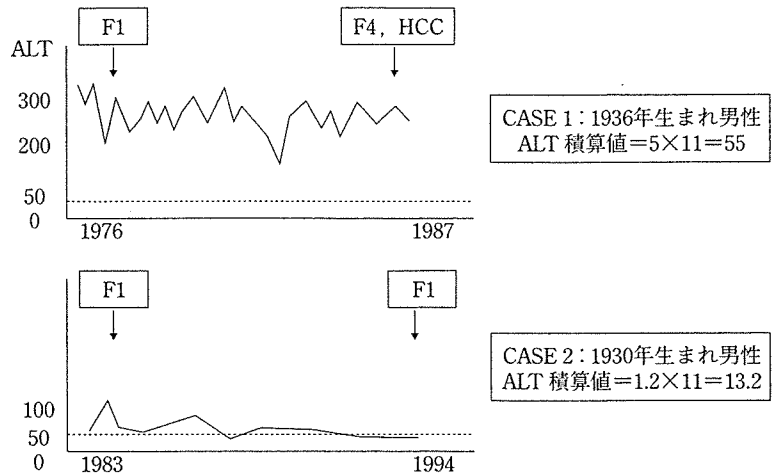


図　2　　ALT 積算値からみた組織学的進展の相違例

表　4　C 型慢性肝炎での初回肝生検像と ALT 積算値からみた繰り返し肝生検例での肝硬変移行率

初回肝生検像 ＼ ALT 積算値	＜10	10≦＜20	20≦＜30	30≦
F1	0/18 (0%)	0/45 (0%)	5/26 (19.2%)	10/21 (47.6%)
F2	3/32 (9.4%)	16/45 (35.6%)	18/27 (66.6%)	11/12 (91.6%)
F3	7/16 (43.8%)	25/29 (86.2%)	4/5 (80%)	2/2 (100%)

表　5　初回肝生検像と ALT 積算値からみた C 型慢性肝炎での肝癌発生率

初回肝生検像 ＼ ALT 積算値	＜10	10≦＜20	20≦＜30	30≦
F1	0/9 (0%)	1/23 (4.3%)	4/26 (15.4%)	14/43 (32.6%)
F2	1/6 (16.7%)	8/17 (47.1%)	9/20 (45%)	17/37 (45.9%)
F3	1/3 (33.3%)	7/18 (38.9%)	7/11 (63.3%)	5/9 (55.6%)

であった。この症例の ALT 積算値は55であった。次に症例２は症例１と同様に初回肝生検診断は F1 であったが，その後 ALT の平均値は60 IU/L で経過し，11年後には ALT 積算値は13.2であった。このことから ALT 積算値が大きい場合には慢性肝炎より肝硬変への移行がおこり，場合によっては肝発癌をきたすと考えられた。

次に図３には初回肝生検診断が F2 であり，肝硬変・肝癌へと進展した例と，F1 への改善がみられた症例とを示した。肝硬変・肝癌へと進展した例では ALT 積算値は22であったのに比し，F1 へ改善した例では ALT 積算値は４と低値であった。

そこで次に C 型慢性肝炎での初回肝生検像と ALT 積算値とからみた繰り返し肝生検例での肝硬変移行率について表４に示した。初回肝生検像が F1 では，ALT 積算値が20以下では肝硬変移行例はみられなかった。一方初回肝生検像が F3 では ALT 積算値が10を越すと80%以上の例が肝硬変まで進展してみられた。

さらに初回肝生検像と ALT 積算値からみた C 型慢性肝炎での肝癌発生率について表５に示した。初回肝生検像が F1 では ALT 積算値が30を越した場合に肝癌発生率は30%を超えたが，F3 では ALT 積算値が10未満でも肝癌発生率は30%を超えていた。

ALT が正常上限の1.5倍で経過した場合に13年余で ALT 積算値は20となり，20年で ALT 積算値30となる。従って初回肝生検像が F1 ならば ALT が平均正常上限の1.5倍以下で経過すればしばらくの間は肝硬変移行，肝癌発生は起こりにくいと考えられる。しかしながら初回肝生検像が F3 にまで至った場合には ALT 積算値が10でも肝硬変移行，肝癌発生率が高いため，F3 症例は ALT を正常内に押さえ込むことが重要となる。

Ⅲ．治　療　法　の　選　択

C 型慢性肝炎に対する治療法として大きく２つに分けられる。その１つはウイルスを排除する為の根治的な療法であり，ウイルス排除の為に抗ウイルス剤を投与する方法である。別の１つは肝細胞をウイルスによる破壊から守ろうとする治療法

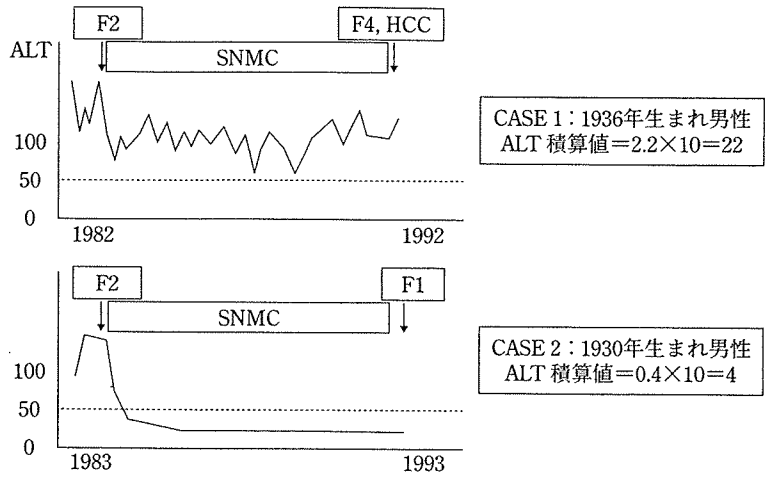


図　3　　ALT 積算値からみた組織学的進展の相違例

表 6　C型慢性肝炎にてインターフェロン治療が禁忌となる場合

- IFN 単独投与での禁忌
- 1.　精神病あるいはその既往
- 2.　血小板，白血球減少
- 3.　網膜症例
- 4.　その他－心疾患例，ネフローゼ例，自己免疫性疾患例，コントロール不良糖尿病例，高齢者

- IFN＋リバビリン併用例での禁忌
- 1.　IFN 単独例で禁忌の場合
- 2.　腎不全例
- 3.　貧血高度例
- 4.　妊娠，分娩予定者およびその配偶者
- 5.　その他－コントロール不良高血圧例

表 7　虎の門病院におけるC型慢性肝炎に対する IFN 単独療法（6ヵ月以内）の genotype 別およびウイルス量別著効率　　(IFN 総治療数 2,897)

genotype / HCV-RNA	1b	2a	2b
100kcopy/ml 以上	54/1,374 (3.9%)	152/375 (40.5%)	80/198 (40.4%)
100kcopy/ml 未満	261/493 (52.9%)	227/310 (73.2%)	77/147 (52.3%)

である。前者の代表がインターフェロン（以下IFN）あるいはIFN＋リバビリンの併用療法である。この治療法は根治的ではあるがその副作用が多くかつ強いため表6に示した病態の際には禁忌となる。後者の肝細胞をウイルスによる破壊から守ろうとする治療法が肝庇護療法であり，この治療法としてグリルチン製剤を含んだ強力ネオミノファーゲンCあるいはウルソ等の投与が行われる。

1.　C型慢性肝炎に対する6ヵ月までのIFN療法

C型肝炎の根治的治療の中心はIFN療法である。IFN療法はウイルスの増殖を抑制することにより，ウイルスを排除する方法である。IFNは現在，天然型IFNのα，β，遺伝子組み替え型のα-IFNが主体である。その投与法は我が国では，2週間連日投与後，週3回の間歇投与で計6ヵ月間投与が標準的で，これによりC型肝炎ウイルスの約30%が治癒する。しかしその治療効果はC型肝炎ウイルスのタイプと量により異なる。現在までのところ本邦では約30万人程度のC型慢性肝炎患者に6ヵ月までのIFN治療がなされてきたと考えられる。表7に当院で6ヵ月までのIFN治療を行った際の治療効果を示す。表5は全国の約1%の患者に相当する成績と考えられるが，genotype1bで高ウイルス量の群ではそのウイルス排除率は数%以下とIFN単独療法での治療効果は不良である。従ってgenotype1bの高ウイルス量の例はIFN単独の6ヵ月までの治療では難治例と考えられるが，本邦においてはこのようなIFN難治例がC型慢性肝炎患者の約50%を占めている。

2.　genotype1/高ウイルス量のC型慢性肝炎に対する抗ウイルス療法

Genotype1で高ウイルス量のC型慢性肝炎はIFN単独療法でのウイルス排除率は不良である。このためIFNにポリエチレングリコール(PEG)を結合させた新しいPEG-IFNが使用されるよう