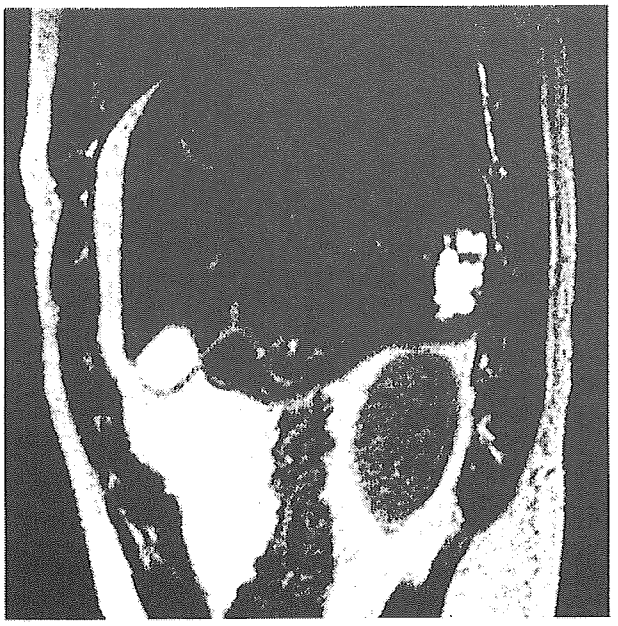

Figure 1. The cystic lesion has high signal intensity on the T2-weighted magnetic resonance image. No other lesion was observed in the liver.

Immunohistochemistry

Immunohistochemical staining was performed on formalin-fixed, paraffin-embedded sections using the avidin-biotin complex method [9]. Sections were cut at 3 μm, deparaffinized in xylene, and the endogenous peroxidase activity was blocked with 0.3% H_2O_2 in methanol. The slides were treated with 0.05% pepsin for 60 min and microwaved [10] at 95°C for 20 min to recover antigenicity, then subjected to immunohistochemical staining for carcinoembryonic antigen (CEA) (1:1000; TAKARA BIO INC, Shiga, Japan), p53 (1:1000; NICHIREI, Tokyo, Japan), and Ki-67 antigen (MIB-1: 1:100; Dako, Glostrup, Denmark).

Results

Macroscopically, the lesion measured 3.6 cm in diameter and was composed of a number of small grayish-white cysts, the diameters of which ranged from 0.1 to 1.2 cm (Figure 2). Green and mucinous fluid was present in the cysts. Neither mural nodules on the cysts nor irregular septum was seen. Microscopically, the lesions were discrete, round in shape, and periportal in location. The constituent cysts were embedded in a fibrous stroma, and some of these cysts had dilated lumens (Figure 3A,B). The cysts were lined by a low columnar or cuboidal epithelium and contained bile-stained material (Figure 3C,D). Immunohistochemistry showed negative results for p53 and CEA monoclonal antibody. Fewer than 1% of cells showed proliferation activity, assessed by staining for Ki-67 antigen (clone MIB-1).

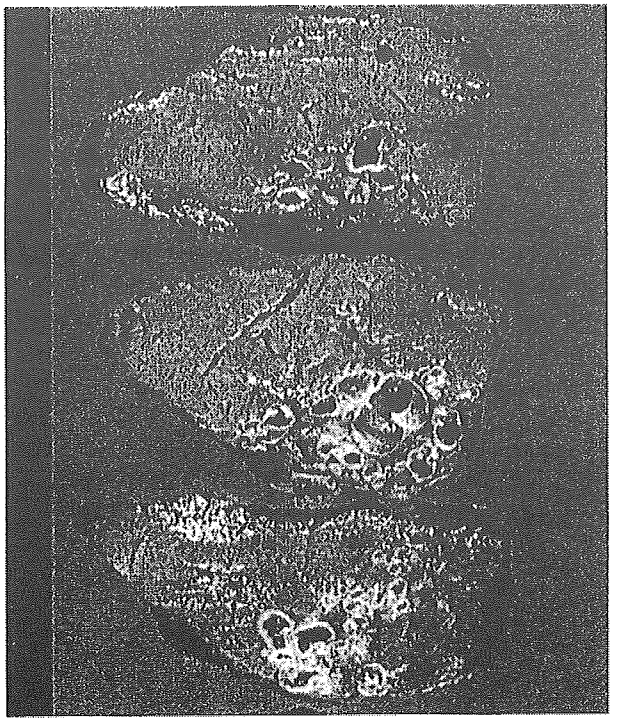

Figure 2. Cut surface of a resected specimen showed that the lesion is composed of a number of small, grayish-white cysts, 3 cm in size, with a green, viscous liquid content. No solid component was evident within the cystic lesion.

Discussion

Bile duct hamartomas consist of focal, disordered collections of bile ducts, lined with a single layer of low columnar or cuboidal epithelium, surrounded by abundant fibrous stroma [1–7]. The bile ducts within the lesions are round to irregular in shape, and contain pink amorphous material or actual bile.

In the present case, the lesion contained both small cysts (<0.5 cm) and larger cysts (0.5–1.2 cm), which were lined by a low columnar or cuboidal epithelium, contained bile-stained material, and were embedded in a fibrous stroma. Microscopic examination and immunohistochemistry did not show any potential for malignancy in the epithelium of the cysts. These findings lead us to conjecture that the lesion was a BDH. Two findings, however, differed from those of the typical BDH: the lesion was solitary and measured 3.6 cm in diameter, whereas typical BDH lesions are multiple and less than 0.5 cm in diameter. Thus, we believe this lesion is a previously unrecognized biliary lesion and propose to designate it as a solitary biliary hamartoma. Solitary BDH is characterized by a solitary nodule composed of small bile ducts of irregular shape, set in a fibrous stroma, and is distinct from typical BDH because of its large size, relatively

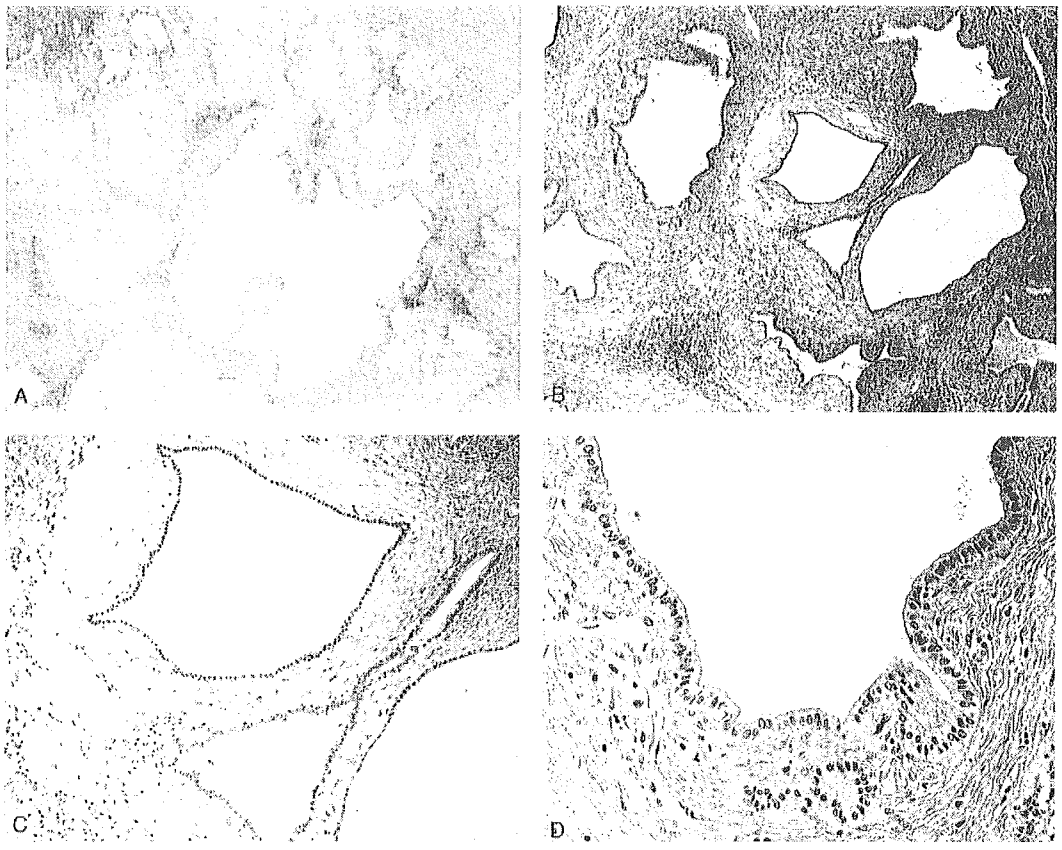

Figure 3. Distinctive features of solitary bile duct hamartoma. A. Microcysts of various size surrounded by fibrous stroma (hematoxylin & eosin; original magnification ×5). B. Microcysts are similar to bile ducts but show abnormal arrangement. Size of cysts tends to be larger than that of typical bile duct hamartoma (hematoxylin & eosin; original magnification ×40). C. & D. Epithelium lining of columnar cells and cuboidal cells shows no cytological abnormalities (hematoxylin & eosin; original magnification C: ×100, D: ×200).

larger bile ducts constituting the lesion, and absence of synchronous biliary hamartomas in the surrounding liver.

In our case, preoperative MRI, which is essential diagnostic imaging for detecting small cystic lesions [11–13], showed no cystic lesions other than the BDH in the liver. In the recent literature, no pathological study of solitary BDH confirmed by preoperative MRI has been reported.

The pathogenesis of BDH is obscure. Bile duct hamartoma is found frequently in patients with adult polycystic kidney disease, Caroli's disease, or congenital hepatic fibrosis [5,8,14–16]. The association with these congenital bile duct diseases suggests that BDH might be a genetic disease. V. J. Desmet hypothesized that BDH is a congenital disease of the intrahepatic bile ducts that results from abnormal remodeling of the embryonic ductal plate, and that BDH is associated with ductal plate malformation of the peripheral interlobular bile ducts in the late phase of bile duct embryogenesis [17]. However, it has been suggested that at least some BDHs are an acquired disease resulting from hepatic ischemia by arteriosclerosis or polyarteritis [18], or from alcoholic liver injury [19].

The pathogenesis of the present case is also unknown. However, ductal plate malformation might not be involved, because the patient had no other congenital bile duct disease, and furthermore the solitary lesion is not consistent with the concept of systemic malformation of the biliary tree. The lesion contained large cysts, and this feature resembles that of BDH resulting from hepatic ischemia reported by Popovsky et al. [18]. The morphological findings of the present case favor pathogenesis other than ductal plate malformation, such as hepatic ischemia, although no history of arteriosclerosis or polyarteritis was identified in the patient.

Bile duct hamartoma is a non-neoplastic lesion. Several papers reported cases of cholangiocarcinoma co-existing with BDHs [20,21]; however, association between cholangiocarcinoma and BDH is still a controversial idea. To date, no treatment is required when solitary BDH is correctly diagnosed. However, solitary BDH should be distinguished from other solitary hepatic lesions. Biliary cystadenoma is a primary cystic neoplasm in liver, and its radiological features resemble those of solitary BDH [22]. Biliary cystadenoma is defined as a multilocular lesion lined by a benign or atypical columnar cuboidal epithelium

usually with an "ovarian-like" stroma, and is rarely encountered in males. Many areas within biliary cystadenoma are composed of a combination of columnar to cuboidal mucinous epithelial cells, and the epithelial cells are positive to cytokeratin, epithelial membrane antigen, and CEA [13,22]. In contrast, neither atypical cells nor mucin-secreting cells were present in the epithelium of solitary BDH, and epithelial cells of solitary BDH were negative with antibody to CEA. The stroma of solitary BDH is relatively rich, but was different from "ovarian-like" stroma. Differentiation between biliary cystadenocarcinoma and solitary BDH is easier. Cystadenocarcinoma is a predominantly cystic but usually solid component macroscopically. Cystic lesions are lined by a proliferation of cytologically malignant epithelium in cystadenocarcinoma. Biliary adenofibroma is a rare benign biliary tumor characterized by microcystic and tubular formations embedded in a fibrous stroma [23,24]. Microscopic findings resemble those of solitary BDH, but its more solid appearance with honeycomb-cut surface is distinct from that of solitary BDH. Furthermore, the epithelium of biliary adenofibroma is positive to keratin AE.3/Cam5.2, CEA, and epithelial membrane antigen and has the marked nuclear p53 immunoreactivity and a low Ki-67 proliferative index ($\leq$10%). In contrast, the epithelium of solitary BDH was negative to CEA and p53, and showed a lower Ki-67 proliferative index ($<$1%).

Our patient underwent surgical resection because biliary cystadenoma could not be excluded in the preoperative diagnosis. Solitary BDH should be added to differential diagnoses of intrahepatic cystic lesions, however, the radiological diagnosis of solitary BDH might be difficult despite careful preoperative study.

In summary, on rare occasions solitary BDH may occur in the liver. Solitary BDH has to be considered as a differential diagnosis of intrahepatic cystic neoplasms.

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HEPATOLOGY

Adverse hepatic events caused by radiotherapy for advanced hepatocellular carcinoma

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Abstract

Background: Radiotherapy is often used to treat patients with unresectable advanced hepatocellular carcinoma (HCC). The present study examines the nature and frequency of adverse events with respect to liver function in such patients after radiotherapy.

Methods: Forty-six patients with HCC who underwent radiotherapy were retrospectively examined. Radiotherapy was applied using coplanar 2–3-beam arrangements to a target dose of 50 Gy/5 weeks. The adverse hepatic events were evaluated according to the National Cancer Institute *Common Toxicity Criteria* and the Radiation Therapy Oncology Group/European Organization for Research and Treatment of Cancer Late Radiation Morbidity Scoring Scheme during the acute phase and the late phase by following the patients for up to 1 year. The influence on survival by adverse hepatic events and other factors was analyzed.

Results: The full irradiation dose of 50 Gy was given to 40 patients (87.0%). Grade 3 or 4 toxicity was observed in 18 (39.1%) within 3 months after radiotherapy and in 11 (33.3%) of 33 thereafter, respectively. The most frequent and serious adverse events were hyperbilirubinemia, hypoalbuminemia, and ascites. The independent adverse prognostic factors for survival were portal vein tumor thrombus ($P = 0.0012$), tumor response ($P = 0.011$), acute adverse hepatic event ($P = 0.012$), and late adverse hepatic event ($P = 0.015$).

Conclusions: Hypoalbuminemia, hyperbilirubinemia, and ascites were important hepatic adverse events that developed after applying radiotherapy to treat advanced HCC. These adverse events seriously affected survival.

Key words: adverse event, external-beam radiotherapy, hepatocellular carcinoma.

INTRODUCTION

Unresectable advanced hepatocellular carcinoma (HCC) has been treated using radiotherapy with and without transcatheter arterial chemoembolization (TACE), and several institutions have reported positive results.[1–7] In contrast, irradiation also injures normal liver tissue and serious, radiation-induced liver disease can develop.[2,4] Most patients with HCC originally had liver dysfunction such as cirrhosis or chronic hepatitis due to hepatitis virus B or C. Therefore, hepatic adverse events that develop after radiotherapy for HCC must be defined.

Adverse events caused by irradiation have been evaluated according to the Radiation Therapy Oncology Group (RTOG) Acute Radiation Morbidity Scoring Criteria or RTOG/European Organization for Research and Treatment of Cancer (EORTC) Late Radiation Morbidity Scoring Scheme[8] and/or the late effects normal tissues (LENT) scoring system and the late effects toxicity scoring system (the SOMA scale).[9,10] However, the RTOG acute radiation morbidity scoring criteria do not mention liver dysfunction at the acute phase of irradiation. However, the National Cancer Institute *Common Toxicity Criteria* (NCI-CTC) can evaluate adverse events after cancer treatment.[11] Therefore, we

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studied the adverse hepatic events using the NCI-CTC in patients with unresectable advanced HCC who underwent radiotherapy, and assessed their impact on survival.

METHODS

We retrospectively studied 46 patients with HCC who underwent radiotherapy for HCC between September 1992 and April 2000 at the National Cancer Center Hospital East. Patient characteristics are shown in Table 1. Hepatocellular carcinoma was clinically diagnosed using ultrasound, dynamic computed tomography (CT), angiography and serum α-fetoprotein concentrations. The main tumors were located in the right lobe in 27 patients, the left lobe in 18 and both lobes in one patient. The main tumor determined by the late phase of dynamic CT was 2.5–15.0 cm in diameter (mean ± SD, 7.3 ± 3.2 cm). Among 37 patients with tumor thrombus of a portal vein, the thrombus was located within the second branch of the portal vein in seven patients, within the first right or left portal vein in 17, and to the portal trunk in 13.

Table 1 Patient characteristics

Variable	No.patients ($n = 46$)
Gender	
Male	38
Female	8
Median age (years) (range)	61 (42–82)
Performance status†	
0	26
1	11
2	9
Child–Pugh classification (Pugh score)	
Grade A (5 or 6)	14
Grade B (7, 8, or 9)	27
Grade C (10–15)	5
Viral markers	
HBs Ag(+), HCV Ab(–)	7
HBs Ag(–), HCV Ab(+)	36
HBs Ag(–), HCV Ab(–)	3
Portal vein tumor thrombus	
(–)	9
(+)	7
(++)	30
Previous treatment	
None	24
TACE	11
Hepatectomy + TACE	5
PEI	3
PEI + TACE	3

PEI, percutaneous ethanol injection; TACE, transcatheter arterial chemoembolization.

†Eastern Cooperative Oncology Group score: 0, fully active; 1, restricted in physically strenuous activity; 2, ambulatory and fully capable of self-care; 3, capable of only limited self-care; and 4, completely disabled. Portal vein tumor thrombus: (–), none; (+), within second branches; (++), beyond second branches.

The target volume of radiotherapy was defined in order to exclude at least one-half of the non-cancerous liver volume outside of the irradiated volume. Accordingly, 34 of the 46 patients received radiotherapy only for their portal vein tumor thrombus and the other 12 patients received radiotherapy for the entire primary HCC and portal vein tumor thrombus. External beam radiotherapy (EBRT) to a target dose of 50 Gy/25 fractions per 5 weeks was planned using 6–21 MV (median 10 MV) of X-rays. The EBRT planning was performed using a CT-based 2-D planning system (CT Port, Toshiba, Tokyo, Japan). Gross tumor volume (GTV) was defined according to the aforementioned restriction regarding irradiated volume of non-cancerous liver, which was based on radiographic findings. Clinical target volume (CTV) encompassed gross tumor volume defined as a radiographically abnormal area with an additional 1–1.5 cm margin. Planning target volume (PTV), consisting of CTV with 1- and 2-cm margins in the cranial and caudal directions, respectively, was defined taking into account both the respiratory motion of the liver and daily set-up error. A 2- or 3-beam arrangement was used in order to minimize not only the irradiated volume of non-cancerous liver, but also the inhomogeneity of dose distribution within the PTV. Radiotherapy dose prescription was defined at the center of the GTV. The irradiation field ranged from 5.4 cm × 6.0 cm to 15.2 cm × 15.7 cm.

Transcatheter arterial chemoembolization was performed in 35 patients with HCC lesions outside the field of irradiation. A mixture of iodized oil (Lipiodol; Andre Guerbet, Aulnay-sous-Bois, France) and epirubicin followed by gelatin sponge particles was injected from the right or left hepatic artery. The TACE was repeated every 2–3 months. It was performed once in 14 patients, twice for 12, three times for three patients, four times for three patients, and 5–10 times for five patients, respectively.

We evaluated hepatic function during the acute phase starting from irradiation until 90 days after, according to the NCI-CTC, and the late phase (from 90 days to 1 year later) according to the RTOG/EORTC Late Radiation Morbidity Scoring Scheme. Adverse events affecting liver function were evaluated by measuring serum bilirubin, serum albumin, serum glutamic oxaloacetic transaminase (SGOT); serum glutamic pyruvic transaminase (SGPT), alkaline phosphatase (ALP), γ-glutamyl transpeptidase (γ-GTP), prothrombin time, ascites, portal vein flow and hepatic enlargement. We evaluated an abnormality that worsened one or more grades in each parameter in patients with liver dysfunction before irradiation as an adverse event. Patients underwent dynamic CT to evaluate the response at 2–3-month intervals after therapy. Computed tomography was performed to obtain contiguous transverse sections using the helical scanning method at a section thickness of 5 mm. Tumor response was assessed according to the WHO criteria.[12]

We statistically analyzed the results using the Kruskal–Wallis Exact test to compare hepatic adverse events among grades determined by the Child–Pugh

classification, between patients with and without TACE, and among grades of portal vein tumor thrombus. Survival was calculated using the Kaplan–Meier method from the start of irradiation. The statistical significance of differences between survival curves was determined according to the log–rank test. The Cox proportional hazards model was used for multivariate analysis of prognostic factors. Differences with $P < 0.05$ were considered significant.

RESULTS

The full 50-Gy irradiation dose was feasible in 40 of the 46 patients (87.0%). Irradiation was stopped because of general deterioration in two patients at 46 and 44 Gy, massive ascites in two at 42 and 36 Gy, bleeding from esophageal varices in one at 36 Gy, and hepatic encephalopathy in one at 18 Gy. Histological examination of the ascites confirmed the absence of malignant cells.

Adverse hepatic events

We identified 28 (60.9%) adverse hepatic events among 46 patients during the acute phase. The most common adverse events were hyperbilirubinemia, hypoalbuminemia, and ascites. Grade 3 or 4 toxicity developed in 18 (39.1%) with hyperbilirubinemia, hypoalbuminemia, ascites, or elevation of SGOT, SGPT, ALP, and/or γ-GTP. SGOT and/or SGPT values were temporarily elevated to grade 3 in three patients and one patient started to bleed from esophageal varices, which elevated the SGOT grade to 4. The ALT and γ-GTP values also transiently increased to grade 3 in one patient.

We evaluated adverse events during the late phase in 33 patients. Eight patients died within 3 months of starting irradiation, and five patients who died within 4 months were excluded because they were terminal during the last month and adverse effects of irradiation could not be accurately evaluated. Adverse events developed in 19 (57.6%) of the 33 patients, the most frequent being ascites, hyperbilirubinemia and hypoalbuminemia. Grade 3 or 4 toxicity appeared in 11 (33.3%) with hyperbilirubinemia, ascites, elevation of SGOT, SGPT, ALP, and/or γ-GTP. Serum glutamic oxaloacetic transaminase, SGPT, ALT, and/or γ-GTP increased to grade 3 in 4 patients, but this was also accompanied by hyperbilirubinemia or ascites.

Relationship between adverse hepatic events and tumor response

Fourteen of the 46 patients achieved a partial response, but no complete responses were observed. The overall response rate was 30.4% (95% confidence interval [CI], 17.7–45.8%). Table 2 shows the relationship between acute adverse hepatic events and tumor response. The frequency of adverse events that developed during the acute phase tended to be higher in patients without tumor response than with tumor response ($P = 0.087$).

Table 2 Relationship between local response and acute hepatic adverse event

		Grade of acute hepatic adverse event Adverse hepatic event		
	n	0–1	≥2	
PR	14	10	4	$P = 0.087$
NC	22	9	13	
PD	10	3	7	

NC, no change; PD, progressive disease; PR, partial response.

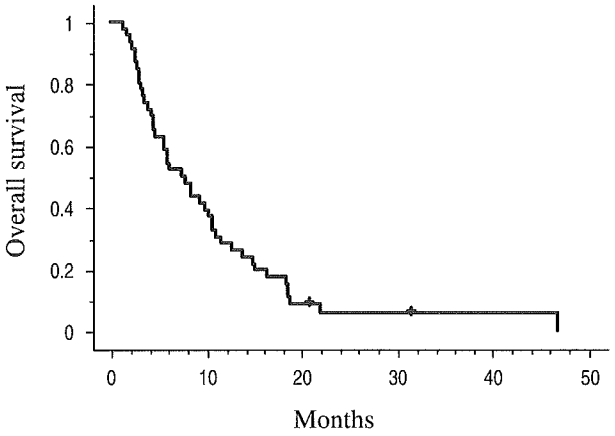

Figure 1 Overall survival rates of all patients ($n = 46$). The median survival time was 7.5 months, and the 1- and 2-year survival rates were 28.3% and 5.8%, respectively.

Relationship between adverse hepatic events and other factors

To determine the influence on adverse hepatic events by other factors, we examined the relationship between the adverse events and these factors: liver function before irradiation, using the Child–Pugh classification (Pugh score) and combined treatment of TACE, and the presence of portal vein tumor thrombi. The frequency of adverse events that developed during the acute phase tended to be higher in patients with more serious liver dysfunction (score 7, 8, or 9 and 10–15) than with mild liver dysfunction (score 5 or 6; $P = 0.070$). There were statistically no differences between patients treated with and without TACE during any period ($P = 0.61$). Regarding portal vein tumor thrombi, adverse events developed more often in patients with than without portal vein tumor thrombi; in particular, the difference during the acute phase was statistically significant ($P = 0.012$).

Survival

The overall survival curve is shown in Figure 1. Of all the 46 patients, 45 have now died except for one patient

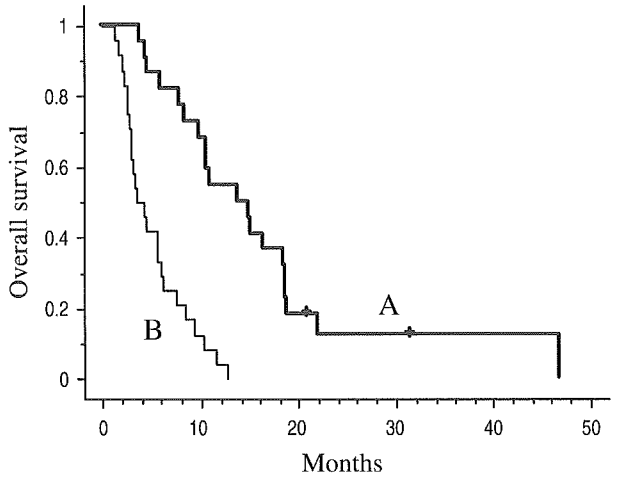

Figure 2 Survival curves according to grades of adverse hepatic event during the acute phase. Curve A, survival of patients with grade 0 or 1 ($n = 24$), median survival time is 13.8 months; curve B, patients with grade ≥2 ($n = 22$), median survival time is 3.5 months. One-year survival rate was 54.5% in grade 0 or 1, 4.2% in grade ≥2. $P < 0.0001$.

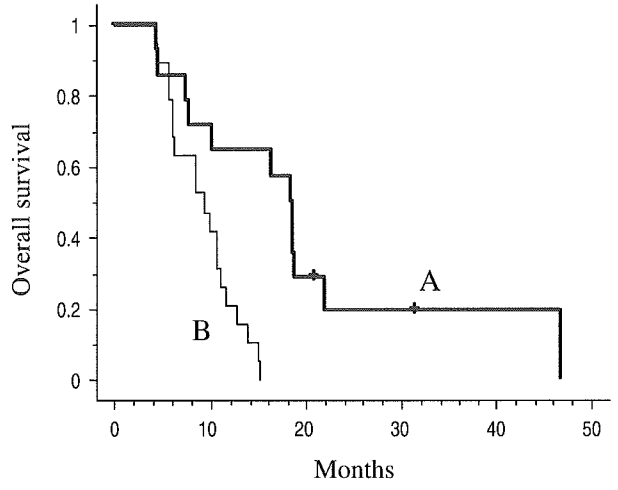

Figure 3 Survival curves according to grades of adverse hepatic event during the late phase. Curve A, survival of patients with grade 0 or 1 ($n = 14$), median survival time 18.5 momths; curve B, patients with grade ≥2 ($n = 19$), median survival time 9.3 months. One-year survival rate was 64.3% in grade 0 or 1, 15.8% in grade ≥2, respectively. $P < 0.0007$.

with whom we have lost contact. The causes of death were tumor progression in 31, liver failure in seven, variceal bleeding in two, pneumonitis due to irradiation in one, and other diseases in four (heart disease in two, renal dysfunction in one, and lung edema in one). Pneumonitis developed 2 months after irradiation. The median survival time (MST) was 7.5 months, and the 1- and 2-year survival rates were 28.3% and 5.8%, respectively. We divided patients with adverse hepatic events during the acute and late phase into the following groups: A, grade 0 or 1; and B, grades 2 or more. We compared survival between the two groups (Figs 2,3), and the survival rates of the two groups statistically differed ($P < 0.0001$, 0.0007).

Prognostic factors related to survival

Performance status, the Child–Pugh classification, the presence of portal vein tumor thrombi, tumor response, and adverse hepatic events during the acute phase and late phase had a prognostic significance for survival (Table 3). Significant prognostic factors identified by univariate analysis were entered into multivariate analysis (Table 4). The independent adverse prognostic factors for survival in patients with HCC treated with radiotherapy were portal vein tumor thrombus ($P = 0.0012$), tumor response ($P = 0.011$), acute adverse hepatic event ($P = 0.012$), and late adverse hepatic event ($P = 0.015$).

DISCUSSION

Although radiotherapy has long been applied to patients with unresectable HCC, it has never been recognized as totally satisfactory. However, local radiation with a limited-field high dose of 48–72.6 Gy is effective for treating HCC.[1–5] In contrast, to achieve positive treatment effects against HCC using radiotherapy, attention should be paid to liver tolerance, because irradiation also injures the normal liver and serious radiation-induced liver disease can develop.[2] Cheng *et al.* reported that radiation-induced liver disease and gastrointestinal bleeding are the most frequent treatment-related toxicities and that six of 25 patients developed radiation-induced liver disease evidenced by elevated ALP and transaminases and non-malignant ascites.[5] Guo *et al.* reported that two patients died of liver failure or variceal bleeding associated with the therapy, and that serum bilirubin, serum transaminase and ascites increased by 13%, 27% and11%, respectively.[6] Although radiation-induced liver dysfunction should be assessed in detail, to our knowledge there are few efforts that have been directed towards this issue.

The present study examined adverse hepatic events caused by radiotherapy in two periods: acute phase within 90 days and the late phase from 90 days to 1 year later. Hypoalbuminemia, hyperbilirubinemia, and ascites were the most frequently seen in grade 3 or 4 of adverse events during both periods. Elevation of SGOT, GPT, ALP, and γ-GTP were almost always accompanied by hyperbilirubinemia, hypoalbuminemia, and/or ascites and increased temporarily. Thus, hyperbilirubinemia, hypoalbuminemia and ascites seem to be important factors when evaluating liver dysfunction with respect to radiotherapy as a treatment for HCC.

Variceal rupture and hepatic encephalopathy were important complications of HCC and/or liver cirrhosis. Variceal bleeding developed during and after irradiation in two of the present patients. Hepatic encephalopathy developed during irradiation in one patient. Each of these patients developed serious liver dysfunction and died early. We therefore believe that the development of

Table 3 Univariate analysis of prognostic factors for survival in patients with advanced hepatocellular carcinoma treated by radiotherapy

Variable	*n*	Median survival (months)	Survival rate (%) 1 year	2 year	*P*
Gender					
Male	38	7.5	26.3	3.9	0.59
Female	8	7.3	37.5	12.5	
Age (years)					
<60	20	5.7	40.0	0	0.98
≥60	26	7.9	19.2	7.7	
Performance status					
0	26	9.8	38.5	10.3	0.016
1, 2	20	5.8	15.0	0	
Child–Pugh classification					
Grade A	14	14.3	57.1	7.1	0.049
Grade B, C	32	5.9	15.6	6.3	
HCV Ab					
Positive	36	7.9	27.8	7.4	0.29
Negative	10	5.0	30.0	0	
Portal vein tumor thrombus					
(–) or (+)	16	12.1	50.0	18.8	0.002
(++)	30	4.6	16.7	0	
TACE					
With	11	11.1	36.4	9.1	0.53
Without	35	6.0	25.7	4.3	
Tumor response					
PR	14	13.8	64.3	7.1	0.007
NC + PD	32	4.5	12.5	6.3	
Adverse hepatic event during acute phase					
Grade 0 or 1	24	13.8	54.5	12.1	<0.0001
Grade ≥2	22	3.5	4.2	0	
Adverse hepatic events during late phase					
Grade 0 or 1	14	18.5	64.3	12.7	0.0007
Grade ≥2	19	9.3	15.8	0	

NC, no change; PD, progressive disease; PR, partial response; TACE, transcatheter arterial chemoembolization.
Portal vein tumor thrombus: (–), none; (+), within second branches; (++), beyond second branches.

Table 4 Independent prognostic factors for survival in patients with advanced HCC treated by radiotherapy

Variable	Category	Relative risk (95%CI)	*P*
Performance status	0	1	0.69
	1 or 2	1.20 (0.50–2.98)	
Child–Pugh classification	A	1	0.95
	B or C	1.03 (0.43–2.50)	
Portal vein tumor thrombus	(–) or (+)	1	0.0012
	(++)	4.91 (1.88–12.84)	
Tumor response	PR	1	0.011
	NC + PD	3.68 (1.35–10.06)	
Adverse hepatic event during acute phase	Grade 0 or 1	1	0.012
	Grade ≥2	3.59 (1.32–9.79)	
Adverse hepatic event during late phase	Grade 0 or 1	1	0.015
	Grade ≥2	3.96 (1.30–12.01)	

CI, confidence interval; HCC, hepatocellular carcinoma; NC, no change; PD, progressive disease; PR, partial response; TACE, transcatheter arterial chemoembolization.
Multivariate analysis using the Cox proportional hazards model.
Portal vein tumor thrombus: (–), none; (+), within second branches; (++), beyond second branches.

the esophageal varices and hepatic encephalopathy, including during the irradiation therapy, should be carefully considered.

We examined the relationship between adverse hepatic events and survival. We confirmed that survival rates worsened according to the grade of adverse events (grades 0 or 1, and 2 or more) and differences between the two groups in each period were statistically significant. On the other hand, tumor characteristics such as α-fetoprotein level, tumor size, number of tumors, and portal involvement have been reported as prognostic factors for HCC.[13–15] Therefore, we examined the independent adverse prognostic factors for survival by multivariate analysis with the Cox proportional hazards model. As for the results, the independent adverse prognostic factors for survival in patients with HCC treated by radiotherapy were portal vein tumor thrombus ($P = 0.0012$), tumor response ($P = 0.011$), acute adverse hepatic events ($P = 0.012$), and late adverse hepatic events ($P = 0.015$). The present results indicated that adverse hepatic event can be a significant prognostic factor in addition to other conventional factors such as the presence of a portal vein tumor thrombus. Particularly, once patients develop grade 2 or more adverse events, the prognosis is potentially very poor, so they require careful follow up.

The present study found that grade 3 or 4 adverse hepatic events developed at a very high rate compared with other reports.[4–6] Thirty-seven (80.4%) of the present patients had tumor thrombi of the portal vein, which may explain why the frequency of adverse hepatic events was so high. We examined the relationship between the grade of the tumor thrombus and adverse hepatic events. Adverse events developed more often in patients with, than without portal vein tumor thrombi, particularly during the acute phase. Acute adverse effects of radiotherapy are generally reversible. However, those adverse effects, particularly hepatic ones, can often be irreversible in patients with advanced HCC. From this point of the view, we considered that hepatic adverse events during the acute phase affected the survival. When it is to be applied to patients who have advanced HCC such as those with portal vein tumor thrombi, radiotherapy should be carefully considered and patients should be closely observed.

The present study has some limitations that should be recognized with respect to the evaluation of the hepatic adverse effects. The present study was a retrospective analysis, and it included two groups of patients who had the different irradiation targets: mainly the portal vein tumor thrombus and the entire HCC lesion including the tumor thrombus. It was difficult to evaluate exactly the hepatic adverse effects, if the volume of the normal liver tissue irradiated was calculated. Recently, 3-D planning has been applied to determine the CTV in patients with HCC.[16,17] In the radiotherapy using this 3-D planning, the dose–volume histogram can be used for calculation of the volume of the normal liver tissue irradiated in radiotherapy for HCC. Because EBRT planning was performed using a 2-D planning system in the present study and the patients were heterogeneous in the radiation targets, we could not examine the volume of the normal liver tissue irradiated. The results of the present study must be understood under these limitations, and a prospective analysis of the hepatic adverse effects should be performed using factors that were important in this study under consideration of the volume of the normal liver tissue irradiated.

In conclusion, we confirm that hyperbilirubinemia, hypoalbuminemia and ascites are important factors that reflect survival. Furthermore, bleeding of an esophageal varix and hepatic encephalopathy also negatively affect prognosis. The evaluation of adverse hepatic events seems to be valuable in predicting prognosis.

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ORIGINAL REPORT

Phase II Study of Radiotherapy Employing Proton Beam for Hepatocellular Carcinoma

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ABSTRACT

Purpose
To evaluate the safety and efficacy of proton beam radiotherapy (PRT) for hepatocellular carcinoma.

Patients and Methods
Eligibility criteria for this study were: solitary hepatocellular carcinoma (HCC); no indication for surgery or local ablation therapy; no ascites; age ≥ 20 years; Zubrod performance status of 0 to 2; no serious comorbidities other than liver cirrhosis; written informed consent. PRT was administered in doses of 76 cobalt gray equivalent in 20 fractions for 5 weeks. No patients received transarterial chemoembolization or local ablation in combination with PRT.

Results
Thirty patients were enrolled between May 1999 and February 2003. There were 20 male and 10 female patients, with a median age of 70 years. Maximum tumor diameter ranged from 25 to 82 mm (median, 45 mm). All patients had liver cirrhosis, the degree of which was Child-Pugh class A in 20, and class B in 10 patients. Acute reactions of PRT were well tolerated, and PRT was completed as planned in all patients. Four patients died of hepatic insufficiency without tumor recurrence at 6 to 9 months. Three of these four patients had pretreatment indocyanine green retention rate at 15 minutes of more than 50%. After a median follow-up period of 31 months (16 to 54 months), only one patient experienced recurrence of the primary tumor, and 2-year actuarial local progression-free rate was 96% (95% CI, 88% to 100%). Actuarial overall survival rate at 2 years was 66% (48% to 84%).

Conclusion
PRT showed excellent control of the primary tumor, with minimal acute toxicity. Further study is warranted to scrutinize adequate patient selection in order to maximize survival benefit of this promising modality.

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INTRODUCTION

Cirrhosis is found in more than 80% of patients with hepatocellular carcinoma (HCC). This precludes more than 70% of the patients from receiving potentially curative treatments, and also contributes eventually to fatal hepatic insufficiency and multifocal tumorigenesis.[1,2] Approximately 50% to 70% and 30% to 50% of 5-year overall survival was achieved with surgery including liver transplantation[3-6] and percutaneous local ablation,[7-9] respectively, for an adequately selected population of patients. However, no standard strategy has been established for patients with unresectable HCC at present.

Partial liver irradiation for HCC using 50 to 70 Gy of megavoltage x-ray with or without transarterial chemoembolizaztion (TACE) for 5 to 7 weeks has been widely applied during the last two decades. This resulted in response rates of 33% to 67%, with a median survival period of 13 to 19

months and 10% to 25% overall survival at 3 years.[10-12] Since 1985, proton radiotherapy (PRT) administered at a median dose of 72 cobalt gray equivalent (Gy_E) in16 fractions during 3 weeks with or without TACE, had been applied in more than 160 patients with HCC at the University of Tsukuba, resulting in a more than 80% local progression-free survival rate with 45% and 25% overall survival at 3 and 5 years, respectively.[13,14] The excellent depth-dose profile of the proton beam enabled us to embark on an aggressive dose escalation while keeping a certain volume of the noncancerous portion of the liver free from receiving any dose of irradiation. This single-institutional, single-arm, prospective study was conducted to confirm encouraging retrospective results of PRT for HCC using our newly installed proton therapy equipment.

PATIENTS AND METHODS

Patient Population

Patients were required to have uni- or bidimensionally measurable solitary HCC of $\leq$ 10 cm in maximum diameter on computed tomography (CT) and/or magnetic resonance (MRI) imaging. In addition, the following eligibility criteria were required: no history of radiotherapy for the abdominal area; no previous treatment for HCC within 4 weeks of inclusion; no evidence of extrahepatic spread of HCC; age $\geq$ 20 years; Zubrod performance status (PS) of 0 to 2; WBC count $\geq$ 2,000/mm^3; hemoglobin level $\geq$ 7.5 g/dL; platelet count $\geq$ 25,000/mm^3; and adequate hepatic function (total bilirubin $\leq$ 3.0 mg/dL; AST and ALT $<$ 5.0$\times$ upper limit of normal; no ascites). Patients who had multicentric HCCs were not considered as candidates for this study, except for those with the following two conditions: (1) multinodular aggregating HCC that could be encompassed by single clinical target volume; (2) lesions other than targeted tumor that were judged as controlled with prior surgery and/or local ablation therapy. Because a planned total dose would result in a significant likelihood of serious bowel complications, patients who had tumors abutting or invading the stomach or intestinal loop were excluded. The protocol was approved by our institutional ethics committee, and written informed consent was obtained from all patients.

Pretreatment Evaluation

All patients underwent indocyanine green clearance test, and the retention rate at 15 minutes (ICG R15) was measured for the purpose of quantitative assessment of hepatic functional reserve. CBC, biochemical profile including total protein, albumin, total cholesterol, electrolytes, kidney and liver function tests, and serological testing for hepatitis B surface antigen and antihepatitis C antibody were done. C-reactive protein and tumor markers including alpha feto-protein and carcinoembryonic antigen were also measured. Chest x-ray was required to exclude lung metastasis. All patients were judged as unresectable by expert hepatobiliary surgeons in our institution, based on their serum bilirubin level, ICG R15, and expected volume of resected liver.[15] Gastrointestinal endoscopy was done to exclude active ulcer and/or inflammatory disease located at the stomach and the duodenum. All patients underwent abdominal ultrasonography, triphasic CT or MRI, CT during arteriography and arterial portography.[16] Diagnosis of HCC was based on radiographic findings on triphasic CT/MRI. Radiologic criteria for HCC definition were as follows: tumor showing high attenuation during hepatic arterial and portal venous phase indicating hypervascular tumor; tumor showing low attenuation during delayed phase indicating rapid wash-out of contrast media. Confirmatory percutaneous fine-needle biopsies were required for all patients unless they had radiologically compatible, postsurgical recurrent HCC. Tumors that broadly abut on the vena cava, portal vein, or hepatic vein that were associated with caliber changes and/or filling defects of these vessels, were tentatively defined as positive for macroscopic vascular invasion. One patient had visible tumor on fluoroscopy because of residual iodized oil contrast medium used in previous TACE. For the other 29 patients, one or two metallic markers (inactive Au grain of which the diameter and length were 1.1 mm and 3.0 mm, respectively) were inserted percutaneously at the periphery of the target tumor.

Treatment Planning

PRT was performed with the Proton Therapy System (Sumitomo Heavy Industries Ltd, Tokyo, Japan), and treatment planning, with the PT-PLAN/NDOSE System (Sumitomo Heavy Industries Ltd). In this system, the proton beam was generated with Cyclotron C235 with an energy of 235 MV at the exit. Gross tumor volume (GTV) was defined using a treatment planning CT scan using X Vision Real CT scanner (Toshiba Co Ltd, Tokyo, Japan), and clinical target volume (CTV) and planning target volume (PTV) were defined as follows: CTV = GTV + 5 mm, and PTV = CTV + 3 mm of lateral, craniocaudal, and anteroposterior margins. Proton beam was delivered with two-beam arrangement to minimize irradiated volume of noncancerous liver using our rotating gantry system. The beam energy and spread-out Bragg peak[13] were fine-tuned so that 90% isodose volume of prescribed dose encompassed PTV. To evaluate the risk of radiation-inducing hepatic insufficiency, dose-volume histogram (DVH) was calculated for all patients.[17]

Scanning of CT images for both treatment planning and irradiation of proton beam were done during the exhalation phase using a Respiration-Gated Irradiation System (ReGIS). Our ReGIS during this study period was composed in the following manner: strain gauge, which converts tension of the abdominal wall into electrical respiratory signal, was put on the abdominal skin of the patient; gating signal triggering CT scanning or proton beam was generated during the exhalation phase.

Treatment

The fractionation and dosage in this study were based on the results of a retrospective study at the University of Tsukuba. A total dose ranging from 50 Gy_E in 10 fractions to 87.5 Gy_E in 30 fractions (median, 72 Gy_E in16 fractions) was administered without serious acute and late adverse events. All patients received PRT to a total dose of 76 Gy_E for 5 weeks in 3.8-Gy_E once-daily fractions, four fractions in a week using 150 to 190 MV proton beam. Relative biologic effectiveness of our proton beam was defined as 1.1. No concomitant treatment (eg, TACE, local ablation, systemic chemotherapy) was allowed during and after the PRT, unless a treatment failure was detected. Verification of patient set-up was done in each fraction using a digital radiography subtraction system. In this system, fluoroscopic images obtained at daily set-up were subtracted by the original image that was taken at the time of treatment planning. Position of the patient couch was adjusted to overlap the diaphragm, inserted metallic markers, and bone landmarks on the original position at the end of the exhalation phase.

PRT was administered 4 days a week, mainly Monday to Thursday, and Friday was reserved for maintenance of the PRT system. Predefined adverse reaction of PRT was dermatitis, pneumonitis, hepatic insufficiency, and gastrointestinal ulcer and/or bleeding. If one of these reactions of grade 3 or higher, or unexpected reactions of grade 4 or higher were observed in three patients, further accrual of patients was defined to be stopped. No further PRT was allowed when grade 4 hematologic toxicity or any of the toxicities of grade 3 or higher were observed at the digestive tract or lung. PRT was delayed up to 2 weeks until recovery when an acute nonhematologic toxicity of grade 3 or higher, other than that described above, was observed. However, when only an elevation of liver enzymes was observed without manifestation of clinically significant signs and symptoms, PRT was allowed to be continued according to the physician's judgment.

Outcomes

It has been reported that the tumor, although achieving a complete response, persisted over a long period, ranging from 3 weeks to 12+ months after the completion of PRT.[18] Therefore, a local progression-free survival rate at 4 weeks after the end of PRT was adopted as the primary end point of this study, where an event was defined as progression of the primary tumor with size increase of more than 25%, in order to facilitate an interim analysis as described in the Statistical Design section below. Assessment of primary tumor response using CT and/or MRI was performed 4 weeks after the completion of PRT. Overall survival and disease-free survival rates were also evaluated as secondary end points. Death of any cause was defined as an event in calculation of overall survival, whereas tumor recurrences at any sites or patient deaths were defined as events for disease-free survival. Adverse events were reviewed weekly during the PRT by means of physical examination, CBC, liver function test, and the other biochemical profiles as indicated. The severity of adverse events was assessed using the National Cancer Institute Common Toxicity Criteria (NCI-CTC) version 2.0. After completion of PRT, reviews monitoring disease status, including CT and/or MRI examinations and long-term toxicity were done at a minimum frequency of once every 3 months.

Statistical Design

The null hypothesis of a true local progression-free rate of 50% or lower was based on average results of photon radiotherapy reported from Japan, in which each study accumulated approximately 20 patients.[11,12] This was tested against the alternative hypothesis of a true rate of 80% or higher with an α level of 5% and a power of 80%, which required 30 patients according to the method by Makuch and Simon.[19] If fewer than five patients experienced local progression-free status within 4 weeks postirradiation at the end of first nine enrollments, the trial would be stopped. Otherwise, if more than 24 patients remained locally progression-free among the total of 30 patients, this would be sufficient to reject the null hypothesis and conclude that PRT warrants further study. Time-to-event analyses were done using Kaplan-Meier estimates, and 95% CIs were calculated. The difference of time-to-event curve was evaluated with the log-rank test. Multivariate analyses were performed with Cox's proportional hazards model.

RESULTS

Patients

Thirty patients were enrolled between May 1999 and February 2003. Patient characteristics at the start of PRT are

Table 1. Characteristics of 30 Enrolled Patients

Characteristic	Patients	
	No.	%
Age, years		
Median	70	
Range	48-87	
Sex		
Male	20	67
Female	10	33
ECOG performance status		
0-1	29	97
2	1	3
Clinical stage (2)		
I	9	30
II	19	63
III	2	7
Positive viral markers		
Hepatitis B virus	3	10
Hepatitis C virus	26	87
Both	1	3
Child-Pugh classification		
A	20	67
B	10	33
C	0	0
Pretreatment indocyanine green clearance at 15 minutes, %		
< 15	0	0
15-40	21	70
40-50	5	17
> 50	4	13
Tumor size, mm		
Median	45	
Range	25-82	
20-50	19*	63
> 50	11	37
Macroscopic vascular invasion		
Yes	12	40
No	18	60
Morphology of primary tumor		
Single nodular	26	87
Multinodular, aggregating	1	3
Diffuse	2	7
Portal vein tumor thrombosis	1*	3
Serum alpha-fetoprotein level, ng/mL		
< 300	21	70
≥ 300	9	30
Histology		
Well-differentiated	10	33
Moderately differentiated	14†	47
Poorly differentiated	2	7
Differentiation not specified	3	10
Negative (radiologic diagnosis only)	1	3
Prior treatment		
No	13	43
Recurrence	6	20
Local ablation/TACE	11	37

Abbreviations: ECOG, Eastern Cooperative Oncology Group; TACE, transarterial chemoembolization.
*Includes one patient whose gross target volume was tumor thrombosis at the posterior branch of right portal vein as a result of postsurgical recurrence.
†Includes two patients with histological diagnoses that were defined in previous surgery.

listed in Table 1. All patients had underlying liver cirrhosis with an initial ICG R15 value of ≥ 15%. Thirteen patients received PRT as a first treatment for their HCC. Six patients had postsurgical recurrences, and 11 received unsuccessful local ablation and/or TACE to the targeted tumor before PRT. Histologic confirmation was not obtained in one patient who had tumor with typical radiographic features compatible with HCC. Vascular invasion was diagnosed as positive in 12 patients. Three patients had HCC of ≤ 3 cm in diameter; however, they were not considered as candidates for local ablation therapy because of tumor locations that were in close proximity to the great vessels or the lung.

Adverse Events

All patients completed the treatment plan and received 76 Gy_E in 20 fractions of PRT with a median duration of 35 days (range, 30 to 64 days). Prolongation of overall treatment time of more than 1 week occurred in four patients: three were due to availability of the proton beam, and one because of fever associated with grade 3 elevation of total bilirubin that spontaneously resolved within 1 week. Adverse events within 90 days from commencement of PRT are listed in Table 2. Decrease of blood cell count was observed most frequently. A total of 10 patients experienced transient grade 3 leukopenia and/or thrombocytopenia without infection or bleeding necessitating treatment. Of note, eight of them already had leuko- and/or thrombocytopenia, which could be ascribable to portal hypertension, before commencement of PRT corresponding to grade 2 in terms of the NCI-CTC criteria. Because none of the five patients experiencing grade 3 elevation of transaminases showed clinical manifestation of hepatic insufficiency and maintained good performance status, PRT was not discontinued. Nevertheless, these events spontaneously resolved within 1 to 2 weeks.

Development of hepatic insufficiency within 6 months after completion of PRT was defined as proton-inducing hepatic insufficiency (PHI), and this was observed in eight patients. Causal relationship between PHI and several factors are described separately below. One patient developed transient skin erosion at 4 months that spontaneously resolved within 2 months. Another patient developed painful subcutaneous fibrosis at 6 months that required nonsteroidal analgesics for approximately 12 months thereafter. Both of these skin changes developed at the area receiving ≥ 90% of the prescribed dose because the targeted tumors were located at the surface of the liver adjacent to the skin. However, they remained free from refractory ulcer, bleeding, or rib fracture.

There were no observations made of gastrointestinal or pulmonary toxicity of grade 2 or greater in all patients. In addition, after percutaneous insertion of metallic markers, no serious adverse events, including bleeding or tumor seeding along the needle tracts, were observed.

Table 2. Adverse Events Within 90 Days From the Start of Proton Beam Radiotherapy

Grade	0	1	2	3	4
Leukopenia	7	2	13	8	0
Thrombocytopenia	2	6	15	7	0
Total bilirubin	20	2	7	1	0
Transaminases	4	8	13	5	0
Nausea/anorexia	23	7	0	0	0
Overall (maximum grade)	0	4	14	12	0

Tumor Control and Survival

At the time of analysis on November 2003, 12 patients had already died because of intrahepatic recurrence of HCC in seven, distant metastasis in two, and hepatic insufficiency without recurrence in three. Eleven of these 12 patients had been free from local progression until death; the durations ranged from 6 to 41 months (median, 8 months). One patient who had a single nodular tumor of 4.2 cm in diameter experienced local recurrence at 5 months and subsequently died of multifocal intrahepatic HCC recurrence. Otherwise, 18 patients were alive at 16 to 54 months (median, 31 months) without local progression. A total of 24 patients achieved complete disappearance of the primary tumor at 5 to 20 months (median, 8 months) post-PRT. Five had residual tumor mass on CT and MRI images for 3 to 35 months (median, 12 months) until the time of death (n = 4) or until last follow-up at 16 months (n = 1). As a whole, 29 of 30 enrolled patients were free from local progression until death or last follow-up, and the local progression-free rate at 2 years was 96% (95% CI, 88% to 100%). Tumor regression was associated with gradual atrophy of the surrounding noncancerous portion of the liver that initially suffered from radiation hepatitis,[20] as shown in Figure 1.

A total of 18 patients developed intrahepatic tumor recurrences that were outside of the PTV at 3 to 35 months (median, 18 months) post-PRT. Five of these occurred within the same segment of the primary tumor. Eight patients received TACE, and four received radiofrequency ablation for recurrent tumors; however, six did not receive any further treatment because of poor general condition in three and refusal in three. Five died without intrahepatic recurrence. Seven patients remained recurrence-free at 16 to 39 months (median, 35 months). Actuarial overall survival rates were 77% (95% CI, 61% to 92%), 66% (95% CI, 48% to 84%), and 62% (95% CI, 44% to 80%), and disease-free survival rates were 60% (95% CI, 42% to 78%), 38% (95% CI, 20% to 56%), and 16% (95% CI, 1% to 31%) at 1, 2, and 3 years, respectively (Fig 2).

Correlation of Survival With Prognostic Factors

Overall survival was evaluated according to 10 factors as listed in Table 3. Univariate analyses revealed that factors

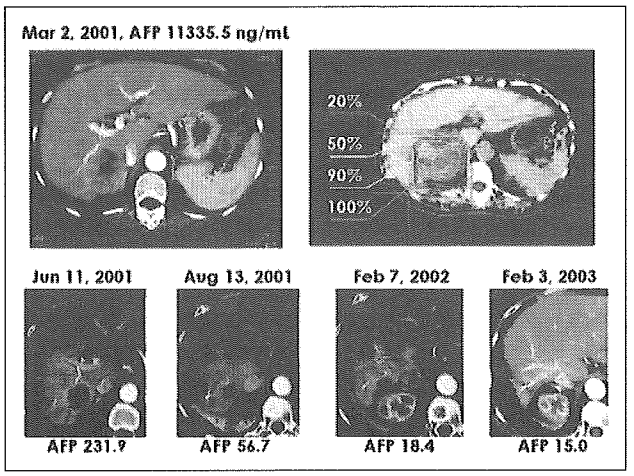

Fig 1. Case presentation: 70-year-old woman who received proton radiotherapy of 76 Gy in 20 fractions for 37 days from April 2, 2001, for her tumor located at the right posterior segment of the liver (left upper panel). Dose distribution was demonstrated in the right upper panel. Two portals from posterior and right lateral directions were used.

related to functional reserve of the liver and tumor size had significant influences on overall survival ($P < .05$). Liver function was the only independent and significant prognostic factor by multivariate analysis, as presented in Table 3. When clinical stage or Child-Pugh classification was substituted for ICG R15 as a covariate for liver function, the results of multivariate analyses were unchanged (data not shown). Overall survival according to pretreatment ICG R15 is shown in Figure 3.

Estimation of the Risk of Proton-Inducing Hepatic Insufficiency by Dose-Volume Histogram Analysis

Eight patients developed PHI and presented with ascites and/or asterixis at 1 to 4 months after completion of PRT, without elevation of serum bilirubin and transaminases in the range of more than 3× the upper limit of normal. Of these, four died without evidence of intrahepatic tumor recurrence at 6 to 9 months; three died with recurrences of HCC at 4, 8, and 22 months; and one was alive at 41 months without tumor recurrence. DVH for hepatic noncancerous portions (entire liver volume minus gross tumor volume) was drawn according to pretreatment ICG R15 values (Fig 4A to C). The results showed that all of the nine patients with ICG R15 less than 20% were free from PHI and alive at 14 to 54 months. Three of the four patients with pretreatment ICG R15 $\geq$ 50% experienced fatal PHI without evidence of HCC recurrence, and another patient died of PHI with intrahepatic and systemic dissemination of HCC at 4 months. Among patients whose ICG R15 values ranged from 20% to 50%, all of the four patients whose percentage of hepatic noncancerous portions receiving $\geq$ 30 Gy_E (V_{30}%) exceeded 25% developed PHI. On the other hand, none of the patients whose V_{30}% was less than 25% experienced PHI, as shown in Figure 4B ($P = .044$, Mann-Whitney U test). Three-year overall survival for patients with either the V_{30}% $\geq$ 25% or ICG R15 $\geq$ 50% (n = 9) was 22% (95% CI, 0% to 50%), whereas it was 79% (95% CI, 60% to 98%) for the remaining 21 patients with favorable risk ($P = .001$).

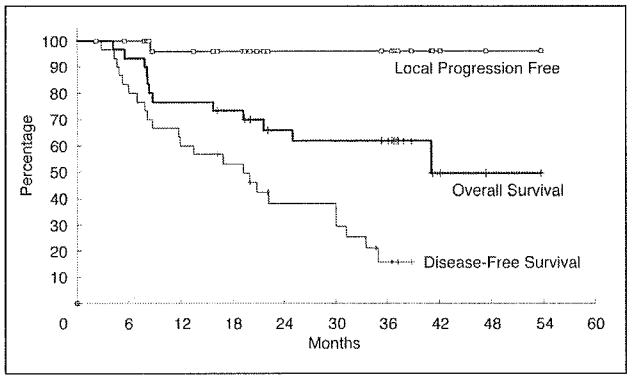

Fig 2. Kaplan-Meier estimate of local progression-free, overall, and disease-free survival rates for all 30 patients enrolled.

DISCUSSION

The principal advantage of PRT lies in its possibility of aggressive dose escalation without prolongation of treatment duration in order to improve local control rate. The liver will be the most appropriate organ for this approach because it has a unique characteristic of developing compensatory hypertrophy when a part of this organ suffers from permanent damage. This study showed that the local control rate of PRT alone for patients with advanced HCC was consistent, as previously reported.[14] Slow regression of tumor volumes associated with gradual atrophy of surrounding noncancerous liver tissue was also in agreement with a previous report.[20] No serious gastrointestinal toxicity occurred, with careful patient selection performed in order to exclude these structures from PTV receiving high PRT dose. Eligibility criteria as to blood cell count in this study were eased up considerably in order to test the safety of PRT for patients with cirrhosis associated with portal hypertension. Nevertheless, no patients experienced serious sequelae relating to leukopenia or thrombocytopenia, which were the most frequently observed adverse events during PRT. All patients were able to complete their PRT basically in an outpatient clinic. Therefore we submit that the safety, accuracy, and efficacy of PRT administering 76 Gy_E/5 weeks using our newly installed Proton Therapy System and ReGIS for selected patients with advanced HCC has been confirmed.

Multivariate analysis suggested that the the functional reserve of the liver had significant influence on overall survival. Recent prospective series of untreated patients with

Table 3. Factors Related to Overall Survival

Factor	No. of Patients	Overall Survival at 2 Years (%)	Univariate *P*	Multivariate *P*	Hazard Ratio	95% CI
Age, years			.263	.665	1.54	0.22 to 10.75
< 70	15	59				
≥ 70	15	71				
Sex			.829	.732	1.44	0.18 to 11.65
Male	20	67				
Female	10	60				
Tumor size, mm			.045	.159	0.34	0.08 to 1.52
20 to 50	19	71				
> 50	11	44				
Pretreatment ICG R15			.006	.026	0.19	0.05 to 0.82
≤ 40%	21	80				
> 40%	9	30				
Clinical stage			< .001			
I	9	73				
II	19	68				
III	2	0				
Child-Pugh classification			.006			
A	20	78				
B	10	38				
Vascular invasion			.930	.650	1.44	0.30 to 7.03
Yes	12	67				
No	18	66				
Serum AFP level, ng/mL			.313	.061	0.20	0.04 to 1.07
< 300	21	67				
≥ 300	9	60				
V_{30}%			.213	.141	0.25	0.04 to 1.58
≤ 25%	24	65				
> 25%	6	40				
Prior treatment			.455	.091	3.63	0.82 to 16.18
No	13	69				
Recurrence	17	60				

Abbreviations: ICG R15, percentage of indocyanine green clearance at 15 minutes; AFP, alpha-fetoprotein; V_{30}%, percentage of hepatic noncancerous portion receiving ≥ 30 cobalt gray equivalent.

advanced HCC and underlying cirrhosis showed that overall survival rate at 3 years ranged from 13% to 38%, and rarely exceeded 50% even for those with most favorable prognostic factors.[1] In this study, actuarial overall survival rate at 3 years for all 30 patients including those who had HCC with vascular invasion and/or severe cirrhosis was 62%. Furthermore, 21 patients with initial ICG R15 of ≤ 50% and V_{30}% of ≤ 25% achieved 79% of overall survival rate at 3 years. All of the eight patients with favorable liver functional reserve (ICG R15, 15% to 20%) were alive at 20 to 54 months as shown in Figure 3. This suggests that adequate local control with PRT provides survival benefit for selected patients with HCC and moderate cirrhosis. On the other hand, prognoses of aggressive PRT were disappointing for patients, with poor functional liver reserve showing an ICG R15 of 50% or worse, and, therefore, indication of PRT for such patients was thought to be extremely limited.

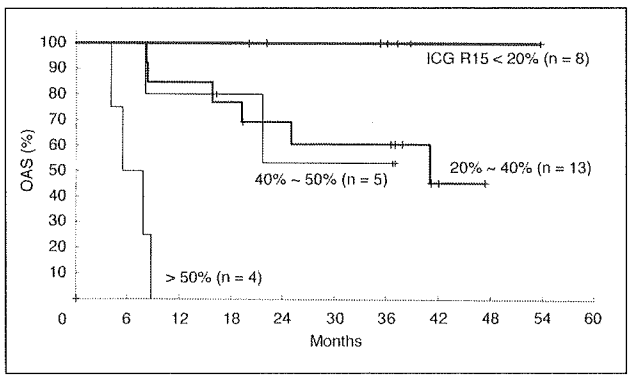

Fig 3. Overall survival (OAS) rates according to pretreatment indocyanine green clearance at 15 minutes (ICG R15).

A part of noncancerous liver suffering from PRT-inducing hepatitis gradually developed dense fibrosis and resulted in almost complete atrophy,[20] whereas the absorbed dose in a large proportion of the remaining liver was 0 Gy_E, as shown in Figures 1 and 4. This change is similar to

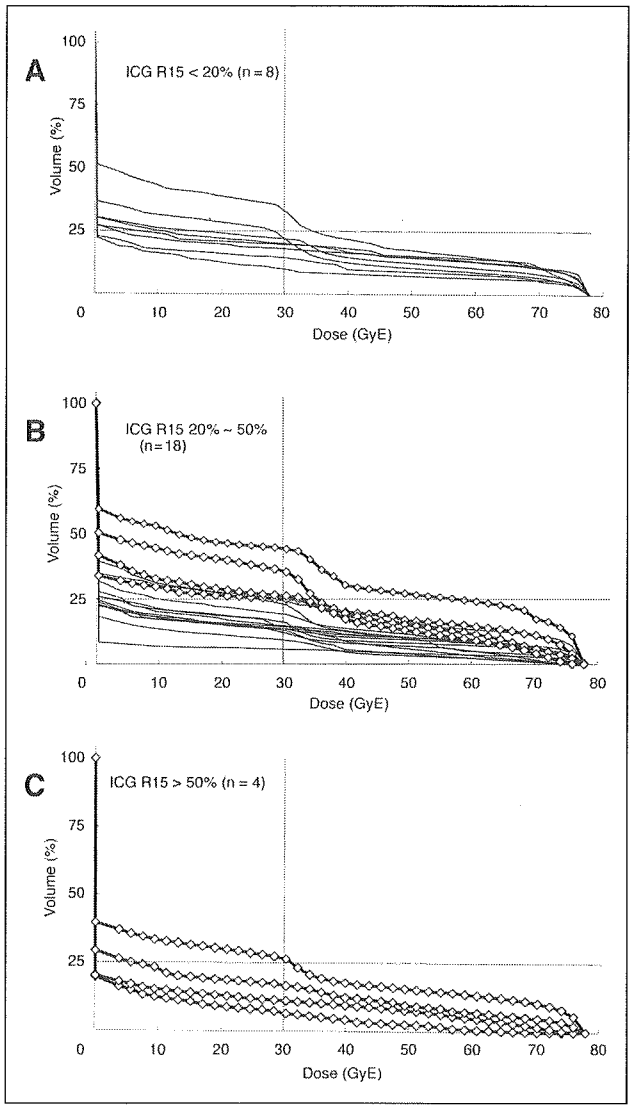

Fig 4. Dose-volume histogram (DVH) for all patients according to their pretreatment ICG R15 values, as noted in panels A, B, and C. Thick line with rhombi represents DVH for patients suffering from hepatic insufficiency within 6 months after completion of proton beam radiotherapy.

that seen in partial liver resection, rather than after 3-dimensional conformal or intensity-modulated radiotherapy delivering a low-dose of x-ray to a large proportion of noncancerous liver. Therefore, estimation of the risk of PRT-inducing hepatic insufficiency should be done with similar guidelines to evaluate liver tolerance to surgery, rather than that with normal tissue complication probability model using a mean dose administered to the entire liver.[21] Remnant liver volume and ICG R15 have been preferred indicators for that estimation, especially in Japan.[15] DVH analyses (Figs 4A to C) suggested that V_{30}% in combination with ICG R15 may be a useful indicator for estimation of liver tolerance to PRT, but no definite quantitative criteria emerged with the limited data obtained at present because of the small number of patients evaluated. The current staging system for HCC is based on survival data obtained in surgical series.[22] There is no reliable system to stratify the prognosis of patients with solitary but unresectable HCC on the assumption that they achieve good local control after PRT. Because of the limited availability of PRT at present, the establishment of particular criteria for patient selection using quantitative parameters of hepatic function such as ICG R15, and volume parameter like V_{30}%, is needed to maximize the cost-effectiveness of PRT.

Applicability of PRT instead of surgery for patients with early-stage disease should be considered with caution. Intraoperative ultrasonography (IOUS) has an important role in detecting small metastatic lesions, which could not be demonstrated in preoperative examinations. The high incidence of intrahepatic recurrences seen outside the PTV might be partly ascribable to the limit of pretreatment imaging studies. Infiltration of HCC to the portal vein and spread via portal blood flow is one of the mechanisms for the development of intrahepatic recurrence.[15] Actually, five recurrences occurred within the same segment of the primary tumor in this study. Although anatomic resection according to the architecture of the portal vein using IOUS offered a better chance of cure only for patients with noncirrhotic livers,[23] systematic segmental PRT based on multimodal imagings such as CT during arterial portography or MRI as well as image fusion technique[24] has a theoretical advantage compared with nonanatomic PRT confined to GTV only. Because there were few potentially curative approaches other than surgery for patients with HCC showing vascular invasion, further study is warranted to scrutinize an efficacy of PRT for patients with HCC of $\geq$ 5 cm in diameter, of which a large majority will demonstrate vascular invasion around the periphery of the tumor,[25] while giving attention to their V_{30}% values.

The risk of this aggressive dose-fractionation for sites such as the gastrointestinal loop, hepatic hilum, skin, or subcutanous tissues must be carefully considered, and more conventional fractionation must be adopted when these structures are critically involved in the PTV.

In conclusion, PRT for localized HCC using an aggressive dose-fractionation scheme (76 Gy_E for 5 weeks) achieved excellent local control rate regardless of vascular invasion or tumor size, if $\leq$10 cm, without devastating acute toxicity. Further study is warranted to scrutinize adequate patient selection according to quantitative parameter of hepatic function, such as ICG R15, and irradiated noncancerous liver volume in order to maximize survival benefit of this promising modality.

Authors' Disclosures of Potential Conflicts of Interest

The authors indicated no potential conflicts of interest.

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平成１７年度　研究報告書

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