ISSN: 0018-2052, CODEN: HIJMAC
Volume 65, Number 2
June 2016

Hiroshima Journal of Medical Sciences

Published by
Hiroshima University
Medical Press

Hiroshima Journal of Medical Sciences is published four times a year, by Hiroshima University Medical Press. Inquiries regarding manuscripts should be addressed to: Editorial Office, School of Medicine, Hiroshima University, 1-2-3 Kasumi, Minami-ku, Hiroshima 734-8551, Japan.

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Hiroshima J. Med. Sci.
Vol. 65, No. 2, 25~33, June, 2016
HIJM 65−5

Assessment of Outcome of Hepatic Arterial Infusion Chemotherapy in Patients with Advanced Hepatocellular Carcinoma by the Combination of RECIST and Tumor Markers

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ABSTRACT

To assess the outcome of stable disease (SD) patients with advanced hepatocellular carcinoma (HCC) by tumor markers after the first course of hepatic arterial infusion chemotherapy (HAIC). The study subjects were 156 HCC patients treated with HAIC and classified as Child Pugh A, with no extrahepatic metastasis, and no history of sorafenib treatment. In the study and validation cohorts, the AFP and DCP ratios of patients who were considered SD to the first course of HAIC were analyzed by AUROC for a prediction of response to the second course of HAIC. The imaging response to the first course of HAIC was classified as partial response (PR), SD and progressive disease (PD) in 29 (18.8%), 80 (51.9%), and 44 (28.6%) patients respectively. For SD patients, the α-fetoprotein (AFP) and des-γ-carboxy prothrombin (DCP) ratios of patients who were considered SD to the first course of HAIC were analyzed by the receiver operating characteristic curve for prediction of response to the second course of HAIC in the study cohorts. The area under the curve of AFP ratio was 0.743. The area under the curve of DCP ratio was 0.695. The cut-off values of AFP and DCP ratios were 1.3 and 1.0, respectively. In the validation cohort, the accuracy of the prediction of response in this validation cohort (71.4%) showed no significant difference compared to that in the study cohort (72.4%) ($p = 1.0$). The results suggested that patients with a high tumor marker ratio could be switched to alternative therapeutic regimens despite the SD response to HAIC.

Key words: *Hepatocellular carcinoma, Hepatic arterial infusion chemotherapy, RECIST, Tumor marker*

Hepatocellular carcinoma (HCC) is the sixth most common cancer and the second leading cause of cancer-related mortality worldwide[6,9)]. Advances in biotechnology have made it possible to develop new diagnostic techniques, such as ultrasonography, computed tomography (CT), magnetic resonance imaging (MRI), and angiography. Similarly, new treatment modalities have been invented, such as surgical resection, radiofrequency ablation (RFA), percutaneous ethanol injection (PEI), transcatheter arterial chemoembolization (TACE), and hepatic arterial infusion chemotherapy (HAIC), which have improved the prognosis of HCC patients[1,3,4,7,8,14,15,23,25)]. However, the survival rates of patients with advanced HCC and associated complications such as portal vein tumor thrombosis (PVTT), venous tumor thrombosis (VTT), and refractoriness to TACE, have not improved enough.

Two phase III clinical trials of sorafenib for advanced HCC showed significant efficacy in terms of overall survival time (OST) compared with a placebo [2,6)]. Based on these studies, sorafenib has become the standard therapy for advanced HCC. Sorafenib contributed to prolonging OST by 2.3-2.8 months and the response rate (RR) by 2.0-3.3%. However, the survival advantage of sorafenib is described as insufficient.

HAIC is widely undergone in Asia, especially Japan. Several studies have shown the survival benefits of HAIC for advanced HCC free of extrahepatic metastasis, with a response rate ranging from 20.8 to 52%, and have reported that median survival time (MST) in responders is 17.6-40.7 months[1,11,19,23,27,28)].

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However, large randomized trials are not demonstrated efficiently. In most of the retrospective studies, survival time was much longer in responders than in non-responders. We have reported that survival after switching HAIC treatment to sorafenib was better than that of continuous HAIC[20]. At present, however, there is no biomarker that can be used to predict the response to HAIC treatment. Such a marker could help in decision making on whether to continue HAIC treatment or not.

The treatment response to HCC is assessed by imaging studies. One of the most common methods for response evaluation is the Response Evaluation Criteria in Solid Tumors (RECIST) [5]. However, it is inefficient to evaluate response to the first course of treatment by imaging studies alone. On the other hand, some studies have shown the usefulness of a-fetoprotein (AFP) and des-γ-carboxy prothrombin (DCP) not only as tumor markers, but also as prognostic factors for HCC[13,17,22,26]. We reported that patients with AFP and DCP ratios of >1 had significantly poorer survival than others (MST 7.4 vs 12.6 months, p=0.014), among patients with stable disease (SD) based on imaging response to first course of chemotherapy[21]. To our knowledge, there are no clear cut-off values for AFP ratio and DCP ratio.

The present retrospective study was designed to analyze the cut-off values of the AFP and DCP ratios for outcome to the first course of chemotherapy in HCC patients with SD.

MATERIALS AND METHODS

Patients. Between June 2000 and March 2015, 364 patients with unresectable HCC were treated with HAIC at our hospital. HAIC was selected as the therapeutic option for patients with advanced HCC who presented also with PVTT and VTT, and refractoriness to TACE. We excluded the following patients from HAIC: *1)* The performance status of the Eastern Cooperative Oncology Group (ECOG) was ≥3 (n=1). *2)* Child–Pugh score of ≥7 (n=109), *3)* extrahepatic metastasis (n=80), *4)* treatment with sorafenib before and after HAIC (n=18). After the exclusion of the above 208 patients, the remaining 156 patients were enrolled in this retrospective cohort study (Fig. 1). The study protocol was approved by the Human Ethics Review Committee of Hiroshima University and a signed consent form was obtained from each subject.

Hepatic arterial infusion chemotherapy (HAIC). Patients were given arterial infusions of anticancer agents via the injection port. Two drug regimens are used in HAIC. We used intra-arterial low-dose cisplatin (CDDP, Nihonkayaku, Tokyo, Japan) with 5- fluorouracil (5FU, Kyowa Hakko, Tokyo) (FP), or intra-arterial 5FU with subcutaneous interferon (5FU+IFN). One course of chemotherapy was undergone for 2 weeks. 5FU (300 mg body weight/day) was administered over 24 hr by using a mechanical infusion pump from days 1 to 5 of the first and second weeks in both regimens. CDDP was injected intra-arterially via a pump at 6 mg/body weight/day on days 1-5 and 8-12. The IFN in the 5FU+IFN regimen was recombinant IFNα-2b [Intron A, Schering-Plough Pharmaceuticals, Osaka, Japan, 3×10^6 U (3 MU)], or natural IFN-α [OIF, Otsuka Pharmaceuticals, Tokyo, 5×10^6 U (5 MU)] administered intramuscularly on days 1, 3, and 5 of each week (total dose, 36 and 60 MU, respectively). We reported previously that recombinant IFNα-2b had an equal effect to natural IFN-α when the combination of 5FU+IFN was used for the treatment of advanced HCC[28].

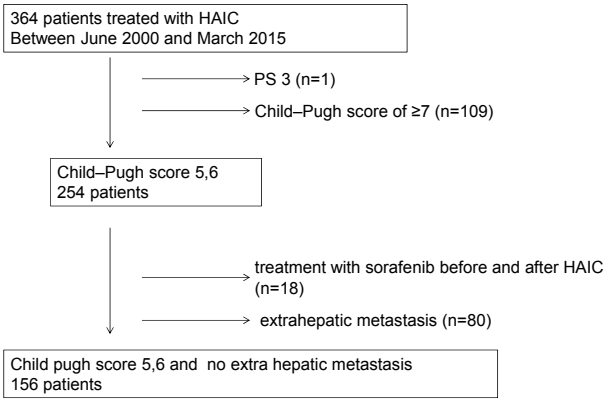

Fig. 1. Patient recruitment process

Assessment of response to HAIC. Each patient underwent dynamic CT before HAIC and also after each course of HAIC. In this study, we defined the terms imaging response and AFP/DCP tumor marker response. The imaging response to HAIC was evaluated by RECIST (version 1.1) on dynamic CT after the first course of HAIC (4 weeks later). A complete response (CR) was defined as the disappearance of all target lesions after one course of HAIC. A partial response (PR) was defined as the sum of the longest diameter reducing by more than at least 30% compared to before HAIC. Progressive disease (PD) was defined as the sum of the longest diameter of the target lesion increasing more than at least 20%. Stable disease (SD) was defined as corresponding neither to the criteria of PR nor PD. HAIC was continued repeatedly as long as the treatment response was better than SD. To evaluate the AFP/DCP tumor marker response, we measured these markers from the serum concentrations after each course of HAIC (each 4 weeks). In our hospital, the normal range of AFP is within 10 ng/ml, while that of DCP is within 30 mAU/ml. The AFP ratio represented the AFP value after one course of HAIC divided by the AFP value before treatment. The DCP ratio was measured similarly. When both tumor mark-

ers were within the normal range before and after treatment, the tumor marker ratio was ≤ 1.

Adverse drug reactions were defined according to the Common Terminology Criteria for Adverse Events version 4.0.

Follow-up and other therapies. Treatment with sorafenib was not administered throughout the clinical course. Instead, other therapies such as RFA, TACE and radiotherapy were used for partial and non-responders. PR patients continued to receive HAIC regularly in combination with other therapies. Patients whose advanced HCC was down-staged to a single tumor ≤ 50 mm in diameter or 1-3 tumors each ≤ 30 mm in diameter following the combination therapy, were considered to receive RFA or hepatectomy. In addition to HAIC, PD patients received TACE. TACE was used after HAIC in the following situations: 1) additional TACE aimed at downstaging HCC when patients showed an effective response to HAIC, and 2) palliative TACE aimed to prevent HCC rupture or rapid growth when patients showed non-response to HAIC. PD patients were also considered for radiotherapy when complicated with portal venous tumor thrombosis (PVTT). For CR patients, the clinical course was observed without adjuvant chemotherapy or additional therapy.

Statistical analysis. Statistical analysis was performed in August 2015. Differences between groups were examined for statistical significance using the Mann-Whitney U test, logistic regression test, or squared test, as appropriate. The cumulative survival rate was calculated from the date of initiation of HAIC, assessed by the Kaplan-Meier life-table method, and differences between groups were evaluated by the log rank test. Univariate analysis of the factors that correlated with survival of patients with HCC treated with HAIC was assessed by the Kaplan-Meier life-table method, and differences were evaluated by the log rank test. Multivariate analysis of the factors that influenced survival was assessed by the Cox proportional hazard model. Statistical significance was defined as p value of less than 0.05.

Cut-off points for continuous variables were determined by analysis of the receiver operating characteristic (ROC) curve based on the minimum balanced error rate (BER)[18]. BER is the average of the proportion of incorrect classifications in each class.

The cut-off value associated with maximum accuracy, sensitivity, and negative and positive predictive values of the PD to the second course of treatment was computed. The chi-squared test was used to compare the accuracy of the prediction score in the study cohort with that of the validation cohort.

All analyses described above were performed with The Statistical Package for Social Sciences software (version 11, SPSS, Chicago, IL).

RESULTS

Baseline characteristics. Patient characteristics are listed in Table 1. The study subjects were 140 men and 16 women, and the median age was 68

Table 1. Clinical characteristics of HCC patients treated with HAIC (n=156)

Age (years) *	68.0 (32-85)
Gender (M/F)	140/16
ECOG performance status (0/1/2)	133/21/2
Child-Pugh score (5/6)	84/72
Etiology (HBV/HCV/others)	42/85/29
Number of HCC tumors (solitary/multiple)	22/134
Size of liver tumor (mm) *	60 (10-180)
HCC stage (II/III/IVa) †	4/49/103
Vp (0/1/2/3/4) §	49/6/24/44/33
Vv (0/1/2/3) ‡	132/1/10/13
Relative tumor size in the liver (<50%/≥50%)	117/39
Platelet count (/mm^3) *	12 (4.6-88.8)
AFP (ng/ml) *	464 (2.9-1895000)
DCP (mAU/ml) *	1733 (7-666480)
HAIC regimen (FP/5FU+IFN)	86/70

*Data are median and (range) values, or number of patients.

†According to the Liver Cancer Group of Japan.

§Portal invasion. ‡Venous invasion.

CP: Child Pugh, ECOG: Eastern Cooperative Oncology Group, HCV: hepatitis C virus, HBV: hepatitis B virus, HCC: hepatocellular carcinoma, Vp2: tumor thrombus in the second branch of the portal vein, Vp3: tumor thrombus in the first branch of the portal vein, Vp4: tumor thrombus in the trunk of the portal vein, Vv2: tumor thrombus in the right, middle or left hepatic vein trunk, posterior inferior hepatic vein trunk or short hepatic vein, Vv3: tumor thrombus in inferior vena cava, AFP: α-fetoprotein, DCP: des-γ-carboxy prothrombin, FP: intra-arterial low-dose cisplatin and 5FU therapy, 5FU+IFN: intra-arterial 5-FU with IFN combination therapy.

years. The Child-Pugh score was 5 points in 84 patients and 6 points in 72 patients. The background liver disease was hepatitis C viral (HCV) infection in 85 patients, hepatitis B viral (HBV) infection in 42, and non-HCV-non-HBV in 29. Solitary HCC was detected in 22 patients and multiple HCCs in 134. HCC was classified as stage II, III and IVa in 4, 49 and 103 patients, respectively. Portal venous invasion was identified in 107 patients and venous invasion in 24 patients. The median value of AFP was 464 ng/ml and that of DCP was 1733 mAU/ml.

Imaging response after first course of HAIC and overall survival. Imaging response by RECIST to the first course of treatment was CR in one (0.6%) patient, PR in 29 (18.8%), SD in 80 (51.9%) and PD in 44 (28.6%) patients. MST varied significantly among the Imaging response groups ($p < 0.0001$) and was 26.6, 12.2 and 5.5 months in the PR, SD and PD groups, respectively (Fig. 2). The percentage of SD patients was more than 50%, and accordingly we examined the factors that could stratify the survival of SD patients to the first course of HAIC. In the median survival time of HAIC, there is no significant difference between FP (MST 11 months) and 5FU+IFN (MST 10 months) (p=0.8).

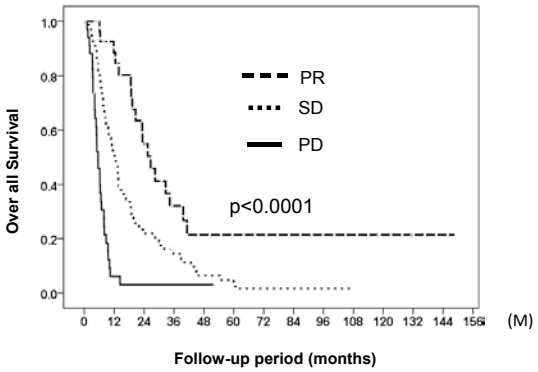

Fig. 2. Cumulative survival rates according to imaging response to the first course of HAIC

Background of SD patients by imaging response to first course. Table 2 lists the background of SD patients according to imaging response to the first course of HAIC. Among the 156 patients, 80 patients were classified as SD by the imaging response.

With regard to AFP and DCP, 23 patients who were treated with warfarin or vitamin K were excluded from analysis. Of the remaining 57 patients, 54 were men and 3 were women, with a median age of 68 years. The background liver disease was HCV infection in 36 and other diseases in 21. The Child-Pugh score was 5 in 28 patients and 6 in 29 patients. HCC was classified as stage II, III, and IVa in 1, 17, and 39 patients, respectively. In this group, the median value of AFP ratio was 1.27 while that of DCP ratio was 0.99. For the study cohort, data from 28 consecutive patients who were treated between 2000 and 2007 with HAIC were collected. Data from 29 patients treated between 2007 and 2015 were also collected as an independent validation cohort (Table 3). There was no significant difference between study cohort and validation cohort in their background.

Imaging response to second course of HAIC and overall survival among SD patients to first course of HAIC. Patients who were considered SD to the first course of HAIC were assessed again by CT after the second course of HAIC. One (1.65%) patient was classified as CR, 11 (17.7%) as PR, 27 (43.5%) as SD and 23 (37.1%) patients as

Table 2. Background of SD patients (n=57) according to the imaging response to the first course of chemotherapy

Age (years) *	68 (34-85)
Gender (M/F)	54/3
ECOG performance status (0/1)	50/7
Child-Pugh score (5/6)	28/29
Etiology (HBV/HCV/others)	13/36/8
Number of HCC tumors	4 (1-100)
Size of liver tumor (mm) *	69 (10-180)
HCC stage (II/III/IVa) †	1/17/39
Vp (0-2/3-4) §	31/26
Vv (0-1/2-3) ‡	50/7
Relative tumor size in the liver (<50%/≥50%)	42/15
Platelet count (/mm^3) *	12.2 (5.1-49.8)
AFP (ng/ml) *	387.3 (2.9-869800)
DCP (mAU/ml) *	1512 (10-120070)
AFP ratio *	1.27 (0.02-29.3)
DCP ratio *	0.99 (0.008-12.4)
Regimen (IFN+5FU/CDDP+5FU)	22/35

For abbreviations see Table 1.

Table 3. Background of SD patients (n=57) according to the imaging response to the first course of chemotherapy in the study cohort and the validation cohort

	Study cohort (n=28)	validation cohort (n=29)	p value
Characteristics			
Age (years) *	67.0 (45-85)	75 (34-84)	0.506
Gender (M/F)	26/2	28/1	0.532
ECOG performance status (0/1)	25/3	25/4	0.723
Child-Pugh score (5/6)	11/17	17/12	0.144
Etiology (HBV/HCV/others)	4/21/3	9/15/5	0.182
Number of liver tumors *	6 (1-40)	5 (1-20)	0.435
Size of liver tumors (mm) *	50 (18-105)	80 (21-180)	0.089
HCC stage (II/III/IVa) †	1/7/20	0/10/19	0.487
Vp (0-2/3-4) §	15/13	16/13	0.903
Vv (0-1/2-3) ‡	25/3	25/4	0.723
Relative tumor size in the liver (<50%/≥50%)	20/8	22/7	0.704
Platelet count (/mm^3) *	10.3 (5.1-49.8)	14.8 (6.1-39.5)	0.143
AFP (ng/ml) *	905 (10-394000)	332 (2.9-869800)	0.893
DCP (mAU/ml) *	1702 (10-120070)	2868 (41-102590)	0.893
HAIC regimen (FP/5FU+IFN)	11/17	11/18	0.916
Response to the first course (CR/PR/SD/PD)	1/8/8/11	0/2/17/10	0.021

For abbreviations see Table 1.

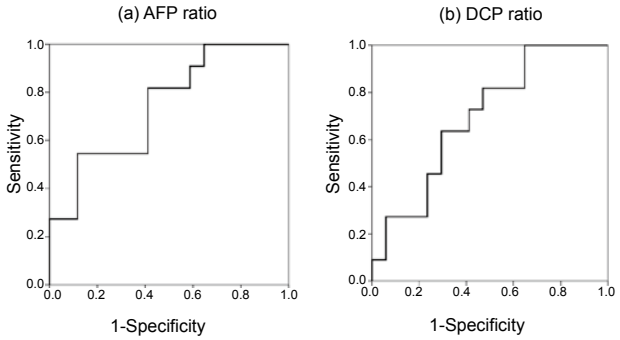

Fig. 3. Area under the receiver operating characteristics curve for (a) AFP ratio and (b) DCP ratio among SD patients to the first course of HAIC in study cohort.
The cut-off value associated with maximum accuracy, sensitivity, and negative and positive predictive values of the PD to the second course of treatment was computed.

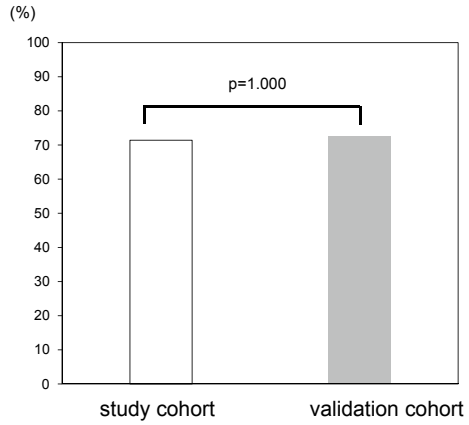

Fig. 4. The accuracy of the prediction of response to the second course of HAIC in study and validation cohort

PD. Among the SD patients to the first course HAIC and MST varied significantly ($p < 0.0001$) and were 32.1, 13.3 and 6.9 months in the PR, SD and PD groups, respectively.

Prediction of response to the second course of HAIC in the study and validation cohorts. First, the AFP and DCP ratios of patients who were considered SD to the first course of HAIC were analyzed by ROC for prediction of response to the second course of HAIC in the study cohort (Fig. 3). The sensitivities and specificities of the AFP and DCP ratios among these patients are shown in Figs. 3a and b. The area under the curve of AFP ratio was 0.743, with a sensitivity of 81.8% and specificity of 58.8%. The area under the curve of the DCP ratio was 0.695, with a sensitivity of 63.6% and specificity of 58.8%. The cut-off values of the AFP and DCP ratios were 1.3 and 1.0, respectively. That is, AFP ratio >1.3 and DCP ratio >1.0 were defined as a prediction of PD to the second course. We next evaluated the accuracy of the prediction score using an independent validation cohort consisting of 29 patients. The positive predictive value (PPV) and negative predictive value (NPV) for PD were 66.7% and 73.6% in the study cohort. On the other hand, the PPV and NPV for PD were 75.0% and 72.0% in the study cohort.

The accuracy of the prediction of response in this validation cohort (71.4%) showed no significant difference compared to that in the study cohort (72.4%) (p=1.0, Fig. 4).

Table 4. Results of multivariate analysis of determinants of survival in SD patients to the first course of HAIC

Parameters	Univariate analysis		Multivariate analysis	
	p value	Hazard ratio	95% CI	p value
All patients				
Age (<65/≥65 years)	0.894			
Gender (M/F)	0.454			
Etiology (HCV/others)	0.05			
AFP ratio >1.3 and DCP ratio >1.0/others	0.0025	2.012	1.36-3.907	0.035
Relative size of liver tumor (50%/≥50%)	0.005			
Vascular invasion (positive/negative)	0.903			
Number of liver tumors (single/multiple)	0.649			
Size of liver tumor (<50 mm/≥50 mm)	0.24			
TACE refractoriness (presence/absence)	0.228			
Regimen (IFN+5FU/CDDP+5FU)	0.176			

For abbreviations see Table 1.

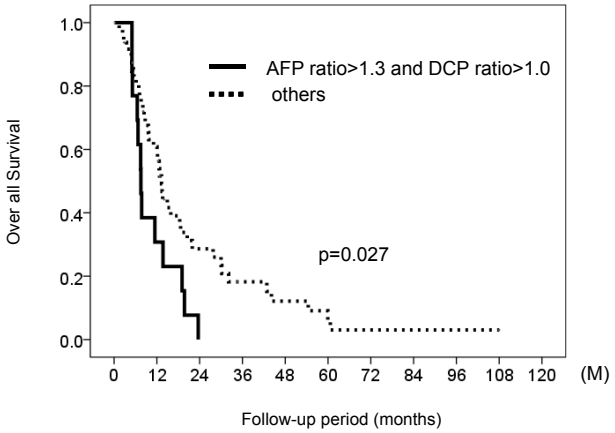

Fig. 5. Overall survival among SD patients to the first course of HAIC, according to tumor markers.
Solid line: patients with AFP ratio > 1.3 and DCP ratio > 1.
Dotted line: others.

Multivariate analysis of factors contributing to overall survival in SD patients by imaging response to first course. Univariate analysis was used to investigate the relationship between overall survival of patients who were considered SD to the first course of HAIC. Overall survival correlated significantly with etiology (p=0.05), AFP ratio > 1.3 and DCP ratio > 1.0 (p=0.0025), and tumor size relative to liver size (p=0.005). Inclusion of the above factors in multivariate analysis showed that an AFP ratio > 1.3 and DCP ratio > 1.0 was the only determinant of overall survival in patients considered SD to the first course of HAIC (p=0.035: hazard ratio 2.012, 95%CI 1.36-3.907) (Table 4).

Overall survival according to AFP and DCP ratios among SD patients to first course of HAIC. The MST of SD patients with AFP ratio of ≤ 1.3 and DCP ratio of ≤ 1, > 1.3 and ≤ 1, ≤ 1.3 and > 1, and > 1.3 and > 1 were 13.3, 12.1, 13.6 and 7.5 months, respectively (p=0.067). We also divided the patients into two groups: AFP ratio of > 1.3 and DCP ratio of > 1, and others. SD patients with AFP ratio of > 1.3 and DCP ratio of > 1 had a significantly poorer survival than others (MST 7.5 vs 13.3 months, p=0.027, Fig. 5). These results indicated that the cut-off values of AFP and DCP ratios could be used to predict the overall survival after the second course of HAIC in SD patients to the first course.

DISCUSSION

The response to HCC treatment is assessed according to RECIST or mRECIST with imaging modalities. In clinical practice, there is no biomarker that can be used to predict the response to HAIC and, accordingly, there are no criteria that can be used for continuation or discontinuation of HAIC. Patients who show a CR or PR response should continue HAIC while those who show PD should be switched to other treatments including sorafenib. However, in our hospital, the number of patients who showed a SD response was more than half of all patients. For this reason, we analyzed their data to identify HCC tumor markers that could predict overall survival. The results showed that patients with the combination of an AFP ratio of > 1.3 and a DCP ratio of > 1 had significantly poorer survival than others among SD patients to the first course of HAIC.

Previous studies analyzed the prognosis of HCC patients treated with HAIC using RECIST or mRECIST with imaging modalities, which is regarded as the gold standard for evaluation of therapeutic response. Sorafenib was introduced recently as a molecular targeting therapy for advanced HCC, though there are no guidelines for assessment of the response to such treatment. In the present study, we analyzed first the treatment response to the first course of HAIC by the combination of RECIST and tumor marker ratios. We also analyzed the data for whether the response to the first course of HAIC can be used to predict the prognosis of patients. The results showed that the

AFP and DCP ratio can be used to determine treatment selection; i.e., continuation or change from HAIC. We reported previously that the survival of patients with AFP and DCP ratios of > 1 was significantly poorer and that the response did not change to CR or PR during the course of treatment in SD patients[21]. However, the cut-off values of AFP and DCP were decided without statistics. That is, we decided the cut-off values of AFP and DCP by only the elevation after treatment. As a result, we decided the cut-off values of AFP and DCP by ROC analysis in the present study. Furthermore, in order to confirm that the cut-off values were appropriate, we studied the cut-off values of AFP and DCP by study cohort and validation cohort in the present study. Therefore, the present study determined the cut-off values of AFP and DCP ratios by statistics. Based on the imaging responses to the second course of HAIC, the median survival time was 6.9 months in the PD groups. Therefore, we used ROC analysis to determine the cut-off values that were associated with the highest accuracy, sensitivity, and negative and positive predictive values of PD to the second course of HAIC. The results showed that the best cut-off values were 1.3 for the AFP ratio and 1.0 for the DCP ratio in the study cohort. We next evaluated the accuracy of the prediction response using an independent validation cohort consisting of 29 patients. The accuracy of the prediction of response in this validation cohort (71.4%) was not significantly different compared to that in the study cohort (72.4%) (p=1.0).

Further analysis showed that patients with an AFP ratio of > 1.3 and DCP ratio of > 1 had significantly poorer survival than others (MST 7.5 vs 13.3 months, p=0.027), indicating that the tumor marker response can accurately predict refractoriness to HAIC.

Saeki et al [24] categorized their patients according to Child-Pugh, AFP and DCP responses after a half course of HAIC (2 weeks) and showed significantly different prognoses. However, they defined AFP- or DCP-positive-response as a reduction in serum AFP or DCP of more than 20% from baseline after half a course of HAIC. However, no reason was given for the selection of 20% reduction. It is possible that some patients showed a good overall survival despite a less than 20% reduction in serum AFP or DCP after a half course of HAIC. In our study, none of the patients who showed PR or CR in the second course had a AFP ratio of > 1.3 and DCP ratio of > 1 because the cut-off was determined by ROC analysis. Furthermore, the study of Saeki et al [24] included patients with extrahepatic metastasis and classified it as Child-Pugh B. It is reported that patients with extrahepatic metastases treated with HAIC show poor overall survival[10]. Furthermore, overall survival is also poor in Child-Pugh B patients treated with sorafenib[12]. Thus, the above study included many biases with the exception of tumor markers. In comparison, our study was limited to tumor markers, excluded patients treated with sorafenib before and after HAIC, extrahepatic metastasis and Child-Pugh B, and thus allowed us to study the prognosis of patients treated by HAIC.

Sorafenib is currently the standard treatment for advanced HCC patients. In two randomized studies, placebo-controlled clinical trials, sorafenib extended overall survival by 2.3-2.8 months and the response rate by 2.0-3.3%[2,16]. Although the effectiveness of HAIC for advanced HCC has been reported in some reports, large randomized trials are lacking. To our knowledge, there is no defined strategy for the standard of treatment with sorafenib and HAIC for advanced HCC patients. We think that it is important to pick up responders to HAIC as early as possible. In other words, HAIC must be switched to sorafenib as early as possible for PD patients of HAIC. Therefore, in this study, the patients were limited to Child-Pugh A patients who could be treated with sorafenib. The efficacy of sorafenib and HAIC on advanced HCC are currently being assessed in a few clinical trials in Japan. We are currently conducting an ongoing HICS study (pilot study of HAIC followed by sorafenib for advanced HCC, UMIN#000009094). Another Japanese clinical study based on the same purpose (HAIC followed by sorafenib) is ongoing: the SCOOP-II trial (Sequential hepatic arterial infusion chemotherapy with cisplatin followed by sorafenib versus sorafenib alone in advanced hepatocellular carcinoma, UMIN#000006147). These studies are designed to pick up refractoriness to HAIC using the combination of imaging response and tumor marker response after every course of therapy.

The present study has certain limitations. First, it was a retrospective cohort study that examined a small population. A prospective study of a larger patient population is needed to confirm the findings. Second, various chemotherapeutic regimens were used in the study population. However, previous studies showed no significant differences in response or survival among these regimens[21]. A validation study of HCC patients treated with a single regimen is required.

In conclusion, we used RECIST to evaluate the response of SD patients to the first course of HAIC and demonstrated that the combination of AFP ratio of > 1.3 and DCP ratio of > 1 could be used to predict the prognosis of patients with advanced HCC. The results emphasize the need to switch to alternative therapies in patients with a high tumor marker ratio despite SD response to HAIC.

(Received April 5, 2016)
(Accepted April 26, 2016)

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Hiroshima J. Med. Sci.
Vol. 65, No. 2, 35~43, June, 2016
HIJM 65−6

High Initial-dose Dependency of Cerebrovascular Disease Mortality among Female Survivors of the Hiroshima Atomic Bomb Exposed in Teens: A Cohort Study, 1970-2010

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ABSTRACT

Several studies have been conducted on cerebrovascular disease mortality in Atomic bomb survivors. Previous studies have investigated the relationship between mortality and initial radiation dose after adjusting for the effects of sex and age at the time of the bombing (ATB), and detected a weak (but statistically significant) dose-response relationship was detected. The objective of the present study was to examine whether the sex- and age ATB-specific cerebrovascular disease mortality among Hiroshima atomic bomb survivors can be explained by the initial radiation dose. At Hiroshima University, a cohort study has been conducted with Hiroshima Atomic Bomb Survivors (ABS) since 1970. We selected 30,378 subjects from the ABS who were exposed at 3.5 km or less from the hypocenter and still alive on January 1, 1970. These subjects were followed up until December 31, 2010. The cohort data were stratified with respect to sex and age ATB into 10-year age groups. For each stratum, using Cox regression, we performed survival analyses of the risk of cerebrovascular mortality using the initial radiation dose and the exposure distance (the ground distance between the exposure location and the hypocenter) as explanatory variables. The results indicated that the risks to females exposed at 10 to 19 years old were highly dependent on the initial radiation dose (hazard ratio: 1.51, $p < 0.001$), while the risks to males were not. There might exist some radiation exposure effects limited to women who were in their teens at the time of exposure. However, the background mechanisms remain unclear, necessitating further study.

Key words: *Atomic bomb survivors, Cerebrovascular disease, Dose-response relationship, Exposure-distance*

Arteriosclerotic cardiovascular disease is now the cause of death of approximately one in three Japanese people. In the 1960s, cerebrovascular disease was the leading cause of death in Japan, but it is now the third leading cause, with a dramatically decreasing tendency since the 1970s[7]. With approximately 70 years having passed since the atomic bombs were dropped, almost all atomic bomb survivors (*hibakusha*) now belong to the generation in which the onset of cardiovascular disease is common, and the effects of radiation on cardiovascular disease have been well studied. However, we have not yet clearly grasped the impact of exposure to radiation from atomic bombs on cerebrovascular disease. Preston et al observed a significant correlation with initial radiation dose after adjusting for the effects of sex and age at the time of the bombing (ATB), reporting an excess relative risk of death due to cerebrovascular disease of 0.12 (90% confidence interval: 0.02–0.22) per Sv of radiation dose[22]. Shimizu et al also observed a non-significant ($p = 0.23$) correlation with radiation dose, reporting an excess relative risk per Gy of 36%, 9%, 15% and 5% for ages < 10, 10-19, 20-39, and ≥ 40[25]. These studies examined the relationship between the initial radiation dose (initial dose) and the mortality risk. Atomic bomb radiation exposure can be divided into two types: direct exposure (gamma-ray as well as immediate and delayed neutron radiations) from the initial explosion and indirect exposure from residual radiation comprising neutron activated radiation in

soil and other materials as well as fallout from the nuclear explosion. The initial dose is determined by the ground distance from the hypocenter to the victim's location at the time of exposure (exposure distance) and shielding conditions assessed from information provided by exposed persons, such as being in buildings or other structures at the time of the bombing. If all of this information is available, the individual direct, initial radiation dose may be estimated. However, it is almost impossible to estimate the individual amount of exposure from residual radiation, because of uncertain or lacking information on individual movements and activities just after the bombing.

The impact of residual radiation as a health hazard has traditionally been assumed to be negligible[12,26]. However, it is becoming clear that the increase in several health risks related to *hibakusha* cannot be explained by the effect of initial dose alone[24]. Tonda et al[28] showed that the geographical distribution of the risk of solid cancer mortality among Hiroshima *hibakusha* is not circular asymmetry around the hypocenter. Recently, Kerr et al[13] reported that the health risk among atomic bomb survivors in Hiroshima and Nagasaki of residual radiation from neutron-activated radionuclides in the airburst's dust stem and pedestal and in uplifted soil might be not negligible. Tonda et al[27] suggested the impact of indirect exposure as a factor in the increased leukemia risk for those who entered Hiroshima City on 6 August 1945. Otani et al[17] reported that the mortality risk for malignant neoplasms (excluding leukemia) was significantly higher for those who entered Hiroshima City on 6 August 1945. The results of both of those studies apply as well to *hibakusha* who were directly exposed age ATB since they were also exposed to residual radiation. Ohtaki et al[16] reported that the mortality rate for solid cancers among *hibakusha* is influenced not only by the initial dose of radiation but also by indirect exposure.

In observational epidemiologic studies, confounding is one of the major limitations. Shimizu et al[25] indicated that smoking, alcohol intake, education, occupation, obesity, and diabetes had had almost no impact on radiation risk estimates for stroke. In our study, it was impossible to analyze the data by adjusting confounding factors such as high blood pressure, a smoking habit, etc. For even if the association between radiation risk and cerebrovascular disease is actually due to some confounding factor so that radiation risk is not causally related to cerebrovascular disease, screening for radiation risk can nevertheless be useful because it permits us to identify people who are at high risk for the disease. The objective of the present study was to examine whether the sex- and age ATB-specific cerebrovascular disease mortality among *hibakusha* can be explained solely by the initial radiation dose.

SUBJECTS AND METHODS

For the present study we used the database of Atomic Bomb Survivors (ABS) that has been managed by the Research Institute for Radiation Biology and Medicine (RIRBM) of Hiroshima University[14]. The ABS differs from the Life Span Study (LSS) of the Radiation Effects Research Foundation (RERF), which that is based on subjects throughout Japan, in that the subjects in the ABS are restricted to Atomic bomb survivors residing in Hiroshima Prefecture. From the ABS, we chose for analysis 30,378 subjects (11,683 males and 18,695 females) who satisfied the following conditions: (i) alive and recognized as an atomic bomb survivor as of January 1, 1970 (the start of the observation period) and (ii) information available on the coordinates of their location at the time of atomic bomb exposure (abbreviated as "location at exposure"). The distance from the hypocenter to the location at exposure (abbreviated as "exposure distance") was within 3.5 km. These subjects were followed until December 31, 2010 for death from cerebrovascular disease (number of deaths: 1,006 among males, 1,945 among females). The death information including the cause of death was obtained from the Vital Statistics Death Schedules which are based

Table 1.1. Number of subjects, events, and censored cases by sex and age at time of exposure (ATB)

(Males)

age ATB (yrs)	number of subjects	number of events	number of censored cases†	number of surviving cases††
[0, 10)	3472	46	555	2871
[10, 20)	2877	121	1179	1577
[20, 30)	1139	105	812	222
[30, 40)	1684	253	1321	110
[40, 50)	1725	318	1322	85
[50, 60)	703	146	526	31
[60, 70)	78	16	61	1
[70, 80)	5	1	4	0
total	11683	1006	5780	4897

(Females)

age ATB (yrs)	number of subjects	number of events	number of censored cases†	number of surviving cases††
[0, 10)	3332	23	1129	2180
[10, 20)	3701	104	1291	2306
[20, 30)	3708	279	2077	1352
[30, 40)	3701	557	2954	190
[40, 50)	2943	638	2301	4
[50, 60)	1112	294	818	0
[60, 70)	188	49	139	0
[70, 80)	10	1	9	0
total	18695	1945	10718	6032

†Number of persons who emigrated out of Hiroshima prefecture or who died from other causes of death.

††Number of persons still alive at 31 Dec 2010.

Table 1.2. Number of subjects, events, and censored cases by sex and exposure distance

(Males)

distance (km)	number of subjects	number of events (CVD)	number of censored cases†	number of surviving cases††
[0.0, 0.8)	1	0	1	0
[0.8, 1.0)	159	14	103	42
[1.0, 1.2)	454	52	257	145
[1.2, 1.4)	967	100	498	369
[1.4, 1.6)	1223	90	610	523
[1.6, 1.8)	994	94	506	394
[1.8, 2.0)	705	66	373	266
[2.0, 2.5)	1083	97	549	437
[2.5, 3.0)	3959	310	1895	1754
[3.0, 3.5)	2138	183	988	967
total	11683	1006	5780	4897

(Females)

distance (km)	number of subjects	number of events (CVD)	number of censored cases†	number of surviving cases††
[0.0, 0.8)	5	0	2	3
[0.8, 1.0)	275	27	200	48
[1.0, 1.2)	909	100	582	227
[1.2, 1.4)	1806	210	1089	507
[1.4, 1.6)	2380	271	1425	684
[1.6, 1.8)	1768	182	1048	538
[1.8, 2.0)	1156	128	650	378
[2.0, 2.5)	1354	136	759	459
[2.5, 3.0)	5526	543	2987	1996
[3.0, 3.5)	3516	348	1976	1192
total	18695	1945	10718	6032

†Number of persons who emigrated out of Hiroshima prefecture or who died from other causes of death.

††Number of persons still alive at 31 Dec 2010.

Table 1.3. Number of subjects, events, and censored cases by sex and dose categories

(Males)

dose(Sv)	number of subjects	number of events (CVD)	number of censored cases†	number of surviving cases††
[0.00, 0.01)	6931	561	3305	3065
[0.01, 0.05)	682	75	385	222
[0.05, 0.1)	731	65	358	308
[0.1, 0.2)	972	84	503	385
[0.2, 0.4)	1027	84	532	411
[0.4, 0.6)	399	37	186	176
[0.6, 0.8)	250	27	116	107
[0.8, 1.0)	190	28	112	50
[1.0, 1.5)	193	16	102	75
[1.5, 2.0)	111	10	62	39
[2.0, 6.0)	197	19	119	59
total	11683	1006	5780	4897

(Females)

dose(Sv)	number of subjects	number of events (CVD)	number of censored cases†	number of surviving cases††
[0.00, 0.01)	9960	975	5466	3519
[0.01, 0.05)	1170	134	684	352
[0.05, 0.1)	1339	139	783	417
[0.1, 0.2)	1833	212	1094	527
[0.2, 0.4)	1941	227	1174	540
[0.4, 0.6)	771	85	444	242
[0.6, 0.8)	448	44	266	138
[0.8, 1.0)	397	48	264	85
[1.0, 1.5)	341	31	216	94
[1.5, 2.0)	173	20	101	52
[2.0, 6.0)	322	30	226	66
total	18695	1945	10718	6032

†Number of persons who emigrated out of Hiroshima prefecture or who died from other causes of death.

††Number of persons still alive at 31 Dec 2010.

on the official death certificates. Subjects who were alive at the end of follow-up, migrated outside Hiroshima Prefecture, or died from other causes were treated as censored (5,780 males and 10,718 females). Numbers of subjects categorized by age ATB and exposure distance are shown in Table 1.1 and 1.2 for each sex, respectively. From these tables, it is indicated that about 40% of the subjects were under 20 years-old at the bombing in this cohort data, and that approximately 5% of the subjects were bombed near the hypocenter with the distance less than 1.2 km.

1. Radiation dosimetry

To access the effect of initial radiation dose on the human body (unit of measurement: Sv), we used the red bone marrow absorbed neutron and gamma doses (unit of measurement: Gy) estimated using the Atomic Bomb Survivor 1993 Dose (which is referred to as ABS93D)[8]. The radiation dose calculated with ABS93D is based on the initial radiation exposure only, as is DS86, and ignores the effects of residual radiation[20-22]. The extent of overlap between survivors in the ABS and the LSS was examined by Hayakawa et al[6] in which

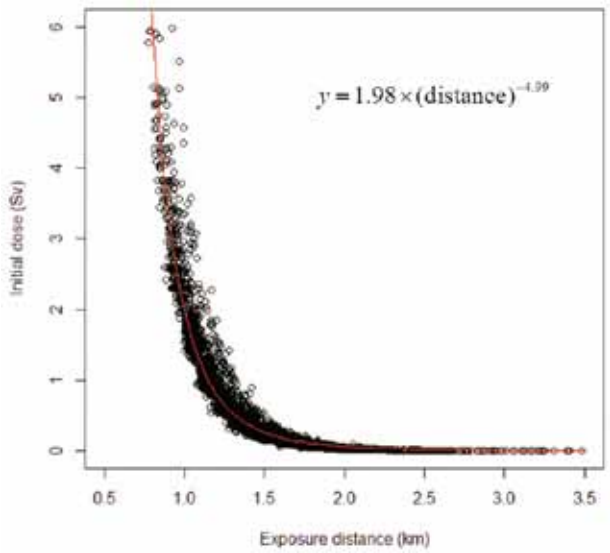

Fig. 1. Scatterplot of initial radiation dose versus exposure distance

it was shown that the dose estimates of the ABS were close to those of the LSS among overlapping subjects. However, it has not yet been investigated

how ABS93D corresponds with DS02. Table 1.3 shows the sex-specific numbers of subjects categorized by initial dose. About 70% of the subjects belong to the low-doses (less than 100 mSv) exposure group and approximately 4% belong to the high-doses (1.0 Sv or more) exposure group. Figure 1 shows a scatterplot of subjects' initial radiation dose versus exposure distance with the fitted curves based on a power function of exposure distance. It is noted that the initial radiation dose for the subjects can be roughly fitted by an inverse of 5th power function of exposure-distance.

2. Kaplan-Meier Curves

Assuming that there would be sex differences in the risk of cardiovascular disease[10] and a dependency on age ATB among *hibakusha*, we stratified the cohort data by sex and age ATB, and calculated the probability of not dying from cerebrovascular diseases (abbreviated simply as "survival probability") by initial dose and exposure distance using the Kaplan-Meier method[11]. Figures 2.1~2.4 show the survival rate for each sex- and age ATB-specific stratum. For these strata, we compared the survival curves of the cohorts exposed to low doses (less than 100 mSv) and high doses (1.0 Sv or more), and the survival curves of the cohorts exposed at short distances (less than 1.2 km) and long distances (2.0 km or more) using the log-rank test.

3. Cox Regression analysis

To quantify the effect of initial dose and exposure distance in detail, we conducted a survival analysis using a mathematical model as defined below. Based on the epidemiological observation that the mortality risk from cerebrovascular disease rises exponentially with increasing age[30], the hazard at attained age t, which incorporates the effect of the initial radiation dose D at age a (the initial-dose model), is expressed by:

$$h(t \mid D, a) = \exp\left(\beta_a D\right) \cdot \exp\left\{g(t, t-a+1945) + \delta\right\},$$

where β_a is the regression coefficient for the impact of the initial dose among *hibakusha* who were age a at the time of exposure, while g (t,y) is a logarithmic function of calendar year (y) and age (t) for the cerebrovascular disease mortality rate in all of Japan during the period 1970 to 2010, which is specified approximately by a quintic polynomial equation of t and y. The parameter δ denotes a coefficient expressing the logarithmic value of the background relative mortality risk of cerebrovascular disease for the Hiroshima *hibakusha* compared with the whole of Japan. Further, to investigate the impact of exposure distance, the following function of exposure distance r (in km, where the value for r = 2 km or more was assumed to be 0) was used as an alternative variable in place of initial radiation dose D,

$$D^*(r \mid \mu) = \begin{cases} \dfrac{\mu - r}{\mu - 1}, & r < \mu, \\ 0 \quad , & r \geq \mu. \end{cases} \qquad (1 < \mu \leq 2)$$

where μ is a parameter denoting the threshold of exposure-distance effect. Thus the distance-function model is expressed by:

$$h(t \mid D^*, a) = \exp\left(\beta_a D^*\right) \cdot \exp\left\{g(t, t-a+1945) + \delta\right\}$$

The cohort data were classified by sex and by age ATB into eight strata—[0, 10), [10, 20), [20, 30), and [30, 80) groups for each sex—and a time-dependent Cox regression analysis[2,3] was applied to each stratum with a hazard model using initial dose D or exposure-distance function D^*, where the estimated regression coefficient β_a expresses the stratum-averaged effect of initial radiation dose or exposure distance and the estimated parameter μ is a location parameter in the-exposure-distance function. To fit the model, we adopted the optimize function[23] in the R software (version 3.0.0). Since it is noted that the model with the minimum AIC has the best goodness of prediction, we used AIC to select the optimal model from the three models (the initial-dose model, the distance-function model and the null model with neither initial dose nor distance function as explanatory variables).

The null model is expressed by:

$$h(t \mid a) = \exp\left\{g(t, t-a+1945) + \delta\right\}$$

Significance tests in all cases were at the 5% level with a two-tailed test.

RESULTS

Kaplan-Meier survival curves by initial dose for men and women are shown in Figs. 2.1 and 2.2, respectively, and those by exposure distance are shown in Figs. 2.3 and 2.4. For men, the log-rank test indicated no significant difference between the low-dose (less than 100 mSv) and high-dose (1.0 Sv or more) exposure groups in any age-ATB stratum (Fig. 2.1). Similarly, no significant differences were observed in survival rates between the short-distance (less than 1.2 km) and long-distance (2.0 km or more) groups in any age-ATB stratum (Fig. 2.3). For women whose age ATB was between 10 and 19, the survival rate of the high-dose (1.0 Sv or more) group was significantly lower than that of the low-dose (less than 100 mSv) group (Fig. 2.2). Further, for women whose age ATB was either between 10 and 19 or 50 or more, the survival rate in the short-distance (less than 1.2 km) group was significantly lower than that in the long-distance (2.0 km or more) group (Fig. 2.4). In all other strata of age ATB, no significant differences in survival rates were observed.

Tables 2.1 and 2.2 display estimated values and significance of the regression coefficient β. In all

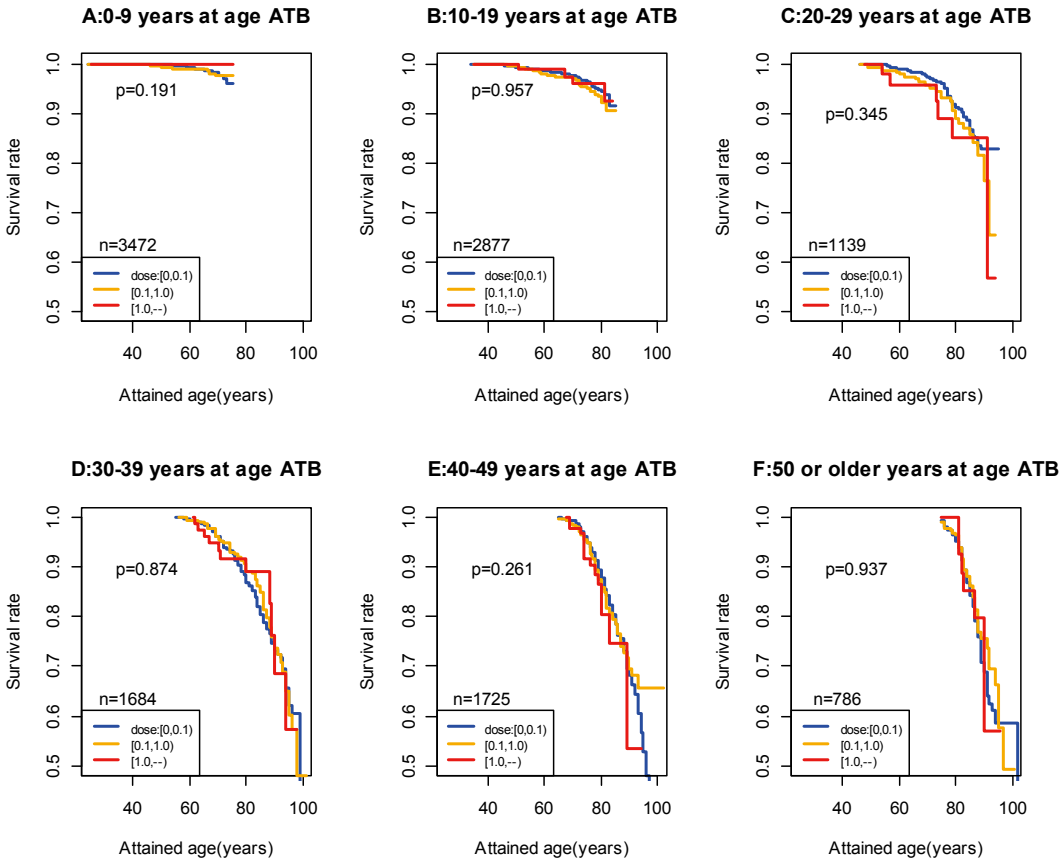

Fig. 2.1. Kaplan-Meier survival curves by initial radiation dose for males stratified by age ATB in 10-year age group. The p-value indicates the statistical significance of the difference between the low-dose group (< 0.1 Sv) and the high-dose group (>1.0 Sv).

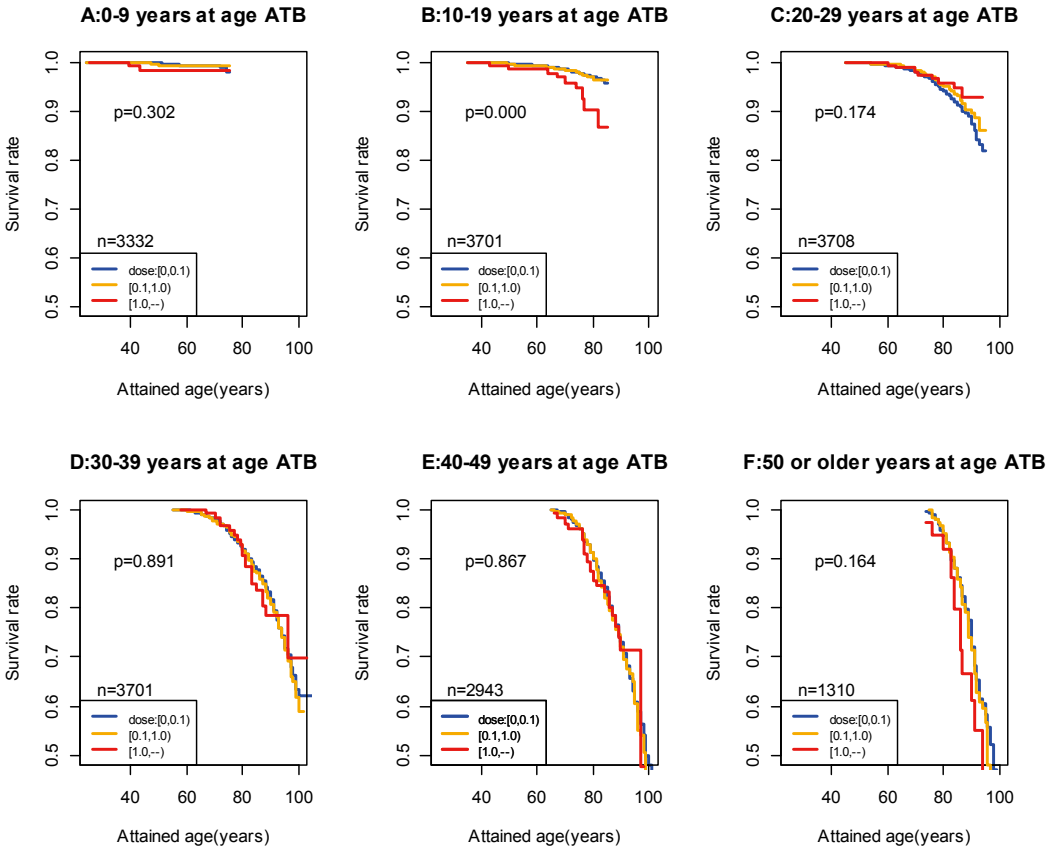

Fig. 2.2. Kaplan-Meier survival curves by initial radiation dose for females stratified by age ATB in 10-year age group. The p-value indicates the statistical significance of the difference between the low-dose group (< 0.1 Sv) and the high-dose group (>1.0 Sv).

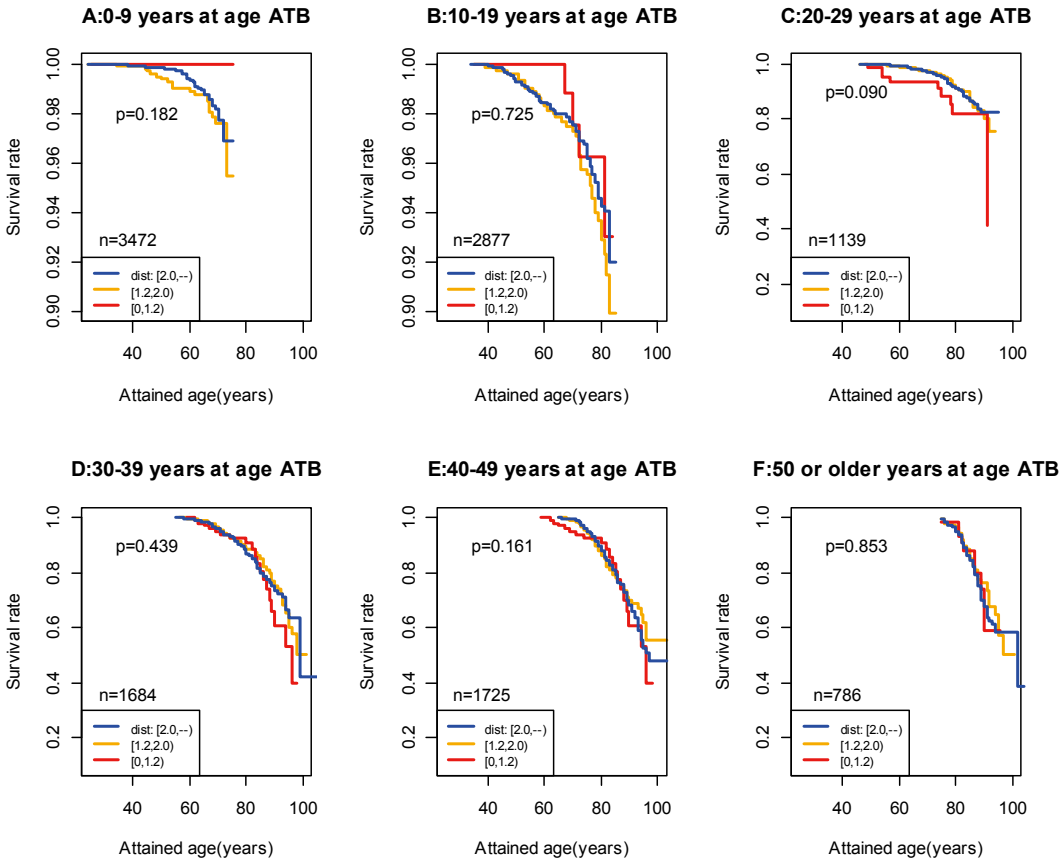

Fig. 2.3. Kaplan-Meier survival curves by exposure-distance for males stratified by age ATB in 10-year age group. The p-value indicates the statistical significance of the difference between the short-distance group (<1.2 km) and the long-distance group (>2.0 km).

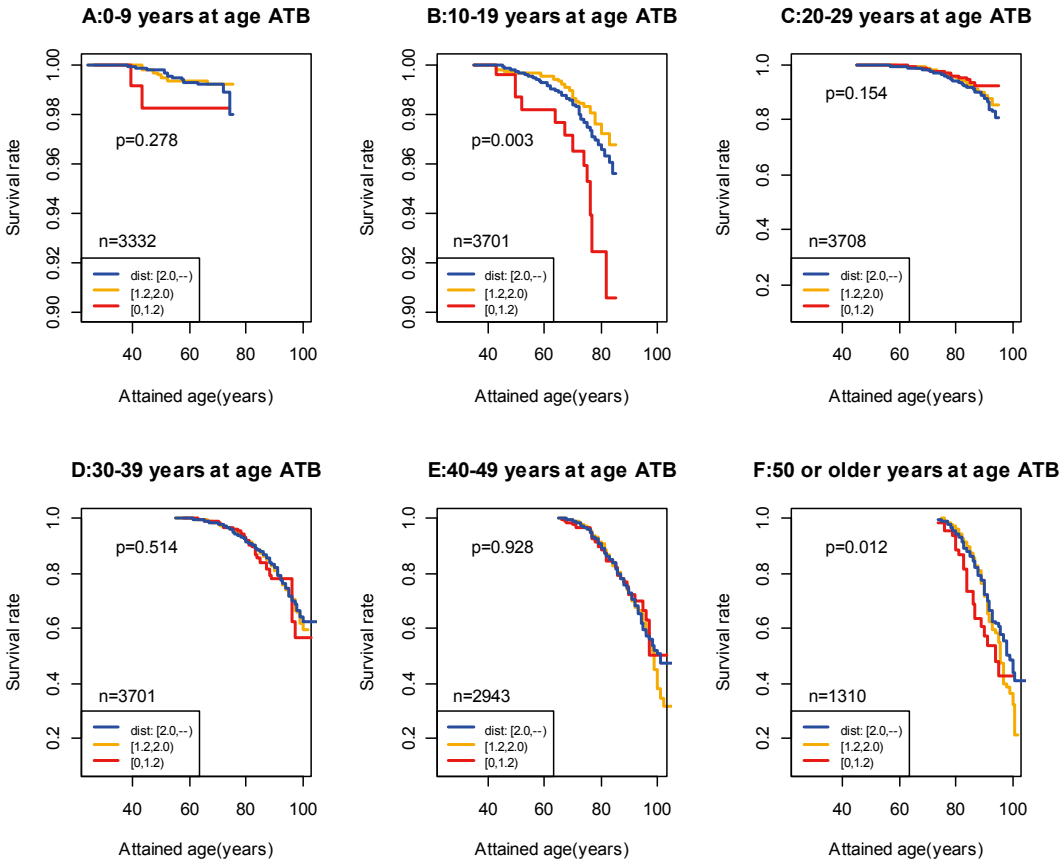

Fig. 2.4. Kaplan-Meier survival curves by exposure-distance for females stratified by age ATB in 10-year age group. The p-value indicates the statistical significance of the difference between the short-distance group (<1.2 km) and long-distance group (>2.0 km).

Table 2.1. Sex- and age ATB-specific estimated coefficients (β) of the dose effect

(Males)

age ATB	coef.	s.e.	95%lower	95%upper	p-value
[0, 10)	-0.513	0.558	-1.606	0.579	0.36
[10, 20)	0.033	0.212	-0.382	0.449	0.87
[20, 30)	0.224	0.173	-0.114	0.562	0.19
[30, 80)	0.107	0.743	-0.038	0.252	0.15

(Females)

age ATB	coef.	s.e.	95%lower	95%upper	p-value
[0, 10)	0.330	0.324	-0.305	0.964	0.31
[10, 20)	0.410**	0.115	0.185	0.636	<0.001
[20, 30)	-0.115	0.142	-0.394	0.163	0.42
[30, 80)	0.031	0.058	-0.083	0.145	0.60

**: $p < 0.01$

Table 2.2. Sex- and age ATB-specific estimated coefficients (β) of the distance effect

(Males)

age ATB	coef.	s.e.	95%lower	95%upper	p-value
[0, 10)	-29.343	63.225	-153.262	94.577	0.64
[10, 20)	0.275	0.322	-0.357	0.906	0.39
[20, 30)	0.437	0.361	-0.270	1.144	0.23
[30, 80)	0.263	0.158	-0.047	0.572	0.10

(Females)

age ATB	coef.	s.e.	95%lower	95%upper	p-value
[0, 10)	1.139	0.708	-0.248	2.526	0.11
[10, 20)	1.071**	0.279	0.524	1.617	< 0.001
[20, 30)	-0.320	0.209	-0.730	0.089	0.12
[30, 80)	0.086	0.098	-0.106	0.278	0.38

**: $p < 0.01$

models for men, no significant contribution was detected for either initial dose or exposure distance. In women, a significantly high contribution was detected for the influence of initial dose (hazard ratio: 1.51, $p < 0.001$) only if their age ATB was between 10 and 19.

Table 3 shows estimated values of the parameter μ of the distance function model. Table 4 shows AIC[1)] for the initial dose model, the exposure-distance model, and the null model, applied to each stratum of age ATB. In men whose age ATB was under 10, the distance-function model had the minimum AIC. In women whose age ATB was between 10 and 19, the initial-dose model had the minimum AIC. The null model had the minimum AIC in other age-ATB strata.

DISCUSSION

The principal risk factors for cerebrovascular disease include aging and arteriosclerosis[15)]. The female sex hormone estrogen works to inhibit arteriosclerosis by suppressing increases in LDL cholesterol and raising HDL cholesterol level[19,29)]. Accordingly, during the period of life with plentiful secretion of female sex hormone, women have a significantly lower risk of cardiovascular disease than men[10)]. Thus, sex is a confounding factor influencing risk of mortality from cerebrovascular disease. It is also assumed that many *hibakusha* inhaled fine radioactive particulate material after the explosion, even if they were inside large buildings or in a basement at the time of the explosion, and that behavioral patterns just after the bombing were largely dependent on sex and age ATB, which leads to the deduction that "dose" due to residual radiation must depend on sex and age ATB. Due to the reasons described above, we stratified the *hibakusha's* cohort data according to sex and age ATB, and analyzed the effect of exposure to radiation, separately. Residual radiation comprises neutron activated radiation in soil and other materials as well as fallout from the nuclear explosion. Radioactive contaminants were generated in the neighborhood of the hypocenter and were dispersed with the bomb blast. Because the dose from residual radiation exposure should not be greatly influenced by the degree of shelter from direct radiation at the time of the explosion, the present study also utilized an analysis based on distance from the hypocenter as an alternate index of exposure. The exposure-distance model is thought to be a good alternative to the initial-dose model, in which effects of shielding from direct Atomic bomb radiation were not taken into account. In the exposure-distance model, we assumed that the effect of an exposure-distance of 2.0 km or more was 0.

In our Cox regression analyses, since the follow-up period for this study covers 41 years from 1970

Table 3. Sex- and age ATB-specific estimated parameter (μ) of the exposure-distance function

age ATB	Males	Females
[0, 10)	1.24	1.19
[10, 20)	2.00	1.23
[20, 30)	1.67	2.00
[30, 80)	1.43	1.68

Table 4. AIC of candidate models and differences in AIC between initial-dose and other models

(Males)

age ATB	dose[(a)]	dist[(b)]	null[(c)]	Δdist†	Δnull††
[0, 10)	680.3	678.9	679.5	-1.4	-0.8
[10, 20)	1755.5	1756.8	1753.5	1.3	-2.0
[20, 30)	1315.8	1317.9	1315.2	2.1	-0.6
[30, 80)	9921.6	9922.9	9921.6	1.3	-0.1

(Females)

age ATB	dose[(a)]	dist[(b)]	null[(c)]	Δdist†	Δnull††
[0, 10)	354.2	355.3	353.0	1.1	-1.2
[10, 20)	1624.8	1625.1	1631.2	0.3	6.4
[20, 30)	4222.9	4223.2	4221.6	0.3	-1.3
[30, 80)	23467.2	23468.7	23465.4	1.5	-1.8

†***(b)*–*(a)***: The difference in AIC between the initial-dose model and distance-function model

††***(c)*–*(a)***: The difference in AIC between initial-dose model and null model

※ Initial radiation dose model has one, distance-function model has two, null model has no unknown parameters.

and the age-adjusted mortality rate of cerebrovascular disease among the general population in Japan decreased dramatically during this period[7], we took these changes of background risk into account in our analysis by allowing for effects of calendar year. Kaplan-Meier curves with log-rank tests and Cox regression analyses results both suggested that, aside from women whose ages were 10 to 19 ATB, the effect of neither initial dose nor exposure distance was statistically significant, which suggests that radiation exposure has no direct impact on mortality due to cerebrovascular disease. In contrast, significant effects of both initial dose and exposure distance were detected among women whose age ATB was between 10 and 19. It is unlikely that radiation exposure had an effect only in this group. The physical and mental damage suffered as a result of the bombings, such as loss of family members and the subsequent deterioration of lifestyle and environment, cannot be ignored. Cerebrovascular diseases are considered to be a type of lifestyle disease, and their onset and progression is influenced by lifestyle factors such as exercise habits, smoking, and drinking[5,15]. For women whose age ATB was in the teens, the estimated effects of initial dose and exposure distance were significant. However, the numbers of female subjects were 2, 55 and 193 for exposure distance < 0.8 km, 0.8 - 1.0 km and 1.0 - 1.2 km. Therefore, it might be accidental that the estimated effect of exposure distance was significant because of the small sample size. There might also exist some effect of radiation exposure limited to women in their teens at the time of exposure; however, the background mechanisms remain unclear, necessitating further study.

Deaths from other diseases were treated as censored in the present study. Strictly speaking, it would be appropriate for these to be treated as competing risks[9]. We may disregard the effects of exposure due to the atomic bomb such as death from explosion including acute radiation symptoms and those due to malignant tumors, etc., which might reduce the observed excess mortality from cerebrovascular disease through the competing risk effect. Ozasa et al[18] reported that the sex-averaged excess relative risk per Gy was 0.42 for all solid cancer at age 70 years-old after exposure at age 30 and that female susceptibility was about twice as high as the male one. In particular, heart disease, which shares many of the same risk factors[4], resulted in many deaths where cerebrovascular disease went undetected. Owing to a competing risk with other causes of death, such as that from heart disease, cerebrovascular disease mortality must be underestimated. In this paper, we showed that the impact was understimated for the young or middle-aged because of their low mortality. The same applies for death in the elderly due to solid cancers. In addition, as the present study was commenced in 1970, 25 years after exposure to the Atomic bomb, it is likely that many deaths occurred prior to the start of the cohort study not only from the effects of the explosion but also due to acute radiation syndrome. Therefore, people with a relatively high risk of death from radiation-related cerebrovascular disease might have been selectively excluded. As a result, we may have underestimated the excess risk for the effect of exposure to the atomic bomb.

Our study had the same limitation as LSS by Ozasa et al[18] in RERF due to analyses without adjusting for the possible effects of confounding factors, since such information was not available in the large cohort studies. For even if some confounding factors mislead the effect of radiation risk to cerebrovascular disease mortality, the result of this study must be a clue toward clarifying how radiation risk affects cerebrovascular disease.

ACKNOWLEDGMENTS

This study was supported in part by Grants-in-Aid for Scientific Research (A) 24249039, Young Scientists (B) 23790694, and 23700337 from the Japanese Ministry of Education, Culture, Sports, Science and Technology.

We declare that we have no conflict of interest.

(Received November 30, 2015)
(Accepted June 14, 2016)

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Hiroshima J. Med. Sci.
Vol. 65, No. 2, 45~51, June, 2016
HIJM 65−7

High Excess Risk of Heart Disease Mortality among Hiroshima Atomic Bomb Male Survivors Exposed Near the Hypocenter

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ABSTRACT

Heart disease (HD) mortality is the second leading cause of death in Japan. The HD mortality risk among Atomic bomb survivors is slightly positive but shows a statistically significant dose-response relationship with initial radiation dose, as reported by the Radiation Effects Research Foundation. In that report, dosimetry was based on initial radiation only, with the effect of indirect radiation dose not taken into consideration. The atomic bomb radiation, however, consisted of both initial and residual radiation. We reevaluated the dose-response relationship for HD mortality using exposure distance (ground distance between the location where exposed and the hypocenter) as a surrogate indicator of radiation dose. At Hiroshima University, a cohort study has been conducted with Hiroshima Atomic Bomb Survivors (ABS) since 1970. We selected 29605 subjects from the ABS who were exposed at 3.5 km or less from the hypocenter and alive on January 1, 1970. These subjects, referred to as "Hiroshima *hibakusha*" in this paper, were followed until December 31, 2010. We stratified the cohort data with respect to sex and age at the time of bombing (ATB) into 10-year age groups. For each stratum, by applying an extended Cox regression model with time-dependent covariates, we analyzed the risk of HD mortality using either initial radiation dose or exposure distance as an explanatory variable. The results indicate a high excess risk in males and older age ATB females who were exposed near the hypocenter. This difference may be explained by the effect of female sex hormone on the circulatory system among young age ATB females. Some unknown risk factor related to exposure distance was also implicated in the elevated risk of HD among the Hiroshima *hibakusha*, especially in males. This necessitates further study.

Key words: *Atomic-bomb survivors, Dose-response relationship, Exposure distance, Heart disease mortality*

Heart disease (HD) was ranked the second leading cause of death in Japan after cancer as of 2010[13]. Among cohort studies of the mortality risk of HD in the general population, the Framingham Heart Study and the Hisayama Study are well known. Sytkowski et al showed that morbidity and mortality among females were comparatively low during the follow-up in Framingham residents from 1950 to 1989[26]. Ueda showed that the rate of development of symptoms of ischemic heart disease rose with increasing age, with rates in males higher than in females in a follow-up study of Hisayama residents from 1961 to 1984[29].

HD mortality risks among atomic bomb survivors have been described in several reports published by the Radiation Effects Research Foundation[19,24]. In those studies, dosimetry was based on initial radiation only[7,14], with the effect of indirect radiation not taken into consideration. In fact, it is known that atomic bomb radiation exposure comprises two types: direct exposure (gamma-ray as well as prompt and delayed neutron radiations) from the initial explosion and indirect exposure from residual radiation comprising neutron activated radiation in soil and other materials as well as fallout from the nuclear explosion. The initial dose is determined by the ground distance from the hypocenter to the victim's location at the time of exposure (exposure distance) and shielding conditions assessed from information provided by ex-

posed persons, such as being in buildings or other structures at the time of the bombing. Lauk et al reported serious heart disease induced by X-ray doses of 10 Gy or more in rats[11]. However, it is becoming clear that the increase in several health risks in *hibakusha* cannot be explained by the effect of initial dose alone[23]. Recently, Kerr et al reported that the health risk among atomic bomb survivors in Hiroshima and Nagasaki for residual radiation from neutron-activated radionuclides in the airburst's dust stem and pedestal and in surface soil might not be negligible[9]. Tonda et al showed that the geographical distribution of solid cancer mortality risk among Hiroshima *hibakusha* is not circular but asymmetric around the hypocenter[28]. Tonda et al suggested the impact of indirect exposure as a factor in the increased leukemia risk for those who entered Hiroshima City on 6 August 1945[27]. Otani et al reported that the risk of mortality from malignant neoplasms (excluding leukemia) was significantly higher for those who entered Hiroshima City on 6 August 1945[18]. Ohtaki et al analyzed solid cancer mortality among the Hiroshima atomic bomb survivors through Cox regression with time-dependent covariates[2,3] using a model with exposure distance as well as initial radiation dose as explanatory variables[15]. In that analysis, the exposure-distance function as a surrogate for non-initial radiation dose was defined by the following formula with the threshold parameter μ:

$$D^*(r \mid \mu) = \begin{cases} \dfrac{\mu - r}{\mu - 1}, & r < \mu, \\ 0, & r \geq \mu, \end{cases} \qquad (1)$$

Here r denotes the ground distance between the location where exposed to the explosion and the hypocenter in Hiroshima. The exposure-distance model had a better fit than the initial-radiation model to the excess relative risk of solid cancer mortality, and the risk increased only in the neighborhood of the hypocenter (within 1.2 km).

The objective of the present study was to examine whether the sex- and age-ATB-specific HD disease mortality among Hiroshima A-bomb survivors can be explained solely through initial radiation dose or not, and to assist in estimating the HD risk among *hibakusha* precisely. We analyzed the HD mortality with the exposure-distance model using $D^*(r \mid \mu)$ defined by (1) as well as the initial radiation model, and compared their performance.

MATERIALS AND METHODS

Subjects

In Hiroshima University, a cohort study of the Hiroshima Atomic Bomb Survivors (ABS) has been conducted since 1968[12]. We chose for analysis 29605 subjects from the ABS who satisfied the following conditions: (i) alive and recognized as an atomic bomb survivor as of January 1, 1970, (ii) having an estimate of initial radiation dose, and (iii) exposed within 3500 m of the hypocenter. These subjects were followed until December 31, 2010. The endpoint was death from HD. Death information including cause of death was obtained from the Vital Statistics Death Schedules, which are based on official death certificates. Tables 1.1 and 1.2 show the sex-specific numbers of subjects categorized by age ATB and exposure distance, respectively.

Radiation dosimetry

To quantify the effect of initial radiation dose in Sieverts (Sv) on the human body, we used the absorbed dose in red bone marrow from neutrons and gamma rays in Gray (Gy) estimated using the Atomic Bomb Survivor 1993 Dose (ABS93D)[7]. The radiation dose calculated with ABS93D is based on the initial radiation only, as is DS86, and ignores residual radiation[21]. The extent of overlap between survivors in the ABS and the LSS was examined by Hayakawa et al in which it was shown that dose estimates of the ABS were close to those of the LSS among the overlapping subjects[6]. However, it has not been investigated the consistency of the ABS93D and DS02, yet.

Table 1.1. Numbers of subjects, events, and censored cases by sex and age categories at time of exposure (ATB)

(Males)

age ATB	number of subjects	number of HD deaths	number of censored cases†† (surviving cases††)
[0, 10)	3401	78	3323 (1985)
[10, 20)	2835	150	2685 (1283)
[20, 30)	1122	141	981 (154)
[30, 40)	1654	256	1398 (25)
[40, 50)	1660	313	1347 (0)
[50, 60)	640	119	521 (0)
[60, 80)	66	16	50 (0)
total	11378	1073	10305 (3447)

(Females)

age ATB	number of subjects	number of HD deaths	number of censored cases†† (surviving cases††)
[0, 10)	3208	30	3178 (2180)
[10, 20)	3657	129	3528 (2306)
[20, 30)	3669	363	3306 (1352)
[30, 40)	3656	728	2928 (190)
[40, 50)	2870	622	2248 (4)
[50, 60)	1011	240	771 (0)
[60, 80)	156	33	123 (0)
total	18227	2145	16082 (6032)

†Numbers of persons who migrated out of Hiroshima prefecture or who died from other causes or who were alive as of 31 Dec 2010.

††Number of persons alive as of 31 Dec 2010.

Table 1.2. Numbers of subjects, events, and censored cases by sex and exposure distance

(Males)

distance (km)	number of subjects	number of HD deaths	number of censored cases†† (surviving cases††)
[0.0, 0.8)	4	1	3 (0)
[0.8, 1.0)	153	26	127 (20)
[1.0, 1.2)	449	40	409 (110)
[1.2, 1.4)	932	88	844 (249)
[1.4, 1.6)	1232	109	1123 (385)
[1.6, 1.8)	950	112	838 (266)
[1.8, 2.0)	667	64	603 (196)
[2.0, 2.5)	1054	88	966 (301)
[2.5, 3.0)	3866	359	3507 (1274)
[3.0, 3.5)	2071	186	1885 (646)
total	11378	1073	10305 (3447)

(Females)

distance (km)	number of subjects	number of HD deaths	number of censored cases†† (surviving cases††)
[0.0, 0.8)	7	1	6 (3)
[0.8, 1.0)	261	28	233 (48)
[1.0, 1.2)	891	97	794 (231)
[1.2, 1.4)	1737	249	1488 (498)
[1.4, 1.6)	2398	298	2100 (698)
[1.6, 1.8)	1717	218	1499 (556)
[1.8, 2.0)	1078	133	945 (345)
[2.0, 2.5)	1343	162	1181 (465)
[2.5, 3.0)	5369	610	4759 (1996)
[3.0, 3.5]	3426	349	3077 (1192)
total	18227	2145	16082 (6032)

†Numbers of persons who migrated out of Hiroshima prefecture or who died from other causes or who were alive as of 31 Dec 2010.

††Number of persons alive as of 31 Dec 2010.

Table 1.3. Numbers of subjects, events, and censored cases by sex and dose categories

(Males)

dose(Sv)	number of subjects	number of HD deaths	number of censored cases†† (surviving cases††)
[0.00, 0.01)	6739	612	6127 (2158)
[0.01, 0.05)	664	63	601 (158)
[0.05, 0.1)	719	76	643 (225)
[0.1, 0.2)	945	97	848 (275)
[0.2, 0.4)	1005	95	910 (289)
[0.4, 0.6)	386	36	350 (121)
[0.6, 0.8)	247	22	225 (65)
[0.8, 1.0)	182	19	163 (38)
[1.0, 1.5)	190	17	173 (56)
[1.5, 2.0)	109	7	102 (25)
[2.0, 6.0)	192	29	163 (37)
total	11378	1073	10305 (3447)

(Females)

dose(Sv)	number of subjects	number of HD deaths	number of censored cases†† (surviving cases††)
[0.00, 0.01)	9687	1063	8624 (3519)
[0.01, 0.05)	1148	152	996 (352)
[0.05, 0.1)	1313	152	1161 (417)
[0.1, 0.2)	1799	256	1543 (527)
[0.2, 0.4)	1894	228	1666 (540)
[0.4, 0.6)	753	109	644 (242)
[0.6, 0.8)	437	45	392 (138)
[0.8, 1.0)	387	64	323 (85)
[1.0, 1.5)	332	33	299 (94)
[1.5, 2.0)	166	10	156 (52)
[2.0, 6.0)	311	33	278 (66)
total	18227	2145	16082 (6032)

†Numbers of persons who migrated out of Hiroshima prefecture or who died from other causes or who were alive as of 31 Dec 2010.

††Number of persons alive as of 31 Dec 2010.

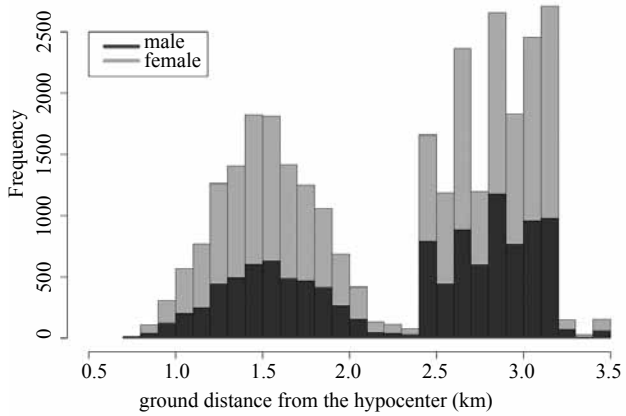

Fig. 1. Histogram of sex-specific numbers of subjects by exposure distance (km) from the hypocenter.

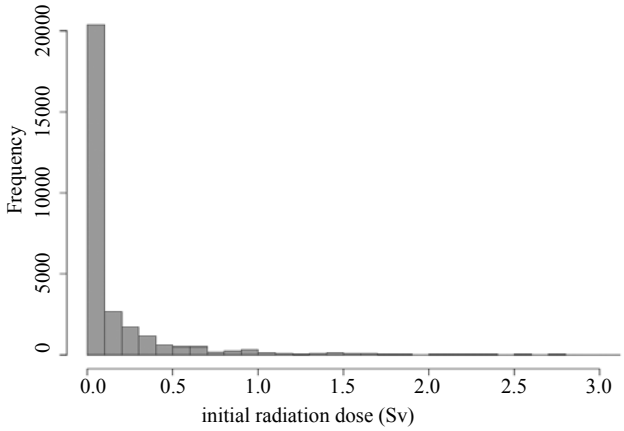

Fig. 2. Histogram of numbers of subjects in the cohort study by initial radiation dose (Sv)

Table 1.3 shows the sex-specific numbers of subjects by categories of initial radiation dose. Histograms of exposure distance and of initial radiation dose are given in Fig. 1 and Fig. 2, respectively. Since we have detailed information about A-bomb survivors' shielding conditions within an exposure distance of 2.0 km, a sufficient number of such subjects were obtained. In cases of an exposure distance beyond 2.5 km, we can assume that their initial radiation doses were zero. However, in the case of an exposure distance between 2.0 km to 2.5 km, only a limited number of samples were available because of the difficulty of estimating the ini-

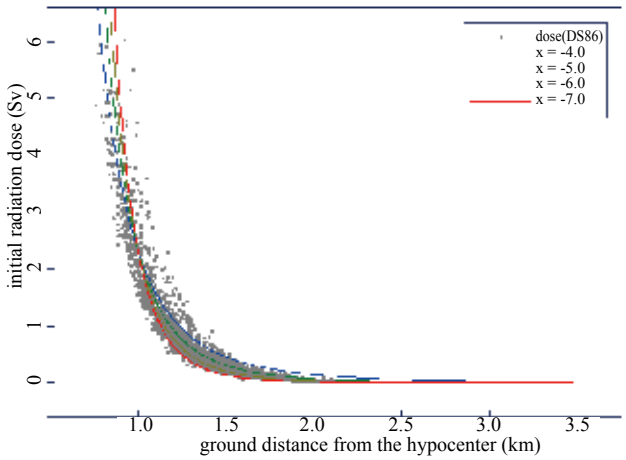

Fig. 3. Scatterplot of individual initial radiation dose versus exposure distance

tial radiation dose. Figure 3 shows the relationship between subjects' initial radiation dose and exposure distance, with fitted curves $f(r \mid x) = 2.3 \times r^{x}$ based on a power function of exposure distance. It is shown that plots for many subjects are located briefly around the dose-distance curves of $f(r \mid -4)$ to $f(r \mid -7)$.

Statistical analysis

Based on epidemiological evidence that HD mortality risk increases exponentially with age[5,17], we assumed that the hazard function of attained age t for a person exposed to an initial radiation dose D at age a can be expressed as

$$h(t \mid D,a) = \exp\big(g(t,t-a+1945)\big) \cdot \exp\big(\delta + \beta_a D\big),$$

where β_a is the regression coefficient for the effect of the initial dose among Hiroshima *hibakusha* who were age a at the time of exposure, while $g(t, y)$ is a logarithmic function of attained age (t) and calendar year (y) for HD mortality risk in all of Japan during the period 1970 to 2010, which is specified approximately by a sextic polynomial equation of t and y. The parameter δ expresses the logarithm of the background relative mortality from HD for Hiroshima *hibakusha* compared with the whole of Japan. The cohort data were stratified by sex and age ATB into eight strata -- 0-9 years (male/female), 10-19 years (male/female), 20-29 years (male/female), and 30 years and over (male/female) -- and the unknown coefficient parameter (β_a) for each stratum was estimated by applying Cox regression analysis with time-dependent covariates[2,3]. To collect information on weight and factors such as smoking and alcohol consumption was not feasible because it would require enormous expense. We noted RERF's report showing that the influence of non-radiation risk factors such as excessive weight, smoking, alcohol consumption and diabetes were significantly low among atomic bomb survivors in Hiroshima and Nagasaki[24], and also that LSS did not deal with these factors in the risk analysis. We also analyzed in a similar way the exposure-distance dependency based on the function $D^{*}(r \mid \mu)$ of exposure distance r defined by (1), in which the threshold parameter μ was estimated by applying the optim function[22]. The model using the exposure distance is expressed as follows: $h(t \mid D^{*},a) = \exp\big(g(t,t-a+1945)\big) \cdot \exp\big(\delta + \beta_a D^{*}\big)$. We used the freeware R (version 3.0.0) to implement the numerical-analyses.

RESULTS

Results of the Cox regression analyses of HD death are shown in Tables 2, 3, and 4. Table 2 shows AIC[1] expressing the goodness of fits of the initial dose model (dose), the exposure-distance model (dist) and a model (null) showing neither dose nor the exposure-distance variables by sex and ATB group. Table 2 also shows the difference in AIC of each model compared to the initial radiation dose model. In males, the null model attained the minimum AIC for ATB 10-19, whereas the exposure-distance model had the minimum AIC for the other ATB groups. In females, the exposure-distance model attained the minimum AIC for ATB 30 and over, whereas the null model had the minimum AIC for the other ATB groups. In no case was the initial-dose model optimal in terms of minimum AIC. Table 3.1 shows the estimated coefficients of the dose effect and its 95% CI bounds for each sex-ATB group, and Table 3.2 shows those of the exposure-distance dependency. From these tables, it was found that the effect of initial dose in males was detected only for the 0-9 age ATB group, whereas large effects of exposure distance were estimated for all age ATB groups except the 10-19 group. On the other hand, in females, the effects of neither initial radiation dose nor exposure distance were significant for any age ATB group except 30 and over. Table 4 shows the optimized values of the threshold parameters in the exposure-distance model. It suggests that in

Table 2. AIC values of candidate models and difference in AIC between the initial-dose and other models. The number of parameters in the dose model, distance model and null model are 1, 2 and 0, respectively.

	age ATB	**dose**(*a*)	**dist**(*b*)	**null**(*c*)	**Δdist**†	**Δnull**††
Males	[0, 10)	1186.82	1185.98	1189.19	-0.84	3.21
	[10, 20)	2236.90	2237.50	2235.49	0.60	-2.01
	[20, 30)	1729.70	1729.29	1730.32	-0.41	1.03
	[30 over)	9909.86	9908.56	9909.90	-1.30	1.33
Females	[0, 10)	461.88	463.91	459.91	2.03	-3.99
	[10, 20)	1997.44	1999.52	1995.54	2.08	-3.98
	[20, 30)	5479.04	5480.42	5478.20	1.38	-2.22
	[30 over)	25559.24	25558.56	25558.98	-0.68	0.42

†(*b*)–(*a*): The difference in AIC between initial-dose model and distance-function model

††(*c*)–(*a*): The difference in AIC between initial-dose model and null model

Table 3.1. Estimated coefficient (β) of the dose effect and its 95% CI

	age ATB	coef.	lower.95	upper.95	p-value
Males	[0, 10)	0.383*	0.079	0.688	0.014
	[10, 20)	0.133	-0.187	0.452	0.416
	[20, 30)	0.262·	-0.026	0.550	0.075
	[30 over)	0.114	-0.036	0.263	0.136
Females	[0, 10)	0.076	-0.695	0.847	0.847
	[10, 20)	0.059	-0.305	0.422	0.752
	[20, 30)	-0.131	-0.380	0.119	0.305
	[30 over)	0.072	-0.032	0.176	0.175

*: $0.01 \leqq p < 0.05$, ·: $0.05 \leqq p < 0.1$

Table 3.2. Estimated coefficient (β) of the exposure-distance dependency and its 95% CI

	age ATB	coef.	lower.95	upper.95	p-value
Males	[0, 10)	0.698**	0.318	1.077	0.000
	[10, 20)	0.429	-0.090	0.948	0.105
	[20, 30)	0.625*	0.100	1.149	0.020
	[30 over)	0.236**	0.058	0.413	0.009
Females	[0, 10)	0.066	-1.694	1.826	0.941
	[10, 20)	0.053	-0.727	0.832	0.894
	[20, 30)	-0.308	-0.773	0.158	0.195
	[30 over)	0.225*	0.021	0.430	0.031

**: $p < 0.01$, *: $0.01 \leqq p < 0.05$

Table 4. Estimated threshold parameter (μ) and its 95% CI

	age ATB	estimate	lower.95	upper.95
Males	[0, 10)	1.05	1.00	1.11
	[10, 20)	1.09	–†	–
	[20, 30)	2.00	1.76	2.24
	[30 over)	1.06	1.01	1.10
Females	[0, 10)	1.50	–	–
	[10, 20)	1.48	–	–
	[20, 30)	1.56	–	–
	[30 over)	1.47	1.37	1.57

† The finite confidence bound was not available because of the non-statistical significance of the corresponding effect of exposure-distance effect. (See. Table 3.2)

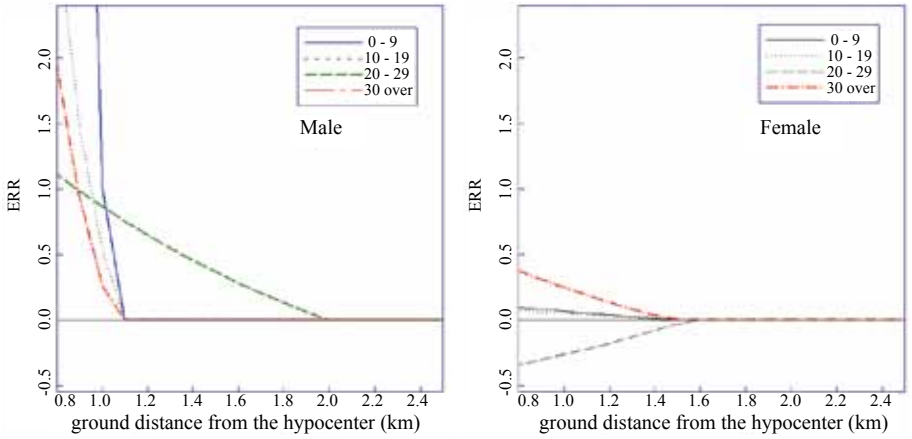

Fig. 4. Fitted exposure-distance dependencies of excess relative risk by sex and age ATB. The colored curves show statistically significant trends.

males the risk became high within about 1.1 km from the hypocenter for all ATB groups except 20-29. Figure 4 shows the fitted exposure-distance dependency of the excess relative risk by sex and age ATB. It indicates that the estimated risk is high for males in the case of being exposed at or near the hypocenter, but no corresponding excess risk can be seen for females.

We also considered fitting a linear-quadratic model of initial radiation dose to our analysis, but the goodness of the fit deteriorated compared with that of the linear initial radiation dose model described above.

DISCUSSION

Several studies have reported the effects of radiation on HD mortality. Ivanov et al analyzed data on the Chernobyl emergency workers and showed that the excess relative risk of ischemic heart disease was 0.41 per Gy[8]. Shimizu et al reported that the sex-averaged excess relative risk per Gy of HD mortality was 14% among atomic bomb survivors in Hiroshima and Nagasaki for the period 1950-2003, and further showed that the initial radiation dose effect was not significant in the low-dose region below 0.5 Gy[24].

We analyzed the relationship between risk of HD mortality and initial radiation dose as well as the exposure distance. It is assumed that many *hibakusha* inhaled fine radioactive particulate material after the explosion, even if they were inside large buildings or in a basement at the time of the explosion, and that behavioral patterns just after the bombing were largely dependent on sex and age ATB. This suggests that dose due to residual radiation should depend on sex and age ATB. Due to these reasons, we stratified the *hiba-*

kusha cohort data according to sex and age ATB to analyze the effect of exposure to radiation. Table 1.2 shows that the number of deaths within 1.2 km was 193 (6.0%) and that of deaths within 2.0 km was 1464 (45.5%). Table 1.3 shows that the number of deaths in under 0.1 Sv was 2118 (65.8%).

We found a large sex difference in the estimated dose-response relationship: significant excess mortality risk of HD was detected for males who were exposed at ages younger than 10 years whereas almost no excess risk was seen for females. A clearer sex difference was found in the relationship between HD mortality risk and exposure distance, which suggests that the dose due to residual radiation was higher for males than females. Although the estimated effects of initial radiation dose and exposure distance in our cohort study were not adjusted for recognized risk factors such as excessive weight, hypercholesterolemia, smoking, alcohol consumption, and others, it may be concluded that there were some risk factors for HD mortality unique to male survivors who were exposed at a short distance from the hypocenter. For example, it is possible that the effects of radioactive particulate materials with short half-lives that were generated just after the explosion, such as[28] Al, affected only short-distance survivors[16]. However, following that reasoning it is difficult to explain why a corresponding excess risk was not found among female survivors. For this sex difference it may be that although non-initial radiation was the main exposure factor, the risk was abated by female sex hormones present in young age ATB females that are not present in males or older age ATB females. Cui et al reported that early menopause is associated with an increased risk of mortality from coronary heart disease (CHD), which can be explained by a protective effect of endogenous estrogen on the development of atherosclerosis[4]. As possible factors other than initial radiation dose or radioactive particulate materials, malnutrition or mental stress caused by the collapse of family due to the devastation might have elevated HD mortality risk among the atomic bomb survivors. Kubzansky and Kawachi showed that negative emotions influence the development of CHD[10]. Shirai et al also reported that men with low perceived enjoyment of life had an increased risk of mortality from CHD and other cardiovascular diseases[25]. Young males might therefore be more sensitive to health deterioration from the collapse of family.

One cause of the complex sex and age ATB- dependency in the dose-response relationship may be the limited follow-up period of our study, which started in 1970. We disregard early effects of exposure to the atomic bomb, such as acute death from the blast and acute radiation sickness, as well as late effects due to malignant tumors etc., which might reduce the observed excess mortality from HD through the competing risk effect. Preston et al showed that the corresponding estimated time-averaged excess relative risks at 1 Sv were 9.1, 3.3 and 6.2 for acute lymphoid leukemia, acute myelogenous leukemia, and chronic myelogenous leukemia, respectively[20]. Matsuura et al showed that the relative risk of leukemia at 1 Gy of bone marrow dose was 2.37, and significantly higher risks were observed for all cancers other than leukemia among survivors who survived for 20 years or more after the bombing[12]. Ozasa et al reported that the sex-averaged excess relative risk per Gy was 0.42 for all solid cancer at age 70 after exposure at age 30, and that the sensitivity was about two times higher in females than in males[19]. We need further investigation into sex and age ATB differences in HD mortality among the atomic bomb survivors.

CONCLUSION

We analyzed HD mortality risk among Hiroshima *hibakusha* using Cox regression analysis. The results suggest that initial radiation dose was not the major risk factor. Some unknown risk factor elevated HD mortality among male Hiroshima *hibakusha*, who were exposed near the hypocenter, while the risk was reduced in females who were exposed at young ages ATB.

ACKNOWLEDGEMENTS

This research was supported in part by Grants-in-Aid for Scientific Research (A) 24249039 (2012–2014), and the Joint Usage/Research Center (RIRBM), Hiroshima University.

We declare that we have no conflict of interest.

(Received November 30, 2015)
(Accepted June 14, 2016)

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