Abbreviations and Acronyms
FDG = ^{18}F fluorodeoxyglucose
NSCLC = non–small cell lung cancer
PET = positron emission tomography
RFI = recurrence-free interval

phy, and blood testing, including tumor markers, 1 month after the initial operation, every 3 to 6 months during the first 3 years, and every half year to 1 year thereafter. If any abnormality was found, we performed computed tomographic scans. We did not routinely perform bone scanning and brain examinations for asymptomatic patients. When a lesion suggesting locoregional or distant recurrence was detected, we scrutinized the whole body radiologically.

Thirty of the 592 patients underwent resection of a solitary recurrent lesion. Among them, 2 patients who had recurrence-free intervals (RFIs) of 1 and 4 months, respectively, were eliminated from this study because they possibly had undetectable "missed" M1 disease at the initial operation. Nine of the 28 patients were symptomatic at the time of recurrence detection. All but 1 patient, who had intrapulmonary recurrence, were asymptomatic. The median period from recurrence detection to the second operation was 2.6 months (range, 0.2-24 months). The follow-up protocols were the same before and after recurrence resection. The median follow-up period after recurrence resection was 33 months, ranging from 11 to 128 months.

For the remaining 562 patients, surgical intervention was not indicated because recurrences were multiple, patients were unfit for further surgical intervention, or both. They underwent palliative chemotherapy, radiotherapy, or best supportive care. Patients with multiple recurrences who underwent palliative operations for symptomatic sites were included in this group.

Prognostic Evaluation

We attempted to identify prognostic factors associated with subsequent survival after resection of a solitary recurrent lesion. We evaluated the following factors: clinical characteristics at recurrence (sex, age, carcinoembryonic antigen level, time from initial resection to recurrence detection [RFI], symptoms at the time of recurrence, site of recurrence, and mode of recurrence [locoregional or distant]) and pathologic findings of the primary lung cancer (histology, tumor size, lymph node status, p-stage, and lymphatic and vascular permeations). We defined locoregional recurrence as recurrence within the ipsilateral thorax and distant recurrence as all other recurrences. Each pathologic specimen was reviewed by a board-certified pathologist who was blinded to the clinical outcome. Histology was specified on the basis of the World Health Organization classification for cell types.[12] Pathologic stages were determined on the basis of the TNM classification of the International Union Against Cancer.[13]

Survival Analysis

Survivals were calculated by using the Kaplan-Meier method and were compared with the log-rank test. Zero time was the date of recurrence identification, and the terminal event was defined as death from any cause. An observation was censored at the last follow-up when the patient was alive or lost to follow-up. Factors with a P value of less than .15 were entered into the multivariate analysis by using the Cox proportional hazards stepwise model. All statistical analyses were performed with a software package (JMP, release 5.0; SAS Institute Inc, Cary, NC).

TABLE 1. Clinicopathologic characteristics of 28 patients with NSCLC who underwent resection of a solitary recurrent lesion

Characteristics	Value	No.
Clinical characteristics at recurrence		
Age at recurrence resection (y)	Median	65
	Range	39-73
Sex	Male	17
	Female	11
RFI (mo)	Median	23
	Range	6-82
CEA level (ng/mL)	Median	3.5
	Range	0.5-3286
Recurrent site	Ipsilateral lung	8
	Contralateral lung	8
	Brain	5
	Adrenal grand	2
	Chest wall	1
	Stomach	1
	Skin	1
	Abdominal lymph node	1
	Bone (malar bone)	1
Pathologic characteristics of primary tumor		
Histology	Adenocarcinoma	21
	Squamous cell carcinoma	5
	Adenosquamous carcinoma	1
	Pleomorphic carcinoma	1
Size of primary tumor (cm)	Mean ± SD	4.1 ± 1.7
p-Stage of primary tumor	IA/IB	4/10
	IIA/IIB	2/6
	IIIA/IIIB	4/2
Nodal status of primary tumor	N0/N1/N2	18/7/3

RFI, Recurrence-free interval; *CEA*, carcinoembryonic antigen; *SD*, standard deviation.

Results

Patient Characteristics

Clinicopathologic characteristics of 28 patients who underwent resection of a solitary recurrent lesion are shown in Table 1. There were 17 men and 11 women, with a median age of 65 years (range, 39-73 years) at the time of resection of the recurrent lesion. At the initial operation, 26 of 28 patients underwent lobectomy and systemic mediastinal lymph node dissection. Two patients underwent limited

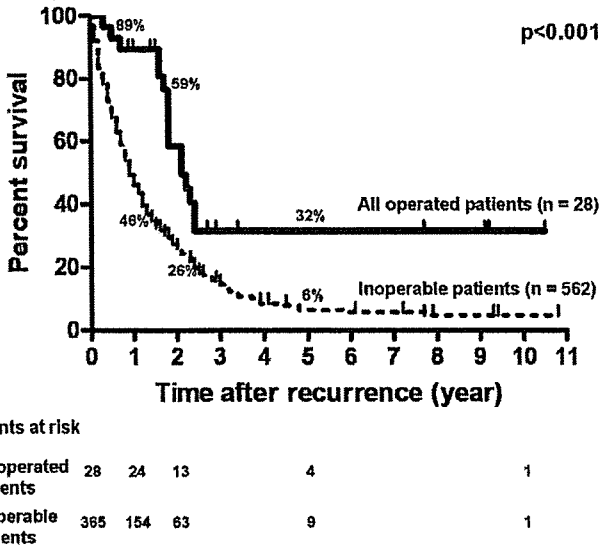

Figure 1. Comparative survival curves among 28 resected patients and 562 patients without resection. The difference in survival probability after recurrence is significant (1-, 2-, and 5-year survivals after recurrence: 89%, 59%, and 32% vs 46%, 26%, and 6%; $P < .001$).

lung resection because of insufficient pulmonary reserve. Neoadjuvant platinum-based chemotherapy was administered to 1 patient because of clinical N2 status. All patients achieved macroscopically complete surgical removal of their primary NSCLC tumor, but the resection margin was pathologically positive in 1 patient. The patient had recurrence in the adrenal gland. RFI was almost 2 years (median, 23 months; range, 6-82 months). The lung (n = 16) was the most frequent site of recurrence. The mode of resection for intrapulmonary recurrences included 3 completion pneumonectomies, 1 lobectomy, and 12 limited resections. Distal gastrectomy was performed for the patient who had gastric recurrence with severe progressive anemia, and open lymph node resection was performed for the patient with pelvic lymph node recurrence. Complete removal of the recurrence was accomplished in all patients. There was no complication after resection of the recurrent lesion. One of 5 patients with brain recurrence received whole-brain irradiation postoperatively. No patients underwent systemic chemotherapy after resection of the recurrent lesion.

Survival and Prognostic Factors After Resection of the Solitary Recurrent Lesion

Figure 1 shows comparative survival curves after recurrence among 28 patients who underwent resection of the solitary recurrent lesion and 562 patients in whom an additional operation was not indicated. Overall 1-, 2-, and 5-year survivals after recurrence were significantly better in patients who underwent resection of a solitary recurrent lesion than in those who did not undergo resection (89%, 59%, and 32% vs 46%, 26%, and 6%; $P < .001$). The median survival times after recurrence were 25 and 11 months, respectively.

Table 2 shows the relationship between survival after resection of the recurrent lesion and the clinicopathologic characteristics of the 28 patients. Multivariate analysis demonstrated that advanced p-stage (stage II-III) of the primary lung cancer was the significant negative prognostic factor associated with survival after recurrence detection (hazard ratio, 6.15; 95% confidential interval, 1.09-30.8; $P = .04$). As shown in Figure 2, the patients with p-stage II or III disease demonstrated survival statistically equivalent to that of patients not undergoing resection after recurrence detection ($P = .11$). In 14 patients with p-stage I disease, 10 and 3 patients survived for more than 2 and 5 years, respectively, after recurrence detection. One with recurrence in the malar bone is surviving for 7 years without a distant failure. In contrast, 3 and 1 of 14 patients with p-stage II or III disease survived for more than 2 and 5 years, respectively, but with a distant failure.

Discussion

Most recurrences after primary NSCLC resection are multiple and disseminated and are usually treated with systemic chemotherapy when patients can tolerate it. Although many studies have shown that systemic chemotherapy prolongs survival in unresectable stage IV NSCLC, there have been no large-scale, randomized prospective trials addressing whether chemotherapy improves survival of patients with recurrence.[14] In an effort to improve long-term tumor control and subsequent survival, attempts have been made to incorporate surgical intervention in selected cases of solitary NSCLC recurrence. Evidence that a solitary recurrent lesion can be effectively treated with surgical intervention exists for malignancies other than lung cancer. For colorectal cancer, melanoma, and thyroid cancer, resection of recurrent lesions can offer prolonged survival.[15-17] For lung cancer, some investigators have reported acceptable survival after resection of the recurrent lesion, but others have contradicted these conclusions. Abrahams and coworkers[4] demonstrated a satisfactory outcome in brain recurrence, with a median survival time of 18 months and a 5-year survival rate of 28.9%. In contrast, Saitoh and associates[2] conducted 24 brain resections, with a 5-year survival rate of only 8.3%. Prognostic factors for survival after resection of the recurrent lesion have not been clarified.

Although our patient population was heterogeneous, with a variety of recurrence sites, the overall survival after resection of the recurrent lesion was acceptable by current standards, with a median survival time of 25 months and a 5-year survival rate of 32%. The patients with a solitary NSCLC recurrence arising from an advanced primary tumor

TABLE 2. Relationship between patient clinicopathologic characteristics and survival after resection of a solitary recurrent lesion

	Factors	No.	MST (mo)	2-y survival (%)	5-y survival (%)	Univariate analysis, *P* value	Multivariate analysis, *P* value
Age at recurrence resection (y)	≥65	14	22	43	26	.41	—
	<65	14	27	75	38		
Sex	Male	17	26	53	31	.48	—
	Female	11	28	67	33		
RFI (y)	≥2	13	26	68	45	.25	—
	<2	15	27	51	22		
CEA level at recurrence (ng/mL)	>5	9	22	30	Not reached	.13	.80
	≤5	19	30	70	38		
Symptoms at recurrence	+	9	26	53	18	.33	—
	−	19	28	60	36		
Site of recurrence	Intrapulmonary	16	30	71	40	.15	.50
	Extrapulmonary	12	22	42	21		
Mode of recurrence	Locoregional	10	26	57	46	.41	—
	Distant	18	27	61	23		
Histology	Ad	21	26	55	31	.73	—
	Non-Ad	7	30	67	33		
Size of primary tumor (cm)	≥4	14	26	51	17	.18	—
	<4	14	28	66	47		
Nodal status of primary tumor	N0 (N−)	18	30	67	40	.08	.40
	N1/N2 (N+)	10	22	41	14		
p-Stage of primary tumor	I	14	—	76	51	.0045	.04
	II/III	14	19	10	10		
Ly in primary tumor	+	10	21	44	Not reached	.32	—
	−	18	27	67	33		
V in primary tumor	+	17	27	57	29	.97	—
	−	11	26	61	36		

MST, Median survival time; *RFI*, recurrence-free interval; *CEA*, carcinoembryonic antigen; *Ad*, adenocarcinoma; *Ly*, lymphatic permeation; *V*, vascular permeation.

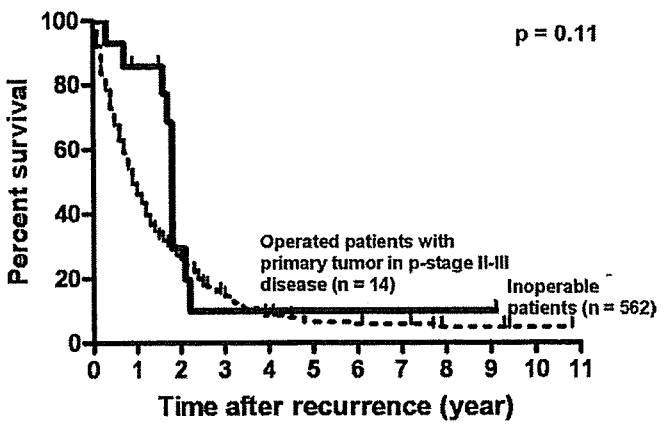

Figure 2. Comparative survival curves among 14 resected patients with p-stage II or III primary non–small cell lung cancer and 562 patients without resection. There is no significant difference in survival probability after recurrence (P = .11).

with p-stage II or III disease, however, had a poor outcome equivalent to that seen in patients with recurrent NSCLC in whom surgical intervention was not indicated. This suggests that advanced stage (ie, II or III) in the primary tumor is a contraindication for surgical intervention in patients with a solitary recurrence. Consistent with our result, Yoshino and coworkers[18] described a strong relationship between pathologic stage and clinical courses after recurrence in patients with NSCLC. They reported that the mean postrecurrent survival time was 590 days in pathologic stage I disease, 381 days in stage II disease, 257 days in stage IIIA disease, and 180 days in stage IIIB disease, with a significant difference being observed between stages I and IIIA (P = .0215). In patients with advanced and biologically aggressive NSCLC, a solitary recurrence might be just the beginning of progressive-disseminated disease.

In our series patients who underwent resection of the intrapulmonary recurrent lesion showed slightly but not significantly better survival than the extrapulmonary recurrence group (P = .15). This might be because some in-

trapulmonary lesions were actually metachronous second primary lung cancers. We can expect better prognosis for metachronous lung cancer compared with intrapulmonary recurrence.[19,20] It can often be hard to discriminate a solitary pulmonary recurrence from a metachronous second primary lung cancer if the 2 lesions are of the same histologic type.[21] Therefore aggressive surgical resection for an intrapulmonary lesion might be justified for patients with adequate pulmonary reserve, regardless of the primary tumor pathology.

Although we did not perform positron emission tomography (PET) with ^{18}F fluorodeoxyglucose (FDG) for the patients in this study, FDG-PET has been reported to be a helpful adjunct in screening for distant metastases but not for brain metastases.[22] Several investigators have reported that FDG-PET could detect unexpected metastatic lesions in 10% to 20% of patients with newly diagnosed NSCLC.[23,24] PET imaging might also be helpful in avoiding surgical intervention in patients who have multiple recurrent lesions.[25] However, it is well known that FDG is not tumor specific and is also taken up in benign lseions.[22] Lardinois and colleagues[26] have reported that 46% of solitary extrapulmonary lesions detected by means of integrated PET and computed tomography were unrelated to lung cancer metastases. Further studies will be needed to clarify whether PET imaging is useful in identifying more clearly the population that benefits from additional surgical intervention and in prolonging subsequent survival.

The limitation of the current study is that the number of enrolled patients, especially in the surgical resection group, was obviously small. Therefore a multi-institutional study would be required to confirm our findings.

In conclusion, long-term survival can be achieved by means of resection of a solitary recurrent lesion in highly selected patients. However, surgical resection might be contraindicated if the primary NSCLC stage is II or III, especially when the recurrent lesion is extrapulmonary.

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Once-Weekly Epoetin-Beta Improves Hemoglobin Levels in Cancer Patients with Chemotherapy-Induced Anemia: A Randomized, Double-Blind, Dose-Finding Study

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Objective: To determine a recommended dose of once-weekly epoetin-beta administration for anemic cancer patients receiving myelosuppressive chemotherapy, we conducted a multicenter, randomized, double-blind trial.

Methods: A total of 86 patients with malignant lymphoma or lung cancer who received chemotherapy containing platinum, taxanes or anthracyclines were enrolled in the study. Patients were randomly assigned into groups that received three dose levels of epoetin-beta (9000, 18 000 or 36 000 IU) administered subcutaneously once a week for 12 weeks. The primary endpoint was change in hemoglobin, while the secondary endpoints were quality of life (QOL) assessed by Functional Assessment of Cancer Therapy-Anemia (FACT-An) questionnaire and transfusion requirements.

Results: Among the 69 patients (per protocol set population) assessable for efficacy, hemoglobin level change in the 36 000 IU group was significantly greater than that in the 9000 IU group (1.75 ± 2.15 versus 0.04 ± 1.98 g/dl; $P = 0.009$), and a significant dose–response relationship was observed for the change in hemoglobin level ($P = 0.003$). Although changes in FACT-An Total Fatigue subscale (Fatigue subscale) scores were similar for the three dosage groups, there was a statistically significant correlation ($r = 0.435$, $P < 0.001$) between the change in hemoglobin levels and the change in Fatigue subscale scores. The proportion of transfused patients was significantly smaller in the 36 000 IU group compared with that in the 9000 IU group ($P = 0.022$, not adjusted for pre-study transfusions). The incidence of adverse events was similar in the three dosage groups.

Conclusions: Once-weekly epoetin-beta 36 000 IU for 12 weeks was well tolerated and significantly increased hemoglobin levels in anemic cancer patients receiving chemotherapy.

Key words: chemotherapy-induced anemia – erythropoietin – lung cancer – malignant lymphoma – quality of life

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INTRODUCTION

Erythropoietin (EPO) is a glycoprotein (MW 30 000) which is the hematologic growth factor produced primarily in the kidney. EPO interacts with erythroid progenitor cells in the bone marrow to increase the peripheral red blood cells (1). Epoetin-beta is recombinant human erythropoietin (rhEPO) (2), which was introduced clinically in the 1990s for the treatment of anemia associated with chronic renal failure, especially in patients receiving hemodialysis.

Cancer patients treated with chemotherapy often suffer from anemia, which is a major contributing factor to fatigue leading to compromised quality of life (QOL) (3,4). In addition, the presence of anemia is associated with shorter survival of patients with malignancies (5). Red blood cell transfusion is the traditional and quickest method of alleviating symptoms of cancer-related anemia. However, the side effects of transfusion such as viral infections have not been completely resolved. Patients tend to decline transfusions, and physicians do not prescribe them in most cases until the hemoglobin levels become <8.0 g/dl. The administration of rhEPO is another choice for the treatment of chemotherapy-inducted anemia. Numerous studies on anemic cancer patients receiving chemotherapy have demonstrated that rhEPO increased hemoglobin levels and reduced the need for transfusions, and some studies reported improvements in QOL as well (6–11). The schedule of rhEPO administration in most trials was three-times per week. This schedule is inconvenient for outpatients receiving chemotherapy. Gabrilove et al. (10) studied a weekly fixed-dose schedule using 40 000–60 000 IU of epoetin-alfa in cancer patients with anemia. The efficacy was comparable with data on the historical regimen of 10 000 IU three-times weekly. Cazzola et al. (12) compared the efficacy and tolerability of epoetin-beta 30 000 IU once-weekly with that of a 10 000 IU three-times weekly regimen in patients with lymphoproliferative malignancies. Their study showed that the once-weekly regimen was as effective as the three-times weekly one in increasing hemoglobin levels and reducing transfusion requirements.

We therefore conducted a multicenter, randomized, double-blind, dose-finding trial of once-weekly epoetin-beta treatment of malignant lymphoma and lung cancer patients receiving platinum-, taxane- or anthracycline-containing chemotherapy. These chemotherapy regimens are the most active and frequently used for the treatment of these malignancies and also produce relatively high incidences of anemia (4). According to the results of this trial, a recommended dose of epoetin-beta was determined for the subsequent randomized placebo-controlled phase III trial in Japan.

PATIENTS AND METHODS

Patient Eligibility

Patients with histologically or cytologically confirmed malignant lymphoma or lung cancer fulfilling the following criteria were enrolled in the study. (i) Age 20–79 years; (ii) Either platinum-, taxane- or anthracycline-based chemotherapy was administered, and more than 2 courses of chemotherapy were scheduled during the study (radiotherapy during the study period was permitted); (iii) Hemoglobin $\leq$ 11 g/dl after chemotherapy administered within 6 weeks before the study, without iron-deficiencies; (iv) Adequate hepatic and renal function (serum total bilirubin $\leq$ 2.0 mg/dl; serum AST $\leq$ 80 IU/l; serum ALT $\leq$ 80 IU/l; serum creatinine < 2.0 mg/dl); (v) Eastern Cooperative Oncology Group (ECOG) performance status (PS) of 0–2; (vi) Life expectancy of at least 12 weeks. Exclusion criteria included uncontrolled hypertension, gastrointestinal bleeding, and a known history of myocardial infarction, cerebral infarction or pulmonary embolism. Patients with known hypersensitivity to rhEPO and previous treatment with rhEPO within 4 weeks before the study were also excluded. Female patients who were pregnant were not eligible. Written informed consent was obtained from all patients before entry into the study.

Study Design and Treatment Schedule

This study was a multicenter, randomized, double-blind, parallel-group comparative trial. The study protocol was approved by the institutional review board for each of the 11 participating centers in Japan. Epoetin-beta was supplied by Chugai Pharmaceutical Co., LTD (Tokyo, Japan).

Enrolled patients were randomly assigned to receive one of the three dose levels of epoetin-beta (9000, 18 000, or 36 000 IU). Randomization was prospectively stratified according to age, PS, disease (lung cancer or malignant lymphoma) and institution. Subcutaneous injection of epoetin-beta was started at the beginning of the subsequent chemotherapy course and continued, thereafter, once a week for 12 weeks. If the hemoglobin level increased to more than 14 g/dl, epoetin-beta was discontinued until the hemoglobin level decreased to <12 g/dl, and then re-administered at the same dose. An oral iron supplementation (200 mg/day) was taken daily during the study period. No specific guidelines for transfusion use were defined.

Assessment of Efficacy and Safety

The primary end point was change in hemoglobin level, and the secondary end points were QOL and red blood cell transfusion requirements. The change in hemoglobin between the baseline and 12 weeks of administration or the last observation was evaluated. If chemotherapy was discontinued within the 12-week period, the change in hemoglobin was evaluated at the last observation; 4 weeks after the beginning of a final-course of chemotherapy. The QOL instrument used in the study was the Functional Assessment of Cancer Therapy-Anemia (FACT-An) questionnaire (13). The Total Fatigue subscale (Fatigue subscale), which consists of 13 items from FACT-An, was mainly analyzed (scores range from 0 to 52). QOL was measured at the baseline, at 7–11 weeks; at the beginning of a chemotherapy course and at 12 weeks after the initiation of epoetin-beta administration.

Adverse events were graded according to the NCI-Common Toxicity Criteria version 2.0 (Japanese edition; Japan Clinical Oncology Group version 1).

Statistical Analysis

Of the enrolled patients, those who received epoetin-beta at least once were included in the safety analysis. For efficacy analysis, the per protocol set (PPS) population was defined as eligible patients who received epoetin-beta without protocol violation. Differences in mean changes in hemoglobin between the groups were assessed by Dunnett's multivariate comparison test (14). Changes in the Fatigue subscale scores were compared by using a *t*-test. Pearson's correlation coefficient was calculated to assess the relationship between change in hemoglobin and change in the Fatigue subscale scores. The potential factors influencing the change in the Fatigue subscale scores were examined by multiple regression analysis.

To determine the required number of patients, Dunnett's multiple comparison test was conducted with the 9000 IU group as the control arm. At 2.0 g/dl of the change in hemoglobin from baseline and with a 1.8 g/dl standard deviation between the 9000 and 36 000 IU groups, the required number of patients was calculated to be 21 per group; this means that 63 in total (two-tailed significance level: 5.0%; power: 90%). In the study, it was planned to use the PPS as the main analysis for efficacy; therefore, the target number of subjects was established as 84 to allow for patient dropout.

RESULTS

Patient Characteristics

A total of 86 patients were enrolled between April 2002 and January 2003, and 83 patients were administered epoetin-beta. All of these 83 patients were eligible for the assessment of safety. For efficacy analysis, 14 patients were then excluded; 13 patients received <7 doses of epoetin-beta with or without <2 courses of chemotherapy mainly due to progression of the disease; and one patient lacked the baseline hemoglobin data. So 69 patients comprised the PPS population evaluated for efficacy. Baseline characteristics of the patients in the PPS population were generally well balanced among the three dosage groups (Table 1), except for transfusion requirements within 4 weeks before the study; in the 9000 IU group, more patients had required transfusions ($P = 0.130$). Table 2 shows the distribution of chemotherapy regimens used during the study.

Hemoglobin Response

Figure 1 shows the mean weekly hemoglobin levels over the 12 weeks of the study for the patients in the PPS population. In the 36 000 IU group, the mean hemoglobin level increased significantly starting from 6 weeks. In contrast, in the 9000 IU group, the mean hemoglobin levels changed little during the study period, despite a higher transfusion rate. The mean changes in hemoglobin level from baseline to last observation for the three dosage groups were summarized in Fig. 2. In 36 000 IU group, a significantly greater increase in the hemoglobin level was observed compared with that in 9000 IU group ($P = 0.009$); however, there was no significant difference between the 18 000 and 9000 IU groups ($P = 0.154$). A significant dose–response relationship for the change in hemoglobin level was observed ($P = 0.003$). As an additional evaluation of efficacy, the proportion of patients who achieved a ≥2 g/dl increase in hemoglobin level during the study was determined. The results were 40.9% (9/22), 66.7% (16/24) and 78.3% (18/23) in the 9000 IU group, 18 000 IU group and 36 000 IU group, respectively. Epoetin-beta was withheld from 16 patients (one patient in 9000 IU, 8 in 18 000 IU and 7 in 36 000 IU) during the study period, whose hemoglobin levels exceeded 14 g/dl.

Red Blood Cell Transfusion Requirements

Five of 22 patients (22.7%) were transfused in the 9000 IU group, 4 of 24 patients (16.7%) in the 18 000 IU group and none of 23 patients in the 36 000 IU group. The proportion of transfused patients was significantly smaller in the 36 000 IU group compared with that in the 9000 IU group ($P = 0.022$). When patients who had received transfusions within 4 weeks before the study were excluded from the analysis; however, there was no significant difference between the three dosage groups.

QOL

Of the PPS population, 69 patients (100%) at baseline, 62 (89.9%) at 7–11 weeks and 61 (88.4%) at 12 weeks were evaluated for QOL scores. No significant mean change in Fatigue subscale scores was observed in any group at 7–11 weeks and 12 weeks. The relationship between change in hemoglobin level and change in the Fatigue subscale score was examined by correlation analysis. There was a statistically significant correlation ($r = 0.435$, $P < 0.001$) between change in hemoglobin levels and change in the Fatigue subscale scores at 7–11 weeks (Fig. 3). Multiple regression analysis was then performed to assess the potential factors contributing to the change in the Fatigue subscale score at 7–11 weeks. The Fatigue subscale score at baseline and change in hemoglobin level were significantly associated with the change in the Fatigue subscale score ($P = 0.001$). Association with other factors such as the weekly dose of epoetin-beta and chemotherapy regimens were not significantly associated. Patients who achieved an increase in hemoglobin of ≥2 g/dl at 7–11 weeks had significant improvements in their Fatigue subscale scores ($P = 0.012$) (Fig. 4).

Safety

The incidence of adverse events was generally similar between the three dosage groups (Table 3). As hematological adverse events, most common were leukocytopenia, neutropenia and thrombocytopenia. As non-hematological adverse events, nausea and appetite loss were commonly observed. One patient

Table 1. Patient characteristics by epoetin-beta dosage group

Patent population	Epoetin-beta dosage groups			
	9000 IU	18 000 IU	36 000 IU	
Randomly assigned patients (*n*)	28	29	29	
Patients evaluated for safety (*n*)	28	27	28	
Patients evaluated for efficacy (PPS) (*n*)	22	24	23	
Characteristic	9000 IU (*n* = 22)	18 000 IU (*n* = 24)	36 000 IU (*n* = 23)	*P*
Age (year)				
Mean ± SD	60.5 ± 16.6	63.0 ± 11.9	61.9 ± 11.7	0.828
Min–Max	22–79	31–76	34–77	
Weight (kg)				
Mean ± SD	53.5 ± 8.7	50.9 ± 7.3	55.1 ± 11.5	0.316
Min–Max	36.1–69	38.8–66.9	34.8–87.5	
Sex male/female (*n*)	13/9	13/11	14/9	0.890
Disease				
Lung cancer (*n*)	11	13	11	0.907
Malignant lymphoma (*n*)	11	11	12	
de novo/relapse (*n*)	17/5	19/5	18/5	0.988
Performance Status 0/1/2 (*n*)	10/11/1	11/12/1	10/12/1	1.000
RBC transfusion before the study (*n*)	5	2	1	0.130
Hemoglobin (g/dl)				
Mean ± SD	10.1 ± 1.3	10.0 ± 1.5	10.2 ± 1.0	0.914
Min–Max	7.4–12.2	7.4–13.2	8.1–11.7	
Serum EPO concentration (mIU/ml)				
Mean ± SD	43.3 ± 38.1	46.8 ± 43.9	30.4 ± 18.4	0.259
Min–Max	13.1–173	14.4–170	7.0–103	
Serum transferrin saturation (%)				
Mean ± SD	31.1 ± 15.9	25.4 ± 11.5	25.5 ± 13.8	0.287
Min–Max	9.4–77.8	10.1–48.0	6.9–77.4	

SD, standard deviation; Min, minimum; Max, maximum; RBC, red blood cell; EPO, erythropoietin.

in the 36 000 IU group experienced deep vein thrombosis, which was evaluated as unrelated to epoetin-beta. When the thrombosis was found, anemia had not improved (baseline hemoglobin level was 9.9 g/dl and that at the onset of thrombosis was 9.2 g/dl); therefore, deep vein thrombosis was considered to be due to prolonged immobility brought on by aggravated malignant lymphoma and PS.

Severe adverse events were reported for 12 patients and were judged by the investigators as unrelated to the administration of epoetin-beta. Of the adverse events, 65 events in 23 patients (27.7%) were considered related to epoetin-beta. The incidence of these events was similar between the three dosage groups (Table 3). An increase of serum ALT was observed in one patient (3.6%) in the 9000 IU group, two (7.4%) in the 18 000 IU group and two (7.1%) in the 36 000 IU group. Hypertension or an increase of blood pressure was observed in one patient (3.6%) in the 9000 IU group, three (11.1%) in the 18 000 IU group and one (3.6%) in the 36 000 IU group. Drug administration was discontinued in one of these patients due to hypertension. No tendency was found in the onset time of hypertension, nor in changes of hemoglobin from baseline at the time hypertension occurred.

Anti-erythropoietin antibody was not detected in any patient, but pure red cell aplasia (PRCA) was reported in one malignant lymphoma (Angioimmunoblastic T-cell Lymphoma) patient over a year after this trial. In this patient, neutralizing anti-erythropoietin antibody was not detected even after PRCA was diagnosed.

Table 2. Chemotherapy regimens used by PPS population during the study

Chemotherapy regimens	Epoetin-beta dosage groups		
	9000 IU ($n = 22$)	18 000 IU ($n = 24$)	36 000 IU ($n = 23$)
Malignant lymphoma			
(R)CHOP	5	6	9
(R)EPOCH	2	3	2
ESHAP	0	2	0
Other regimens	4	0	1
Lung cancer			
Platinum + Paclitaxel	4	2	2
Platinum + Irinotecan	1	4	3
Platinum + Etoposide	3	2	1
Platinum + Vinorelbine	1	2	1
Other regimens	2	3	4

PPS, Per Protocol Set; (R)CHOP, (Rituximab) + Cyclophosphamide + Doxorubicin + Vincristine + Prednisolone; (R)EPOCH, (Rituximab) + Etoposide + Doxorubicin + Vincristine + Cyclophosphamide + Prednisolone; ESHAP, Etoposide + Methylprednisolone + High Dose Ara C (Cytarabine) + Cisplatin.

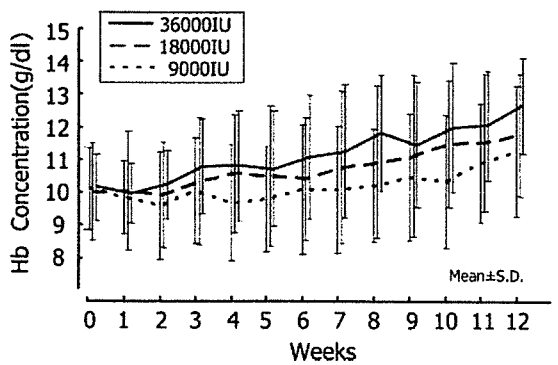

Figure 1. Mean weekly hemoglobin levels for Per Protocol Set population by epoetin-beta dosage Group. Hb, hemoglobin; SD, standard deviation.

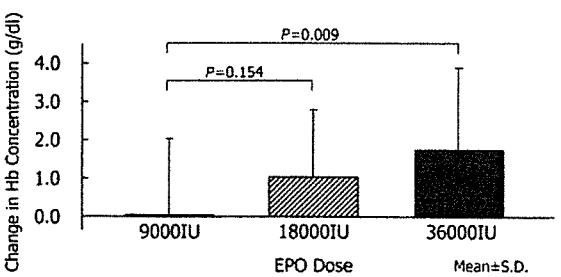

Figure 2. Mean change in hemoglobin level from baseline to last observation (at 12 weeks or 4 weeks after the beginning of a final-course of chemotherapy) by epoetin-beta dosage group (Per Protocol Set population). Hb, hemoglobin; EPO, epoetin-beta; SD, standard deviation.

DISCUSSION

Recently, results of several clinical studies have demonstrated the efficacy and safety of weekly rhEPO for the treatment of cancer-related anemia (10,12,15,16). In a large, nonrandomized, community-based study reported by Gabrilove et al. (10), once-weekly dosing of epoetin-alfa was as effective as three-times weekly dosing in increasing hemoglobin levels and improving QOL. Cazzola et al. (12) reported a randomized study of epoetin-beta that compared the efficacy and tolerability of 30 000 IU once-weekly with the conventional 10 000 IU three-times weekly regimen in patients with lymphoproliferative malignancies. Between these two dosing regimens, there was no significant difference in time-adjusted area under the hemoglobin curve and increase in hemoglobin. Two randomized phase III studies using 40 000 IU once-weekly epoetin-alfa also support the use of epoetin-alfa as an ameliorative agent for cancer-related anemia (15,16).

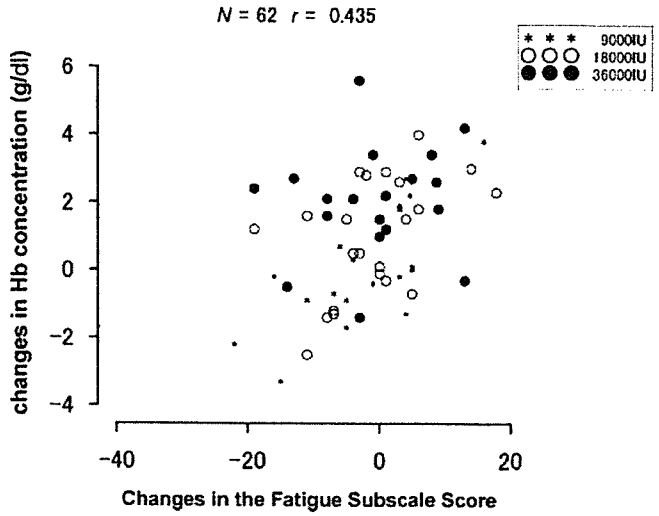

Figure 3. Correlation between change in hemoglobin levels and change in the Functional Assessment of Cancer Therapy—Anemia total Fatigue subscale scores at 7–11 weeks (Per Protocol Set population). Hb, hemoglobin.

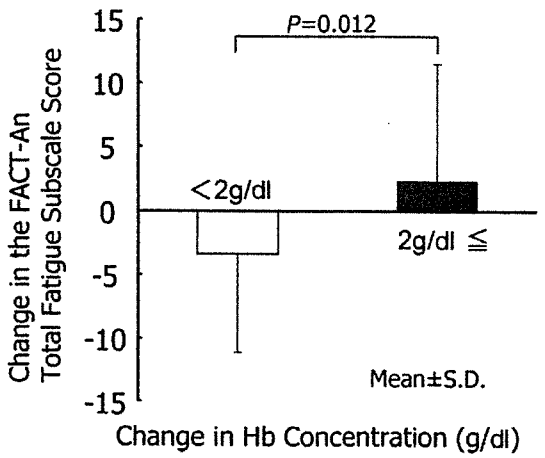

Figure 4. Change in mean Functional Assessment of Cancer Therapy—Anemia total Fatigue subscale score between baseline and 7–11 weeks by change in hemoglobin level (change in hemoglobin of ≥2 g/dl or <2 g/dl from baseline). FACT-An, Functional Assessment of Cancer Therapy–Anemia; Hb, hemoglobin; SD, standard deviation.

Table 3. Incidence of most common adverse events by epoetin-beta dosage group (safety population)

	Epoetin-beta dosage groups							
	9000 IU ($n = 28$)		18 000 IU ($n = 27$)		36 000 IU ($n = 28$)		All patients ($n = 83$)	
	No.	%	No.	%	No.	%	No.	%
Adverse events (incidence > 20%, any grade)								
Leukopenia	23	82.1	24	88.9	23	82.1	70	84.3
Neutropenia	20	71.4	19	70.4	15	53.6	54	65.1
Nausea	15	53.6	19	70.4	16	57.1	50	60.2
Thrombocytopenia	17	60.7	13	48.1	15	53.6	45	54.2
Anorexia	18	64.3	17	63.0	8	28.6	43	51.8
Fever	10	35.7	6	22.2	12	42.9	28	33.7
Vomiting	8	28.6	9	33.3	11	39.3	28	33.7
Malaise	9	32.1	10	37.0	7	25.0	26	31.3
Increased ALT	7	25.0	8	29.6	10	35.7	25	30.1
Diarrhea	8	28.6	10	37.0	6	21.4	24	28.9
Lymphopenia	13	46.4	6	22.2	5	17.9	24	28.9
Fatigue	8	28.6	7	25.9	8	28.6	23	27.7
Increased AST	5	17.9	6	22.2	9	32.1	20	24.1
Increased LDH	3	10.7	11	40.7	6	21.4	20	24.1
Constipation	5	17.9	6	22.2	6	21.4	17	20.5
Adverse events related to epoetin beta (incidence > 3%, any grade)								
Total number of patients	9	32.1	8	29.6	6	21.4	23	27.7
Total number of events	16		32		17		65	
Hypertension/increased blood pressure	1	3.6	3	11.1	1	3.6	5	6.0
Increased ALT	1	3.6	2	7.4	2	7.1	5	6.0

ALT, alanine aminotransferase.

This is the first prospective randomized dose-finding study of once-weekly epoetin-beta in anemic cancer patients treated with chemotherapy. The study demonstrated that the mean increase in hemoglobin level in the 36 000 IU group was significantly higher than that in the 9000 IU group, while the mean increase in hemoglobin level in the 18 000 IU group was not significantly higher than that in the 9000 IU group. In the present study, epoetin-beta 36 000 IU once-weekly administration showed the same efficacy (an increase in hemoglobin level) as a 200 IU/kg thrice-weekly regimen studied in lung cancer patients receiving chemotherapy (6). It is noteworthy to point out that once-weekly epoetin-beta can be conveniently used in an outpatient-based chemotherapy regimen.

FACT-An is one of the widely used QOL assessment tools, which comprises the FACT-General and a 20-item Anemia subscale, 13 items of which make up a Fatigue subscale. Many reports indicated that chemotherapy-induced anemia increased the ease of a patient becoming fatigued and resulted in decreased patient QOL (17–19). The administration of 36 000 IU epoetin-beta did not significantly improve the patients' Fatigue subscale score in spite of increased hemoglobin levels. As a primary goal of the study was to determine a recommended dose of epoetin-beta, the study design was not planned and did not have adequate statistical power to determine whether epoetin-beta would improve the Fatigue subscale scores. According to the results of the study by Hedenus et al. (20), patients with the lowest baseline Fatigue subscale scores (baseline scores of <24) reported the largest improvement in Fatigue subscale scores after the treatment with darbepoetin alfa. In contrast, patients with baseline Fatigue subscale scores of >36 did not show any improvement. In the subset analysis of our study, among the patients with baseline Fatigue subscale scores of ≤36, a mean improvement in the Fatigue subscale scores at 7–11 weeks were −1.8 for the 9000 IU group, +1.9 for the 18 000 IU group and +4.3 for the 36 000 IU group (36 000 IU versus 9000 IU $P = 0.183$). Our data also demonstrated a significant correlation between change in Fatigue subscale score and change in hemoglobin level and showed that the patients who responded

with a hemoglobin increase of $\geq$2 g/dl showed significant improvements in the Fatigue subscale scores. In conjunction with these findings, the administration of epoetin-beta may not be beneficial for the patients with relatively high hemoglobin levels and/or less symptomatic even in an anemic state. Thus, the actual hemoglobin level for initiation of epotein beta will be critical for its optimal use. The ASCO/ASH clinical practice guideline in 2002 recommends the use of rhEPO for chemotherapy-associated anemia patients with the hemoglobin level of $\leq$10 g/dl and that the use of rhEPO for patients with the hemoglobin level of 10–12 g/dl should be determined by clinical circumstances (21).

Most of the adverse events in the present study were considered to be related to concomitant chemotherapy, and the incidence of side effects was similar among the three dosage groups. Two large randomized studies (22,23) targeting higher hemoglobin levels raised concerns about the safety of rhEPO, because of the increased thrombovascular events and worsening survival of cancer patients. In our study, one patient in the 36 000 IU group experienced deep vein thrombosis, which was evaluated as unrelated to epoetin-beta. Stimulated tumor growth is another possible mechanism for worsened survival in the rhEPO studies. A meta-analysis of 27 randomized trials of rhEPO showed suggestive but inconclusive evidence for improved overall survival in patients who received rhEPO (8). Further large scale randomized studies are necessary to confirm the effect of rhEPO on tumor outcome and overall survival.

In conclusion, once-weekly epoetin-beta 36 000 IU for 12 weeks was well tolerated and significantly increased hemoglobin levels in anemic cancer patients receiving chemotherapy. Therefore, the weekly dose of 36 000 IU epoetin-beta was determined as a recommended dose for a subsequent randomized, placebo-controlled, phase III study in Japan.

Part of the results in this report was contributed as Abstract No. 8169 at the 2004 American Society of Clinical Oncology Annual Meeting.

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Association of polymorphisms in the *MTH1* gene with small cell lung carcinoma risk

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Fifty single-nucleotide polymorphisms (SNPs) associated with amino acid changes in 36 genes involved in diverse DNA repair pathways were assessed for associations with risk for small cell lung carcinoma (SCLC) by a case–control study consisting of 211 SCLC cases and 685 controls. An SNP, Val83Met, in the *MTH1* (mutT homolog 1) gene encoding a triphosphatase that hydrolyzes pro-mutagenic oxidized nucleoside triphosphates, such as 8-hydroxy-dGTP and 2-hydroxy-dATP, showed the strongest and a significant association with SCLC risk [odds ratio (OR) = 1.6, 95% confidence interval (CI): 1.2–2.2, $P = 0.004$], while three other SNPs in the *TP53, BLM* and *SNM1* genes, respectively, also showed marginal associations ($0.05 < P < 0.1$). Another SNP, which causes a nucleotide change in the 5′-UTR of *MTH1* transcripts leading to alternative translation initiation, was additionally examined and the SNP also showed a significant association (OR = 1.7, 95% CI: 1.2–2.3, $P = 0.002$). The two SNPs in the *MTH1* gene were in linkage disequilibrium, and the OR for carrying a copy of the haplotype consisting of both the risky SNP alleles was 2.0 (95% CI: 1.2–3.2, $P = 0.002$). The present results indicate that inter-individual differences in MTH1 activities due to SNPs are involved in susceptibility to SCLC.

Introduction

Lung cancer is the leading cause of cancer-related deaths in the world, and is comprised of a group of four histologically distinct types: adenocarcinoma (ADC), squamous cell carcinoma (SQC), large cell carcinoma (LCC) and small cell lung carcinoma (SCLC) (1). SCLC accounts for ~20% of all lung cancer cases and has clinical and biological characteristics distinct from non-small cell lung carcinoma (NSCLC). More than 90% of patients at the time of diagnosis are stage III or stage IV owing to its early and wide dissemination. Although, in most cases tumors initially respond to chemotherapy, >95% of patients eventually die from the cancer. Accordingly, the prognosis of patients with SCLC is poor, and 5-year survival of SCLC is <10% (1–3). Thus, SCLC is the most aggressive type of lung cancer. Genes responsible for the susceptibility to SCLC have been searched for to establish novel and efficient ways of preventing the disease. On the basis of the fact that smoking contributes to SCLC development, polymorphisms in metabolic genes encoding enzymes that activate or detoxify carcinogens in tobacco smoke are being studied for association with SCLC risk by case–control studies. Up to the present, a few metabolic genes, such as *CYP1A1, CYP2A6* and *GSTM1*, have been found to be associated with SCLC risk (4–7). Thus, it is possible that polymorphisms in several metabolic genes are involved in SCLC susceptibility.

Polymorphisms in DNA repair genes have been considered to be involved in susceptibility to cancers, since they are thought to cause inter-individual differences in the capacity for preventing mutagenesis (8–12). In fact, single-nucleotide polymorphisms (SNPs) in several DNA repair genes have been shown to be associated with the risk for several types of cancers (12,13). Carcinogens in cigarette smoke are thought to cause a variety of pro-mutagenic DNA adducts, including benzo[*a*]pyrene-diol-epoxide (BPDE) and 8-hydroxyguanine (8OHG), which are repaired by nucleotide excision repair (NER) and base excision repair (BER) (12). Lung cancer patients were indicated to have lower activities of NER and BER than healthy individuals (9,14). Mice deficient in BER were reported to predispose to lung cancer (15). These results support the fact that inter-individual variations of DNA repair activity are involved in lung cancer susceptibility. We recently identified 50 non-synonymous (associated with amino acid change) SNPs in 36 DNA repair genes involved in diverse intracellular processes that maintain genome

Abbreviations: ADC, adenocarcinoma; CI, confidence interval; LCC, large cell carcinoma; *MTH1*, mutT homolog 1; NSCLC, non-small cell lung carcinoma; OR, odds ratio; SCLC, small cell lung carcinoma; SNPs, single-nucleotide polymorphisms; SQC, squamous cell carcinoma.

[†]These authors contributed equally to this work.

integrity (13) (see Table II). These 50 SNPs were examined for association with NSCLC risk in a case–control study consisting of 752 ADC cases, 250 SQC cases and 685 controls, and four of them, LIG4-Ile658Val, TP53-Arg72Pro, POLI-Thr706Ala and REV1-Phe257Ser, were found associated with NSCLC risk. The results suggested that polymorphisms in genes involved in a variety of DNA repair pathways contribute to NSCLC susceptibility. However, to our knowledge, association of SNPs in DNA repair genes with SCLC risk has not been extensively investigated; therefore, their involvements in SCLC susceptibility is unknown. Thus, in the present study, allele distributions for 50 SNPs in 36 DNA repair genes were examined in 211 SCLC cases to investigate association of the SNPs with SCLC risk. Furthermore, DNA repair genes commonly or specifically involved in susceptibility to SCLC and NSCLC were investigated by comparing the present results with our previous results on NSCLC.

Subjects and method

Case–control study

All cases and controls were Japanese. The cases consisted of 211 SCLC patients of four hospitals located in the Kanto area of Japan (i.e. Tokyo and surrounding prefectures) from 1999 to 2004. The hospitals were the National Cancer Center Hospital (NCCH) (113 patients), the National Cancer Center Hospital East (NCCHE) (81 patients), the National Nishigunma Hospital (NNGH) (16 patients) and the Gunma Prefecture Cancer Center Hospital (1 patient). All SCLC cases, from whom informed consent as well as blood samples were obtained, were consecutively included in this study without any particular exclusion criteria. All the cases were diagnosed by cytological and/or histological examinations according to WHO classification (16). From each individual, a 10 or 20 ml whole-blood sample was obtained. Genomic DNAs for all the cases and the controls were isolated from the samples, and 10 ng of genomic DNA was subjected to genotyping by pyrosequencing as described previously (13). Information on the primer sequences and conditions for PCR were described previously (13).

Genotypes for the 50 SNPs of 685 controls had been determined by the same method as used in the present study (13). The information on the controls was described previously (13). Briefly, the controls consisted of patients of two hospitals, NCCH and NNGH, in which SCLC cases were enrolled, and 302 healthy volunteers of Keio University, located in Tokyo. All of the control subjects were selected with a criterion of no history of any cancer.

Smoking history of cases and controls was obtained via interview using a questionnaire. Smoking habit was expressed by pack-years, which was defined as the number of cigarette packs smoked daily multiplied by years of smoking, both in current smokers and former smokers. Smokers were defined as those who had smoked regularly for 12 months or longer at any time in their life, while non-smokers were defined as those who had not. The study was approved by the institutional review boards of the National Cancer Center, the Nishigunma Hospital, the Gunma Prefecture Cancer Center and Keio University.

Statistical analysis

Differences in the allele distributions for the 50 SNPs between the cases and controls were tested by the χ^2-test. Hardy–Weinberg equilibrium (HWE) tests were performed using the TFPGA software (http://bioweb.usu.edu/mpmbio/). Calculation of the D' and r^2 values and haplotype estimation were undertaken using the EM algorithm. The strength of association of *MTH1* (mutT homolog 1) genotypes and haplotypes with SCLC risk was measured as crude odds ratios (ORs), and ORs were adjusted for gender, age (49, 50–59, 60–69, 70) and smoking dosage (pack-years: 0, 1–49, 50) with 95% confidence intervals (CIs) by unconditional logistic regression analysis (17). ORs for carrying a copy of a haplotype were also calculated by the bootstrap method with 5000 resampling. All the statistical analyses were performed using the SAS version 9 software (SAS Institute, NC, USA).

Results

We conducted a case–control study consisting of 211 SCLC cases and 685 controls (Table I). The SCLC cases consisted of patients enrolled in four hospitals in Tokyo and surrounding prefectures. The 685 controls consisted of patients/outpatients and healthy volunteers without a history of cancer enrolled in two hospitals and a university in the same area. Most of the SCLC cases were males and had a smoking habit, as has been reported (18,19). Therefore, the SCLC cases showed a higher fraction of males and smokers than the controls, and the mean smoking dosage of the SCLC cases was larger than that of the controls.

All the 685 controls were genotyped for the 50 SNPs with a success rate of 99.98% in our previous study (13). The 211 SCLC cases were genotyped for the same 50 SNPs in the present study, and the success rate was 99.94% (Table II). The allele distribution in the SCLC cases was compared with that in the 685 controls. None of the 50 SNPs deviated from HWE in cases and controls ($P \geq 0.05$). A significant difference in the allele distribution between the controls and cases was observed in one of the 50 SNPs, MTH1-Val83Met (OR for the MTH1-83Met allele = 1.6, 95% CI: 1.2–2.2, $P = 0.004$) (Table II). Marginally significant ($0.5 \leq P < 0.1$) allele differentiations were observed in three SNPs, SNM1-His317Asp, TP53-Arg72Pro and BLM-Thr298Met. Allele distributions for the other 46 SNPs were not significantly or were marginally significantly different between the controls and cases.

The relative risks of the genotypes for the four SNPs, which showed significant or marginally significant allele differentiations, were calculated as crude and adjusted ORs. Heterozygotes, homozygotes for the MTH1-83Met allele and carriers of the allele showed significantly increased

Table I. SCLC cases and controls for case–control study

Subject	Institution[a]	No.	Gender (%)		Age	Smoking habit (%)			Pack-years of smokers
			Male	Female	(Mean ± SD)	Non-smoker	Smoker	Unknown	(Mean ± SD)
Case	NCCH	113	88 (78)	25 (22)	61 ± 10	8 (7)	105 (93)	0 (0)	62 ± 31
	NCCHE	81	68 (84)	13 (16)	65 ± 8	0 (0)	77 (95)	4 (5)	57 ± 30
	NNGH[b]	17	16 (94)	1 (6)	68 ± 8	0 (0)	17 (100)	0 (0)	55 ± 25
	Total	211	172 (82)	39 (18)	63 ± 9	8 (4)	199 (94)	4 (2)	59 ± 30
Control	NCCH	242	129 (53)	113 (47)	60 ± 16	138 (57)	102 (42)	2 (1)	36 ± 32
	NNGH	141	100 (71)	41 (29)	65 ± 14	46 (33)	91 (65)	4 (3)	46 ± 35
	KEIO	302	254 (84)	48 (16)	48 ± 10	202 (67)	94 (31)	6 (2)	22 ± 20
	Total	685	483 (71)	202 (29)	55 ± 13	386 (56)	287 (42)	12 (2)	35 ± 31

[a]NCCH, National Cancer Center Hospital; NCCHE, National Cancer Center Hospital East; NNGH, National Nishigunma Hospital; KEIO, Keio university.
[b]Including a case of Gunma Prefecture Cancer Center Hospital.

Table II. Allele frequencies of 50 SNPs in 36 DNA repair genes in controls and cases

DNA repair	Gene	SNP	Amino acid change	dbSNP ID	Minor allele frequency[a]		
					Control[b]	Case	
BER	*PARP/ADPRT*	T2444C	Val762Ala	rs1805412	0.40	0.37	
		A2978G	Lys940Arg	rs1136471	0.05	0.04	
	APEX/APE1	A395G	Ile64Val	rs2307486	0.04	0.05	
		T649G	Asp148Glu	rs3136820	0.38	0.41	
	MBD4	G1212A	Glu346Lys	rs140693	0.35	0.36	
	MTH1/NUDT1	G273A	Val83Met	rs4866	0.09	0.15	($P = 0.004$)
	OGG1	C2243G	Ser326Cys	rs1052133	0.48	0.46	
	XRCC1	C685T	Arg194Trp	rs1799782	0.33	0.30	
		G944A	Arg280His	rs25489	0.09	0.08	
		G1301A	Arg399Gln	rs25487	0.25	0.24	
NER	*XPG/ERCC5*	C3507G	His1104Asp	rs17655	0.42	0.46	
	CSB/ERCC6	G1275A	Gly399Asp	rs2228528	0.45	0.43	
	XPC	A2655C	Lys939Gln	rs2228001	0.40	0.38	
	XPD/ERCC2	G1615A	Asp312Asn	rs1799793	0.04	0.04	
		A2932C	Lys751Gln	rs1052559	0.05	0.05	
Mismatch repair	*MLH1*	A676G	Ile219Val	rs1799977	0.05	0.03	
	MLH3	C2645T	Pro844Leu	rs175080	0.18	0.16	
		C2939T	Thr942Ile	rs17102999	0.05	0.06	
	MSH2	C91T	Thr8Met	rs17217716	0.02	0.02	
	MSH3	A3122G	Thr1036Ala	rs26279	0.24	0.27	
	MSH6	G203A	Gly39Glu	rs1042821	0.32	0.31	
DNA double-strand break repair	*BRCA2*	A1342C	Asn372His	rs144848	0.22	0.21	
	SNM1/KIAA0086	C1867G	His317Asp	rs3750898	0.26	0.30	($P = 0.08$)
	LIG4	A2245G[c]	Ile658Val	rs2232641	0.04	0.06	
	NBS1	C605G	Gln185Glu	rs1805794	0.50	0.46	
	RAD51L3/RAD51D	G501A	Arg165Gln	rs4796033	0.04	0.03	
	RAD54L	A551G	Lys151Glu	rs2295466	0.02	0.01	
	RINT-1	G33C	Glu4Gln	rs818620	0.07	0.09	
	XRCC3	C1075T	Thr241Met	rs861539	0.09	0.09	
DNA damage response	*TP53*	G466C[c]	Arg72Pro	rs1042522	0.33	0.38	($P = 0.097$)
DNA polymerase	*POLD1*	G409A	Arg119His	rs1726801	0.20	0.22	
	POLH/XPV/RAD30	A1840G	Lys535Glu	-	0.03	0.04	
	POLI/RAD30B	A2180G[c]	Thr706Ala	rs8305	0.25	0.24	
	POLL	C1683T	Arg438Trp	rs3730477	0.01	0.012	
	REV1	T982C[c]	Phe257Ser	rs3087386	0.33	0.32	
	A1330G	Asn373Ser	rs3087399	0.04	0.04		
	POLZ/REV3	C4259T	Thr1146Ile	rs462779	0.35	0.37	
Other pathways	*BLM*	C967T	Thr298Met	rs28384991	0.09	0.12	($P = 0.09$)
		G4035A	Val1321Ile	rs7167216	0.04	0.04	
	FANCA	G827A	Ala266Thr	rs17232400	0.03	0.03	
		G1080A	Arg350Gln	rs17233497	0.01	0.01	
		A1532G	Ser501Gly	rs2239359	0.17	0.16	
		A2457G	Asp809Gly	rs7195066	0.03	0.03	
		C3294T	Ser1088Phe	rs7190823	0.02	0.02	
	FANCE	G451T	Arg89Leu	-	0.01	0.00	
		G1213A	Arg343Gln	-	0.04	0.04	
	FANCF	A983G	Lys324Glu	-	0.003	0.002	
	FANCG/XRCC9	C1382T	Thr297Ile	rs2237857	0.12	0.13	
	WRN	C2573T	Thr781Ile	rs17847568	0.03	0.03	
		T4330C	Cys1367Arg	rs1346044	0.09	0.07	

[a]*P*-values by χ^2-test against the control population are shown, when they are <0.1.
[b]Determined in our previous study (12).
[c]Significantly associated with SQC and/or ADC risks in our previous study (12).

ORs, when homozygotes for the 83Val allele were used as a reference, respectively (Table III). On the other hand, ORs of genotypes for the remaining three SNPs, SNM1-His317Asp, TP53-Arg72Pro and BLM-Thr298Met, did not show significant increases or decreases in SCLC cases (data not shown); therefore, these SNPs were not further investigated in the present study.

The *MTH1* gene, whose SNP, Val83Met, showed a significant association as described above, encodes a triphosphatase that hydrolyzes oxidized purine nucleoside triphosphates, such as 8-hydroxy-dGTP and 2-hydroxy-dATP (20). The activity of the MTH1-83Met protein was reported to be more thermolabile than that of the MTH1-83Val protein (20–22). The mitochondrial translocation of the MTH1-83Met protein was reported to be less efficient than that of the MTH1-83Val protein (23). Thus, it was suggested that the MTH1-83Met protein is less active than the MTH1-83Val protein. Previously, another SNP was found in a non-coding exon of *MTH1* (i.e. the T/C SNP in exon 2) 7.0 kb distal to the MTH1-Val83Met SNP, and the C allele in exon 2 leads to the production of an additional translation start site, resulting in the production of a longer MTH polypeptide in addition to commonly produced MTH polypeptides (21). This T/C SNP of the *MTH1* gene was

Table III. MTH1 genotypes and SCLC risk

SNP	Genotype	No. of controls (%)	No. of cases (%)	Crude OR (95% CI, P)	Adjusted OR[a](95% CI, P)
Val83Met	Val/Val	558 (82)	154 (73)	Reference	Reference
	Val/Met	117 (17)	53 (25)	1.6 (1.1–2.4, 0.009)	1.7 (1.1–2.7, 0.03)
	Met/Met	6 (1)	4 (2)	2.4 (0.7–8.7, 0.2)	6.5 (1.3–32.1, 0.02)
	Val/Met + Met/Met	123 (18)	57 (27)	1.7 (1.2–2.4, 0.005)	1.8 (1.2–2.9, 0.01)
T/C in exon 2	T/T	560 (82)	154 (73)	Reference	Reference
	T/C	118 (17)	53 (25)	1.6 (1.1–2.4, 0.009)	1.7 (1.1–2.7, 0.03)
	C/C	3 (0)	4 (2)	4.8 (1.1–21.9, 0.04)	15.7 (2.5–100.6, 0.004)
	T/C + C/C	121 (17)	57 (27)	1.7 (1.2–2.5, 0.004)	1.9 (1.2–3.0, 0.008)

[a]Adjusted for gender, age and smoking dosage.

not included in our 50 SNP set, because it was located in a non-coding exon. However, the above result prompted us to genotype this SNP in the same SCLC and control subjects. Since genotype data for the MTH1-Val83Met SNP were obtained from 681 of the 685 controls and all 211 cases, the genotypes for the T/C SNP were also determined for the same 681 controls and the 211 SCLC cases. Significant allele differentiations were also observed in the T/C SNP (OR for the C allele = 1.7, 95% CI: 1.2–2.3, $P = 0.002$). ORs of the heterozygotes, homozygotes for the exon 2-C allele and carriers of the allele were also significantly increased, when homozygotes for the exon 2-T allele were used as a reference, respectively (Table III).

Since both the case and control subjects in the present case–control study were enrolled in several institutions, it was possible that differences in the institutions lead to the observed allele differentiations due to population stratification. Therefore, we compared allele frequencies for the MTH1-Val83Met and exon 2-T/C SNPs among SCLC cases and controls of each institution (Figure 1). Allele frequencies for the MTH1-Val83Met SNP had been also reported in two other populations consisting of healthy Japanese volunteers (21,24); therefore, the frequencies in those studies were also compared. Frequencies of the 83Met and exon 2-C alleles in any SCLC populations were higher than those in any of the control populations. Allele frequencies for these SNPs were not significantly different among control populations and among case populations ($P > 0.05$ by χ^2-test). We also compared the frequencies of genotypes for the two SNPs, and they were not significantly different among control populations and among case populations, either ($P > 0.05$ by χ^2-test). Thus, it was indicated that the 83Met and exon 2-C alleles were associated with the SCLC risk beyond institutional differences.

Both the SNPs of *MTH1* were found to be in linkage disequilibrium with each other ($D' = 0.97$, $r^2 = 0.91$). Thus, we further evaluated the haplotype differentiation between the SCLC cases and the controls (Table IV). The haplotype consisting of the two risky alleles (i.e., haplotype #2 consisting of the 83Met and C alleles in Table IV) was significantly over-represented in the SCLC population (OR = 1.7, 95% CI: 1.2–2.4, $P = 0.001$), and the OR for haplotype #2 was similar to those for individual 83Met and C allele, respectively. In addition, by taking into account the estimation error of haplotype frequency, crude and adjusted ORs for carrying one copy of haplotype #2 were calculated on the basis of the estimated number of haplotypes for each subject by the bootstrap method, and they were 1.8 (95% CI: 1.2–2.5, $P = 0.0004$) and 2.0 (95% CI: 1.2–3.2, $P = 0.002$), respectively.

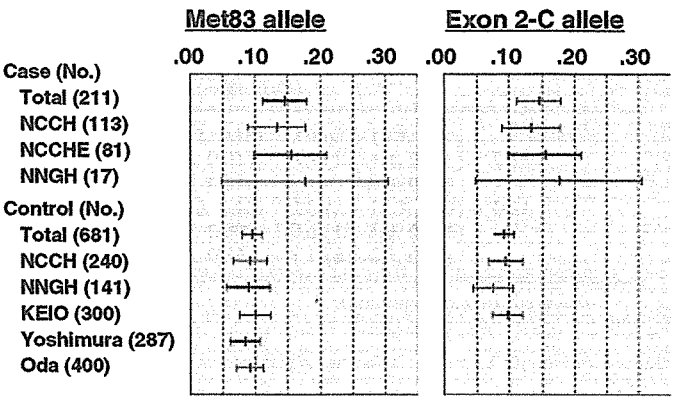

Fig. 1. Frequencies of the MTH-83Met and exon 2-C alleles in cases and controls. Allele frequency is shown with its sampling variations estimated by 95% CI. Frequencies of the MTH-83Met allele in two control populations reported by Yoshimura *et al.* (24) and Oda *et al.* (21) are also shown.

We next assessed the effect of smoking on the contribution of the MTH1-Val83Met and exon 2-T/C SNPs to the SCLC risk. ORs in light (PY < 50) smokers and heavy (PY ≥ 50) smokers were compared (Table V). The number of non-smokers in the case subjects was small (i.e. $N < 10$); therefore, they were excluded from the analysis. Increases of ORs for the 83Met and exon 2-C alleles were more evident in light smokers than in heavy smokers, and the ORs were statistically significant in light smokers but not in heavy smokers. *P*-values for interaction of the Val83Met and exon 2-T/C genotypes on the SCLC risk with smoking were 0.15 and 0.11, respectively. *P*-value for interaction of haplotype #2 on the SCLC risk by smoking was calculated as being 0.095.

Discussion

The *MTH1* gene was cloned as a human homolog for the *Escherichia coli mutT* gene, encoding an enzyme hydrolyzing 8-hydroxy-dGTP, an oxidized dNTP causing A:T to C:G transversion (20). It has been shown that MTH1 protein hydrolyzes not only 8-hydroxy-dGTP but also several other oxidatively damaged dNTPs, such as 2-hydroxy-dATP, thereby preventing multiple mutations including A:T to C:G, G:C to T:A and G:C to A:T mutations (20). *Mth1* nullizygous mice are susceptible to tumor development in lung and other tissues (25). Thus, it has been assumed that inter-individual differences in *MTH1* activity are associated with risks for cancers by causing inter-individual differences

Table IV. Association of MTH1 haplotypes and SCLC risk

Haplotype	SNP		Haplotype frequency		OR (95% CI)	*P*
	Val83Met	T/C in exon 2	Control (95% CI)	Case (95% CI)		
1	Val	T	0.90 (0.89–0.92)	0.85 (0.82–0.89)	Reference	
2	Met	C	0.089 (0.073–0.10)	0.14 (0.11–0.18)	1.7 (1.2-2.4)	0.001
3	Met	T	0.0067 (0.0023–0.011)	0.0024 (0–0.0070)	0.4 (0.05–3.0)	0.3
4	Val	C	0.0030 (0–0.0059)	0.0024 (0–0.0070)	0.9 (0.1–7.7)	0.9

Table V. OR for MTH1 genotypes by smoking dosage and age

SNP	Stratification	No of controls (%)		No of cases (%)		Crude OR	Adjusted OR[a]	*P* for interaction[a]
		Major homozygote	Minor allele carrier	Major homozygote	Minor allele carrier	(95% CI, *P*)	(95% CI, *P*)	
Val83Met	py = 0	319 (83)	67 (17)	5 (63)	3 (38)	2.8 (0.7–12.2, 0.16)	2.9 (0.7–12.7, 0.16)	
	0 < py < 50	178 (82)	38 (18)	59 (69)	26 (31)	2.1 (1.2–3.7, 0.014)	2.3 (1.2–4.4, 0.011)	0.15
	py ≥ 50	54 (78)	15 (22)	88 (77)	26 (23)	1.1 (0.5–2.2, 0.87)	1.1 (0.5–2.3, 0.85)	
T/C in exon 2	py = 0	316 (82)	68 (18)	5 (63)	3 (38)	2.8 (0.7–12.0, 0.16)	2.8 (0.6–12.3, 0.17)	0.11
	0 < py < 50	181 (84)	35 (16)	59 (69)	26 (31)	2.3 (1.3–4.1, 0.006)	2.6 (1.3–4.9, 0.005)	
	py ≥ 50	54 (78)	15 (22)	88 (77)	26 (23)	1.1 (0.5–2.2, 0.87)	1.1 (0.5–2.3, 0.85)	

[a]Adjusted for gender and age.

in the capacity to prevent mutations of the cancer-related genes caused by incorporation of oxidatively damaged dNTPs during DNA replication (20). In the present study, SNPs in the *MTH1* gene were found to be associated with SCLC risk. To the best of our knowledge, SNPs in the *MTH1* gene were found for the first time as being associated with risks for human cancers by a case–control study. However, the possibility of false positives (type I statistical errors) must be considered. We performed 50 separate tests of significance in the analysis. A consecutive Bonferroni adjustment to yield an experiment-wide type I error rate of 0.05 would demand a test-wise *P*-value of 0.001. Therefore, the association of the MTH1-Val83Met SNP would not be considered significant on an experiment-wide level after Bonferroni adjustment. Thus, the association requires confirmation in other population samples, although the present study proposed *MTH1* as a candidate gene responsible for SCLC susceptibility.

The two *MTH1* SNPs, Val83Met and exon 2-T/C, examined in the present study were suggested to cause functional differences, although their effects on mutation suppression efficiency against oxidative DNA damages are unknown (20–22). These two SNPs were in linkage disequilibrium, and the risky allele of each SNP (i.e. the 83Met and exon 2-C alleles) was on the same haplotype (haplotype #2) in most of the Japanese population. Thus, at present, it is unclear whether both or one of the two SNPs are responsible for the SCLC susceptibility. It is also possible that other SNPs consisting of the haplotype are responsible. Further biological and genetic analyses of the *MTH1* SNPs will elucidate the issue.

Interestingly, ORs for carriers of the 83Met and C alleles were more evidently increased in light smokers than in heavy smokers. Tobacco smoke is known to cause oxidative damages on genomic DNA and nucleoside triphosphates (26). Therefore, individuals carrying the 83Met and C alleles might be more prone to acquiring gene mutations even by a low-dose exposure of carcinogens, and therefore, the effects of *MTH* SNPs might have more prominently appeared under the condition of a low-dose exposure of tobacco smoke. On the other hand, the effects of the SNPs might be masked under the condition of a high-dose exposure of tobacco smoke, since, under such a condition, environmental factors (i.e. carcinogens in tobacco smoke) rather than genetic factors predominantly determine the risk for SCLC. However, the interaction of *MTH1* SNPs with smoking on SCLC risk in the present study was not statistically significant; therefore, further case–control studies are necessary to elucidate how *MTH1* SNPs contribute to SCLC risk of smokers.

We previously examined the same 50 SNP set for associations with lung SQC and ADC risk using the same controls (13). In the study, frequencies of the MTH1-83Met allele in SQC and ADC cases, respectively, were slightly higher than that in controls. However, ORs of the carriers of the allele was not significantly increased (Figure 2). Thus, the MTH1-Val83Met SNP was thought to be associated with SCLC risk but not with NSCLC risk. In the previous study, an SNP, TP53-Arg72Pro, in the *p53* gene was associated with SQC risk, and the association remained significant after Bonferroni adjustment. Association of the SNP with NSCLC and overall lung cancer risks have been observed in several other case–control studies (28–31). The association was also supported by a report that TP53-72Pro protein has a weaker activity than TP53-72Arg protein in inducing apoptosis of human cells suffering from DNA damages (32). Interestingly, the TP53-72Pro allele was marginally significantly over-represented in SCLC cases in the present study. ORs of the homozygotes for the carriers of the TP53-72Pro allele were increased in SCLC cases, although the increase was not statistically significant (Figure 2). Thus, it is possible that the TP53-72Pro allele confers increased susceptibility both to SCLC and NSCLC. In the present study, marginally significant associations with SCLC risk were observed for two other SNPs, BLM-Thr298Met and SNM1-His317Asp. However, such associations were not detected in ADC and

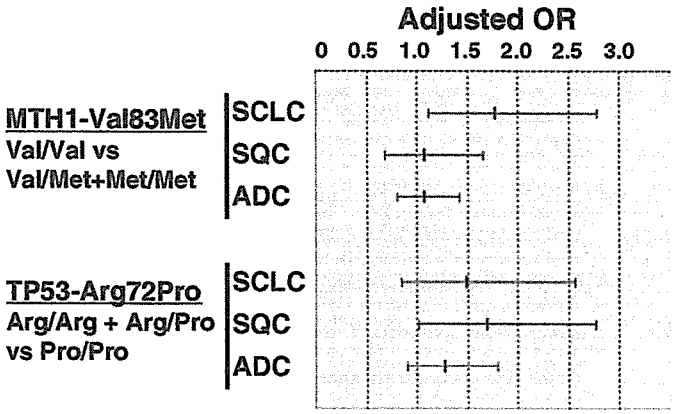

Fig. 2. ORs of the MTH-83Met allele carriers against homozygotes for the MTH-83Val allele and those of homozygotes for the TP53-72Pro allele against others. ORs adjusted for gender, age and smoking dosage are shown. ORs in SQC and ADC cases are from our previous report (13).

SQC (Table II). SNPs that showed association with SQC or ADC risk, such as LIG4-Ile658Val, POLI-Thr706Ala and REV1-Phe257Ser, were not associated with SCLC risk in this study. Thus, genes involved in the susceptibility might be overlapped but different between SCLC and NSCLC.

In the present and previous studies (13), we examined the associations of 50 SNPs in 36 DNA repair genes with SCLC and NSCLC risks. The studies led us to identify several DNA repair genes commonly or specifically involved in the susceptibility to SCLC and NSCLC. The results supported the idea that inherited variations in DNA repair genes are involved in susceptibility to lung cancer of each individual. Further examination of SNPs in DNA repair genes in the present and also in other sets of subjects will help us understand genetic factors responsible for the susceptibility to lung cancer. In addition, studies up to the present suggested that polymorphisms of genes involved in metabolism of carcinogens in cigarette smoke, such as *CYP1A1, CYP2A6* and *GSTM1*, are also responsible for the susceptibility to lung cancer (4–7). It is possible that such polymorphisms modify the effect of SNPs in DNA repair genes on risk for lung cancer. Therefore, combined effects of polymorphisms in DNA repair genes and metabolic genes on risks for SCLC and NSCLC should be also further investigated.

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Docetaxel Consolidation Therapy Following Cisplatin, Vinorelbine, and Concurrent Thoracic Radiotherapy in Patients with Unresectable Stage III Non-small Cell Lung Cancer

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Background: To evaluate the feasibility and efficacy of docetaxel consolidation therapy after concurrent chemoradiotherapy for unresectable stage III non–small cell lung cancer (NSCLC).

Patients and Methods: The eligibility criteria included unresectable stage III NSCLC, no previous treatment, age between 20 and 74 years, and performance status 0 or 1. Treatment consisted of cisplatin (80 mg/m^2 on days 1, 29, and 57), vinorelbine (20 mg/m^2 on days 1, 8, 29, 36, 57, and 64), and thoracic radiotherapy (TRT) (60 Gy/30 fractions over 6 weeks starting on day 2), followed by consolidation docetaxel (60 mg/m^2 every 3 to 4 weeks for three cycles).

Results: Of 97 patients who were enrolled in this study between 2001 and 2003, 93 (76 males and 17 females with a median age of 60) could be evaluated. Chemoradiotherapy was well tolerated; three cycles of chemotherapy and 60 Gy of TRT were administered in 80 (86%) and 87 (94%) patients, respectively. Grade 3 or 4 neutropenia, esophagitis, and pneumonitis developed in 62, 11, and 3 patients, respectively. Docetaxel consolidation was administered in 59 (63%) patients, but three cycles were completed in only 34 (37%) patients. The most common reason for discontinuation was pneumonitis, which developed in 14 (24%) of the 59 patients. During consolidation therapy, grade 3 or 4 neutropenia, esophagitis, and pneumonitis developed in 51, 2, and 4 patients, respectively. A total of four patients died of pneumonitis. We calculated a V_{20} (the percent volume of the normal lung receiving 20 Gy or more) on a dose–volume histogram in 25 patients. Of these, five patients developed grade 3 or more severe radiation pneumonitis. A median V_{20} for these five patients was 35% (range, 26–40%), whereas the median V_{20} for the remaining 20 patients was 30% (range, 17–35%) (p = 0.035 by a Mann–Whitney test). The response rate was 81.7% (95% confidence interval [CI], 72.7–88.0%), with 5 complete and 71 partial responses. The median progression-free survival was 12.8 (CI, 10.2–15.4) months, and median survival was 30.4 (CI, 24.5–36.3) months. The 1-, 2-, and 3-year survival rates were 80.7, 60.2, and 42.6%, respectively.

Conclusion: This regimen produced promising overall survival in patients with stage III NSCLC, but the vast majority of patients could not continue with the docetaxel consolidation because of toxicity.

Key Words: Non-small cell lung cancer, Chemoradiotherapy, Consolidation, Docetaxel.

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Locally advanced unresectable non–small cell lung cancer (NSCLC), stage IIIA with bulky N2 and stage IIIB disease without pleural effusion, is characterized by large primary lesions and/or involvement of the mediastinal or supraclavicular lymph nodes and occult systemic micrometastases. A combination of thoracic radiotherapy and chemotherapy is the standard medical treatment for this disease, but the optimal combination has not been established.[1] Although the available data are insufficient to accurately define the size of a potential benefit,[2] concurrent chemoradiotherapy using a platinum doublet has been shown to be superior to the sequential approach in phase III trials of this disease.[3–5] However, third-generation cytotoxic agents, which have provided better patient survival with extrathoracic spread than the old-generation agents, must be reduced when administered concurrently with thoracic radiotherapy.[6] Thus, it has been hypothesized that the addition of systemic dose chemotherapy with a new cytotoxic agent to concurrent chemoradiotherapy, either as induction or as consolidation chemotherapy, might further improve patient survival.[1]

The consolidation chemotherapy with docetaxel was based on the observation that this drug was highly active in the primary treatment of metastatic NSCLC, producing a response rate (RR) as high as 20% after platinum-based chemotherapy failed.[7–9] Highly encouraging results of a me-

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dian survival time (MST) of more than 2 years and a 3-year survival rate of nearly 40% were obtained in a phase II trial of docetaxel consolidation after chemoradiotherapy with cisplatin and etoposide in patients with stage IIIB NSCLC (SWOG study S9504).[10]

We have developed a combination chemotherapy schedule with cisplatin and vinorelbine concurrently administered with thoracic radiotherapy at a total dose of 60 Gy in 30 fractions in patients with unresectable stage III NSCLC. The results of a phase I study in 18 patients were very promising, with a RR of 83%, a MST of 30 months, and a 3-year survival rate of 50%.[6] Thus, addition of docetaxel consolidation to this regimen is a particularly interesting therapeutic strategy. The objectives of the current study were to evaluate the feasibility of docetaxel consolidation therapy after concurrent chemoradiotherapy with cisplatin and vinorelbine and to evaluate the efficacy and safety of the whole treatment regimen including both the chemoradiotherapy and consolidation therapy in patients with unresectable stage IIIA and IIIB NSCLC.

PATIENTS AND METHODS

Patient Selection

The eligibility criteria were histologically or cytologically proven NSCLC; unresectable stage IIIA or IIIB disease; no previous treatment; measurable disease; tumor within an estimated irradiation field no larger than half the hemithorax; age between 20 and 74 years; Eastern Cooperative Oncology Group performance status (PS) of 0 or 1; adequate bone marrow function (12.0×10^9/liter ≥ white blood cell [WBC] count ≥4.0×10^9/liter, neutrophil count ≥2.0×10^9/liter, hemoglobin ≥10.0 g/dl, and platelet count ≥100×10^9/liter), liver function (total bilirubin ≤1.5 mg/dl and transaminase no more than twice the upper limit of the normal value), and renal function (serum creatinine ≤1.5 mg/dl and creatinine clearance ≥60 ml per minute); and a PaO_2 of 70 torr or more under room air conditions. Patients were excluded if they had malignant pleural or pericardial effusion, active double cancer, a concomitant serious illness such as uncontrolled angina pectoris, myocardial infarction in the previous 3 months, heart failure, uncontrolled diabetes mellitus, uncontrolled hypertension, interstitial pneumonia or lung fibrosis identified by a chest x-ray, chronic obstructive lung disease, infection or other diseases contraindicating chemotherapy or radiotherapy, pregnancy, or if they were breast feeding. All patients gave their written informed consent.

Pretreatment Evaluation

The pretreatment assessment included a complete blood cell count and differential count, routine chemistry determinations, creatinine clearance, blood gas analysis, electrocardiogram, lung function testing, chest x-rays, chest computed tomographic (CT) scan, brain CT scan or magnetic resonance imaging, abdominal CT scan or ultrasonography, and radionuclide bone scan.

Treatment Schedule

Treatment consisted of a chemoradiotherapy phase with three cycles of cisplatin and vinorelbine followed by a consolidation phase with three cycles of docetaxel (Figure 1). Cisplatin 80 mg/m^2 was administered on days 1, 29, and 57 by intravenous infusion for 60 minutes with 2500 to 3000 ml of fluid for hydration. Vinorelbine diluted in 50 ml of normal saline was administered intravenously on days 1, 8, 29, 36, 57, and 64. All patients received prophylactic antiemetic therapy consisting of a $5HT_3$-antagonist and a steroid.

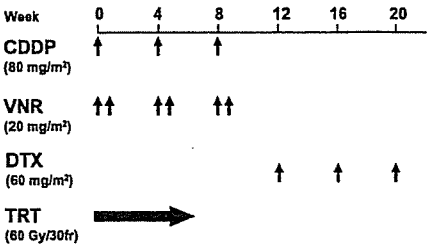

FIGURE 1. Treatment schema. CDDP, cisplatin; DTX, docetaxel; TRT, thoracic radiotherapy; VNR, vinorelbine.

Radiation therapy was delivered with megavoltage equipment (≥6 MV) using anterior/posterior opposed fields up to 40 Gy in 20 fractions including the primary tumor, the metastatic lymph nodes, and the regional nodes. A booster dose of 20 Gy in 10 fractions was given to the primary tumor and the metastatic lymph nodes for a total dose of 60 Gy using bilateral oblique fields. A CT scan–based treatment planning was used in all patients. The clinical target volume (CTV) for the primary tumor was defined as the gross tumor volume (GTV) plus 1 cm taking account of subclinical extension. CTV and GTV for the metastatic nodes (>1 cm in shortest dimension) were the same. Regional nodes, excluding the contralateral hilar and supraclavicular nodes, were included in the CTV, but the lower mediastinal nodes were included only if the primary tumor was located in the lower lobe of the lung. The planning target volumes for the primary tumor, the metastatic lymph nodes, and regional nodes were determined as CTVs plus 0.5- to 1.0-cm margins laterally and 1.0- to 2.0-cm margins craniocaudally, taking account of setup variations and internal organ motion. Lung heterogeneity corrections were not used.

The criteria for starting consolidation chemotherapy were completion of three cycles of cisplatin and vinorelbine and a full dose of thoracic radiotherapy, the absence of progressive disease, adequate general condition within 6 weeks of the start of the third cycle of cisplatin and vinorelbine (PS 0 or 1, WBC count ≥3.0×10^9/liter, neutrophil count ≥1.5×10^9/liter, hemoglobin ≥9.0 g/dl and platelet count ≥100×10^9/liter, total bilirubin ≤1.5 mg/dl and transaminase no more than twice the upper limit of the normal value, and a PaO_2 of 70 torr or more at room air). Docetaxel (60 mg/m^2) was administered intravenously for 1 hour every 3 to 4 weeks for three cycles.

Toxicity Assessment and Treatment Modification

Complete blood cell counts and differential counts, routine chemistry determinations, and a chest x-ray were performed once a week during the course of treatment. Acute toxicity was graded according to the NCI Common Toxicity Criteria, and late toxicity associated with thoracic radiother-