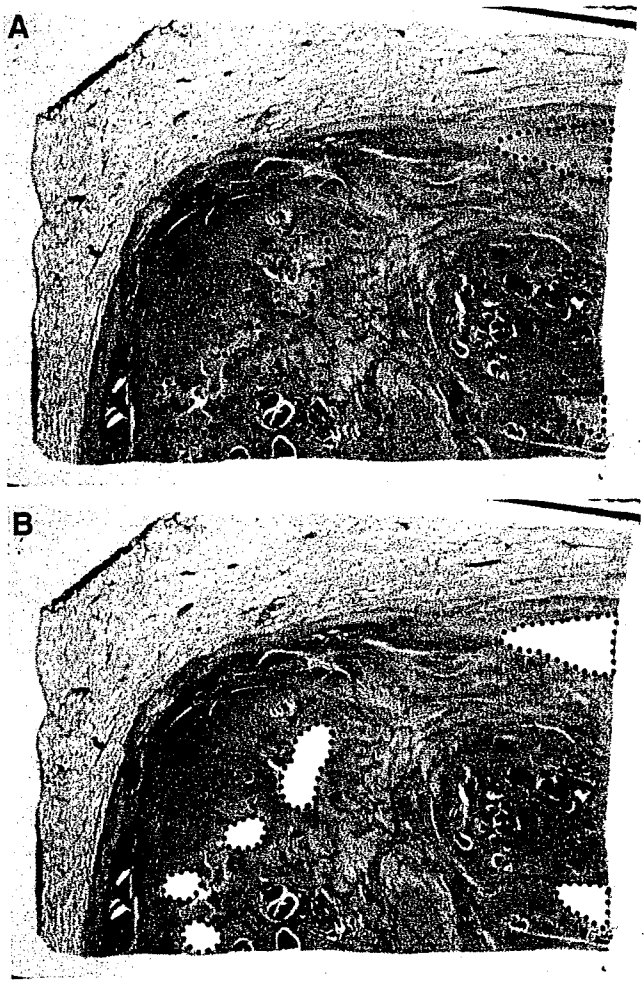

FIGURE 2. Measurement of ART. *A*, We observed the area of the residual tumor (ART) cells on slides under a microscope and marked it with a marker pen. *B*, We photographed the slides and traced the areas that had been marked with image analysis software (shown in yellow).

separate group. We photographed the slides and traced the areas marked in advance with image analysis software Win ROOF version 5.0 (MITANI CORPORATION) (Figure 2*B*).

Statistical Analyses

We analyzed the correlations between the histologic features and whether the patients had received neoadjuvant therapy with Fisher's exact test. We then analyzed the correlations between the histologic features and the clinical response with Fisher's exact test.

We also analyzed the relationships between the histologic features, the ART, ypT, ypN, and pleural invasion and the outcome. These histopathological factors were entered into univariate and multivariate analyses to determine whether they had a significant effect on overall survival. The survival rates were calculated by the Kaplan-Meier method, and the differences were analyzed by means of the log-rank test. The multivariate analysis was performed by means of Cox's proportional hazards model with commercial StatView Version 5.0 statistical software (SAS Institute, Cary, NC).

RESULTS

Clinicopathological Characteristics of the Patients Who Received Neoadjuvant Therapy

Table 1 shows the clinicopathological characteristics of the patients who received neoadjuvant therapy. There were 42 men (79%) and 11 women (21%), and their median age was 60 years (range: 32–74 years). The histologic type was adenocarcinoma in 28 patients, squamous cell carcinoma in 17 patients, large cell carcinoma in four patients, adenosquamous carcinoma in two patients, and pleomorphic carcinoma in two patients. The histologic type in the six patients whose resected specimen contained no residual vital tumor cells was classified by histodiagnosis or cytodiagnosis of specimens obtained by bronchoscopic or transcutaneous needle biopsy before neoadjuvant therapy. Forty-two patients received chemotherapy, 10 patients received chemoradiotherapy, and one patient received radiotherapy before surgery. As neoadjuvant chemotherapy, 17 patients had received mitomycin, vindesine, and cisplatin, and 19 patients had received some kind of platinum-based combination chemotherapy, such as cisplatin plus vindesine, cisplatin plus vinorelbine, cisplatin plus docetaxel, cisplatin plus gemcitabine, or carboplatin plus

TABLE 1. Clinicopathological Characteristics of the Patients Who Received Neoadjuvant Therapy (n = 53)

Characteristic	No. of Patients
Gender	
Male/female	42/11
Age (yr)	
Median (range)	60 (32–74)
Histology	
Adenocarcinoma	28
Squamous cell carcinoma	17
Large cell carcinoma	4
Adenosquamous carcinoma	2
Pleomorphic carcinoma	2
Clinical stage	
IA/IB/IIA/IIB/IIIA/IIIB/IV	0/10/0/18/16/4/5
c-T: T1/T2/T3/T4	1/18/29/5
c-N: N0/N1/N2/N3	29/9/13/2
Pathological stage	
0/IA/IB/IIA/IIB/IIIA/IIIB/IV	6/4/10/1/13/12/7/0
yp-T: T0/T1/T2/T3/T4	6/6/17/17/7
yp-N: N0/N1/N2/N3	32/10/11/0
Neoadjuvant therapy	
Chemotherapy	42
Chemotherapy + radiotherapy	10
Radiotherapy	1
Clinical response	
Complete response	1
Partial response	27
Sable disease	22
Progressive disease	3

TABLE 2. The Comparison of Histological Features of NSCLC According to Whether the Patients had Received Neoadjuvant Therapy

Histological Feature	Neoadjuvant Therapy (+) N = 69 (%)	(−) N = 138 (%)	p
Coagulation necrosis	47 (68)	91 (66)	0.875
Bizarre nucleus in more than 50% of the cancer cells	12 (17)	0 (0)	<0.001
Cholesterin clefts	41 (59)	31 (22)	<0.001
Foam cell infiltration	46 (67)	91 (66)	>0.999
Foreign body reactive giant cells	39 (57)	40 (29)	<0.001
Stromal hyalinosis	46 (67)	37 (27)	<0.001
Foam cell infiltration around the necrotic foci	21 (30)	35 (25)	0.507

NSCLC, non-small cell lung cancer.

TABLE 3. Histological Features and Clinical Response

Histological Feature		CR + PR (n)	SD + PD (n)	p
Bizarre nucleus in more than 50% of the cancer cells	+	5	7	0.533
	−	26	23	
Cholesterin clefts	+	20	16	0.440
	−	11	14	
Foreign body reactive giant cells	+	21	14	0.123
	−	10	16	
Stromal hyalinosis	+	21	19	0.791
	−	10	11	

CR, complete response; PR, partial response; SD, stable disease; PD, progressive disease.

TABLE 4. Univariate Analyses of the Prognostic Factors of NSCLC Treated by Neoadjuvant Therapy (n = 53)

Prognostic Factor		n	5-Yr Survival (%)	p
Histological features				
Bizarre nuclei	+	11	48.5	0.532
	−	42	42.3	
Cholesterin clefts	+	29	40.3	0.976
	−	24	47.4	
Foreign body reactive giant cells	+	29	36.5	0.986
	−	24	50.0	
Stromal hyalinosis	+	33	30.6	0.056
	−	20	64.3	
ART	≤400	27	58.1	0.014
	>400	26	29.6	
ypT	T0–1	12	61.9	0.135
	T2–4	41	39.5	
ypN	N0	32	54.6	0.119
	N1–3	21	31.4	
Pleural invasion	−	20	75.2	0.003
	+	33	28.3	

Bizarre nuclei, Bizarre nucleus in more than 50% of the cancer cells; ART, the area of residual tumor, NSCLC, non-small cell lung cancer.

TABLE 5. Multivariate Analyses of the Prognostic Factors of NSCLC Treated by Neoadjuvant Therapy

Variable	Hazard Ratio	95% CI	p
ART >400	2.063	0.919–4.630	0.079
Pleural invasion (+)	3.600	1.221–10.614	0.020

CI, confidence interval; ART, the area of residual tumor.

paclitaxcel. Only four patients had received docetaxel alone, and two patients had received gefitinib alone. The chemotherapy regimens with radiotherapy were mitomycin, vindesine, and cisplatin or cisplatin plus vinorelbine. The median cycles of chemotherapy was two cycles (range: 1–4). The median total dose of radiotherapy was 45 Gy (range: 28–50). The clinical responses according to Response Evaluation Criteria in Solid Tumors were a complete response (CR) in one patient (2%), partial response (PR) in 27 patients (51%), stable disease in 22 patients (42%), and progressive disease in three patients (6%).

Histologic Features of NSCLC Treated with and without Neoadjuvant Therapy

Table 2 compares the histologic features of NSCLC according to whether the patients had received neoadjuvant therapy. There were no significant differences in the rates of "coagulation necrosis," "foam cell infiltration," or "foam cell infiltration around necrotic foci." "Bizarre nucleus in more than 50% of the cancer cells," "cholesterin clefts," "foreign body reactive giant cells," and "stromal hyalinosis" were observed in a significantly higher proportion of cases in the neoadjuvant group than in the surgery alone group. We then analyzed the correlations between these histologic features and the clinical response, but no significant correlations were found (Table 3).

The Prognostic Factors of NSCLC Treated by Neoadjuvant Therapy

We analyzed the relationships between the histologic features, the ART, ypT, ypN, and pleural invasion and the outcome. Table 4 shows the results of the univariate analyses of the prognostic factors of NSCLC treated by neoadjuvant therapy. ART (>400 mm^2), and pleural invasion (+) were significant prognostic factors for poorer overall survival (p = 0.014 and p = 0.003, respectively). On the other hand, "bizarre nucleus in more than 50% of the cancer cells," "cholesterin clefts," "foreign body reactive giant cells," "stromal hyalinosis," ypT, and ypN did not have any significant prognostic value for overall survival. As shown in Table 5, the multivariate analysis demonstrated pleural invasion (+) to be independent prognostic factor, and the hazard ratio was 3.600 (p = 0.020). ART (>400 mm^2) showed a tendency for poorer overall survival, and the hazard ratio was 2.063; however, it was not significant poor prognosis factor (p = 0.079).

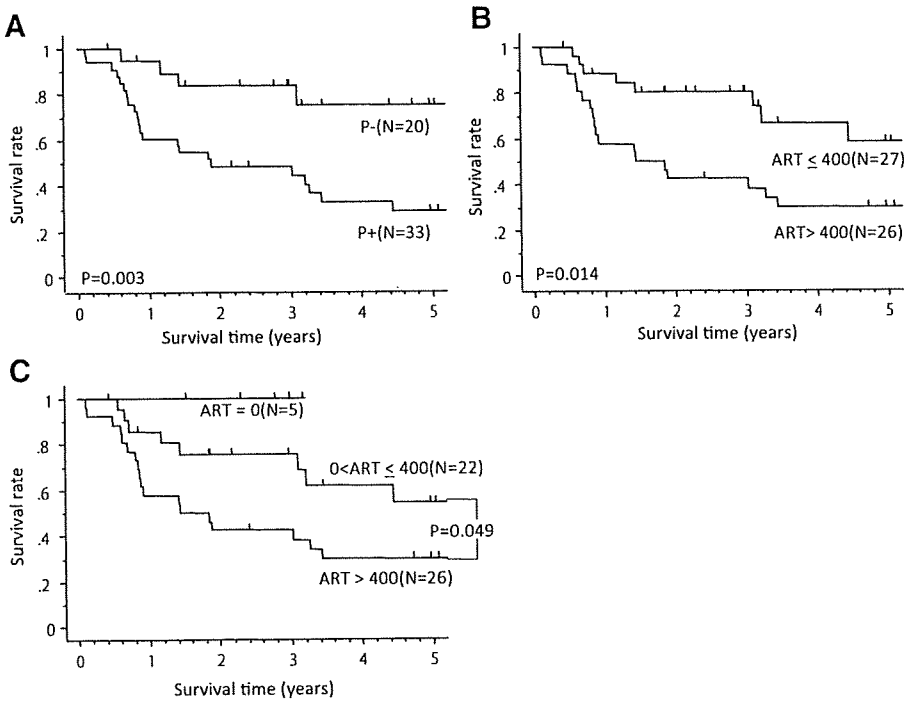

FIGURE 3. Overall survival curves of the patients with non-small cell lung cancer (NSCLC) who received neoadjuvant therapy. Overall survival curves according to (*A*) whether positive or negative for pleural invasion, (*B*) whether ART $\leq$400 mm^2 or >400 mm^2, (*C*) whether ART = 0, ART >0 mm^2 but ART $\leq$400 mm^2, or >400 mm.^{2}

ART Predicts the Outcome of NSCLC Treated by Neoadjuvant Therapy

Figures 3*A*, *B* show survival curves according to pleural invasion, ART, respectively. We identified 27 patients with an ART value $\leq$400 mm^2 as a group of patients who had a better outcome. The 5-year survival rate of those 27 patients was 58.1% as opposed to 29.6% for the 26 patients in the group whose ART value was >400 mm^2 (Figure 3*B*). Furthermore, the five patients with an ART value of 0 had survived without recurrence, and the survival rate of the group of 22 patients with ART value >0 mm^2 but $\leq$400 mm^2 was significantly higher than in the group the 26 patients with ART values >400 mm^2 (Figure 3*C*).

DISCUSSION

The degree of tumor regression based on the histologic findings after neoadjuvant therapy has been considered an objective parameter and has been studied in patients with osteosarcoma,[12] carcinoma of the prostate,[13] esophagus,[14] breast,[15] and head and neck,[16] gastric carcinoma,[17] and NSCLC.[11] Several histologic features have been considered to reflect tumor regression and the prognosis. However, we found that the presence of some histologic features in NSCLC including "coagulation necrosis," "foam cell infiltration," and "foam cell infiltration around necrotic foci" were unrelated to whether the patient received neoadjuvant therapy. Other histologic features including "bizarre nucleus in more than 50% of the cancer cells," "cholesterin clefts," "foreign body reactive giant cells," and "stromal hyalinosis" were observed in higher proportions of NSCLC treated by neoadjuvant therapy, but there were no significant correlations between the presence of these histologic features and clinical response, i.e., tumor reduction assessed radiographically. It has also been reported that there is no association between clinical response and histologic regression.[18,19] Furthermore, these histologic features were not related to a better outcome.

Junker et al.[11] used their own grading system and showed that the grade of therapy-induced tumor regression is a significant prognostic factor in NSCLC. The same grading system was used in our study, and the survival of the grade IIB or III group was significantly better than in the grade I or IIA group (5-year survival rate 62.2 versus 34.8%, $p = 0.031$, data not shown). Becker et al.[17] used a similar grading system and also reported finding that histologic tumor regression grade was an objective measure of the effects of neoadjuvant therapy in patients with gastric carcinoma and that it was significantly correlated with survival. Mandard et al.[14] used a similar grading system and reported grade of tumor regression of esophageal carcinoma treated by neoadjuvant therapy was strongly correlated with disease-free survival. Evaluation of pathologically CRs (ART = 0) is easy, and it is reported that a pathologic CR predicts excellent survival in patients with locally advanced NSCLC who receive neoadjuvant therapy.[20] However, it is sometimes difficult to determine the ratio of residual viable tumor tissue in the primary tumor tissue. We sometimes found that tumor cells remained in the form of islands in the necrotic or fibrotic tissue, and it was difficult to measure the size of tumors after neoadjuvant therapy, and ypT does not always reflect residual tumor size or volume. We found that ypT was not prognosis index in this study also. Measuring ART can be used to overcome these problems.

Although ypT did not have any significant effect on overall survival, ART predicted the outcome. We think it

is because ypT does not always reflect residual tumor size. Because tumor size reflects the prognosis of patients with NSCLC who do not receive neoadjuvant therapy, it is reasonable to think that ART reflects the prognosis of patients with NSCLC who receive neoadjuvant therapy but ypT does not.

Junker et al.[18] found no correlation between clinical response and survival. Our study had same findings (5-year survival rate: CR + PR 47.8 versus SD + PR 34.9%, $p = 0.424$). This observation suggests that pathologic assessment is of potentially greater utility in predicting the patient's prognosis whereas imaging studies may be less useful.

Liu-Jarin et al.[19] found a significantly higher rate of response of patients with squamous cell carcinoma to neoadjuvant chemotherapy compared with patients with adenocarcinoma, according to histologic tumor regression grade. Such a finding could have important implications in the selection criteria of patients with lung cancer to receive neoadjuvant therapy. However, in this study, there was no correlation between histologic type and histologic tumor regression grade. The response rate of histologic tumor regression grade was 28.5% (8 of 28) in adenocarcinoma and 29.4% (5 of 17) in squamous cell carcinoma ($p = 0.951$). When we show the correlation between histologic tumor regression grade or ART and the type of neoadjuvant therapy, the response rate of histologic tumor regression grade was 16.6% (7 of 42) in chemotherapy group and 100% (10 of 10) in chemoradiotherapy group. The rate of patients with ART ≤ 400 mm^2 was 42.8% (18 of 42) in chemotherapy group and 80% (8 of 10) in chemoradiotherapy group.

We also tried measuring ART manually (manual ART) by using a ruler to measure the perpendicular diameters of residual tumor nests on all slides containing the maximum surface area of the tumor. When the sum of the products of the perpendicular diameters was used as the "manual ART" value, the group of patients who had a manual ART value >400 mm^2 had a poorer outcome (5-year survival rate 31.0% versus 60.8%, $p = 0.023$, data not shown). Therefore, manual measurements of ART can also be used in clinical practice.

In this study, we also analyzed the relationship between pleural invasion and outcome, because many cases with pleural invasion were included in the study. Pleural invasion was also found to be a significant prognostic factor for overall survival and the multivariate analysis demonstrated pleural invasion as independent prognostic factor. Pleural invasion has been identified as a predictor of a poor outcome in NSCLC,[21] and we found that pleural invasion was also a prognostic factor for NSCLC in patients who received neoadjuvant therapy.

In conclusion, we found that ART and pleural invasion predict the outcome of NSCLC treated by neoadjuvant therapy. We think that ART is a novel histopathological evaluation method for predicting the outcome of NSCLC treated by neoadjuvant therapy and that it can also serve as a guide to treatment after surgical resection in patients who have received neoadjuvant therapy.

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Differences in the Quality of Information on the Internet about Lung Cancer between the United States and Japan

Yasushi Goto, MD, Ikuo Sekine, MD, PhD,* Hiroshi Sekiguchi, MD,† Kazuhiko Yamada, MD,* Hiroshi Nokihara, MD,* Noboru Yamamoto, MD,* Hideo Kunitoh, MD,* Yuichiro Ohe, MD,* and Tomohide Tamura, MD**

Introduction: Quality of information available over the Internet has been a cause for concern. Our goal was to evaluate the quality of information available on lung cancer in the United States and Japan and assess the differences between the two.

Methods: We conducted a prospective, observational Web review by searching the word "lung cancer" in Japanese and English, using Google Japan (Google-J), Google United States (Google-U), and Yahoo Japan (Yahoo-J). The first 50 Web sites displayed were evaluated from the ethical perspective and for the validity of the information. The administrator of each Web site was also investigated.

Results: Ethical policies were generally well described in the Web sites displayed by Google-U but less well so in the sites displayed by Google-J and Yahoo-J. The differences in the validity of the information available was more striking, in that 80% of the Web sites generated by Google-U described the most appropriate treatment methods, whereas less than 50% of the Web sites displayed by Google-J and Yahoo-J recommended the standard therapy, and more than 10% advertised alternative therapy. Nonprofit organizations and public institutions were the primary Web site administrators in the United States, whereas commercial or personal Web sites were more frequent in Japan.

Conclusion: Differences in the quality of information on lung cancer available over the Internet were apparent between Japan and the United States. The reasons for such differences might be tracked to the administrators of the Web sites. Nonprofit organizations and public institutions are the up-and-coming Web site administrators for relaying reliable medical information.

Key Words: Internet, Information quality, Lung cancer.

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*Division of Internal Medicine and Thoracic Oncology, National Cancer Center Hospital, Chuo-ku, Tokyo, Japan; and †Department of Pulmonary and Critical Care Medicine, Mayo Clinic Minnesota, Rochester, Minnesota.

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Address for correspondence: Yasushi Goto, MD, Division of Internal Medicine and Thoracic Oncology, National Cancer Center Hospital, Tsukiji 5-1-1, Chuo-ku, Tokyo 104-0045, Japan. E-mail: ygoto-tky@umin.net

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The Internet has given rise to an information revolution of unprecedented magnitude. Whereas the Internet has great potential in marshaling the large volume of health information resources available, it is becoming increasingly difficult to discern which of the resources are reliable and accurate or appropriate for the users.[1–6] This issue has become a cause for great concern, especially in the field of oncology, and many studies have evaluated the pros and cons of obtaining information from the Internet.[2–6] Meanwhile, the medical community is being increasingly faced with patients asking us about the medical information available on the Internet. We can no longer neglect the public importance of the information available and have to use it effectively for patients to better understand their disease.

Although one of the main characteristics of the Internet is its worldwide accessibility, differences in language use around the world serve as a bottleneck for collecting information from the Internet. The estimated number of people using the Internet is about the same in the United States and Japan (70 and 67%,[7,8] respectively), and 80% of patients obtain health information via the Internet in the United States.[9] Until now, most studies that have evaluated the quality of the health care information available over the Internet are from the English-speaking community, and very few studies have been conducted in relation to information available in Japanese.[10,11] Furthermore, only a limited number of studies evaluating the differences in the quality of information available between two languages have been published,[12] and no such study comparing such information in the English and Japanese languages has been published.

Our goal was to imitate the search for medical information by the general population in Japan and United States and to evaluate the differences in the process between the two countries. We also investigated the administrators of the Web sites and attempted to identify any correlation existing between the Web site administrators and the quality of information available on the Internet. We focused on information available on lung cancer, which is the leading cause of cancer-related death in both the Unites States and Japan.[13,14] Because search engines are the leading tools to obtain any kind of information, whether general or medical, on the Internet,[15] we used Google and Yahoo, which are the two most commonly used search engines for Web search in both the United States and Japan.

METHODS

Web Site Search

We conducted a prospective, observational Web review by performing keyword searches using Google in both Japanese and English, and Yahoo in Japanese. Japanese searches were conducted by author YG in Japan (Tokyo) on May 29, 2007, and the English search was conducted by author HS in the United States (New York) on May 25, 2007. We used "Hai-gan (both letters in Chinese characters)," "Hai (Chinese character)-gan (hiragana)," and "Hai (Chinese character)-gan (katakana)," for the Japanese search, and "lung cancer" and "lung carcinoma" for the English search. The search word that resulted in the largest number of search results was chosen for the subsequent study.

The first 50 Web sites displayed by Google and Yahoo in Japanese, and Google in English, excluding the advertisement area, were used for further evaluation. Web sites that were inaccessible, not designed to provide health information (i.e., news and advertisement of books), or displayed for the second (or more) time were excluded from the subsequent evaluation. Samples from the Yahoo in English were supplemented to compare the search utility on January 21, 2009.

Site Characteristics

Author YG evaluated the Web sites within a week of the original search. We evaluated the Web sites based on criteria known as the "JAMA" benchmark[16]: display of authorship (authors and contributors, their affiliations, and relevant credentials), attribution (references and sources for all content and all relevant copyright information), disclosure (Web site ownership, sponsorship, advertising, commercial funding arrangements or support, or potential conflicts of interest), and currency (dates on which the contents were posted and updated). We considered each criterion as fulfilled when it was fully displayed. For further evaluation, we focused on the description about the treatment of advanced non-small lung cancer. To our knowledge, there is no established tool-based instrument to evaluate the information available on cancer treatment. Therefore, we classified the information into three categories: acceptable (description of systematic reviews, such as guidelines from authorized facilities,[17–20] links to systematic reviews, or abstracts of systematic reviews), unacceptable (recommendation of alternative medicine or a generally unapproved treatment), and inevaluable (lack of adequate description). The administrators of the Web sites were classified into five categories: nonprofit organization (NPO) or public institution, medical institution, commercial (for specific treatments), personal (pages made by patients or their families), and others.

Analysis

Descriptive statistics were used to determine the numbers and percentages related to the characteristics of the Web sites. To compare the differences between two countries in view of user experience and search utility, Web sites displayed in Google-U was compared with that of Yahoo-J and Google-J, respectively. The χ^2 test or Fisher's exact test was used as appropriate.

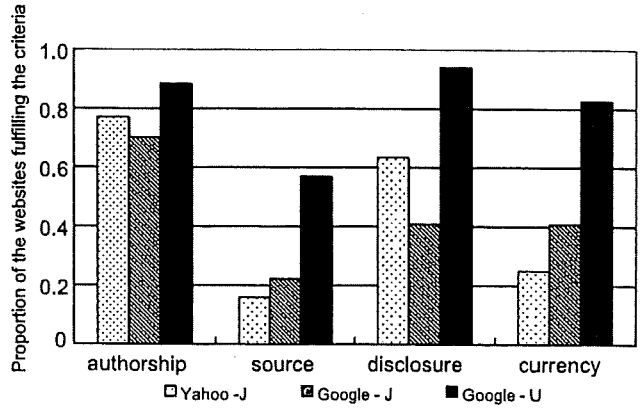

FIGURE 1. JAMA benchmark: Description of the JAMA benchmark[16] is listed by the search engines; display of authorship (authors and contributors, their affiliations, and relevant credentials); attribution (references and sources for all content, and all relevant copyright information); disclosure (Web site ownership, sponsorship, advertising, commercial funding arrangements or support, or potential conflicts of interest); and currency (dates on which the contents were posted and updated).

RESULTS

Differences by Notation

In Google Japan, search using the word "Hai-gan (both letters in Chinese characters)" resulted in a display of approximately 7.7 million Web sites, and in Google United States, search using the phrase "lung cancer" threw up approximately 52 million Web sites. These notations were, therefore, used for the subsequent evaluation. After excluding Web sites that were inaccessible, were not designed to provide health information, or ranked for the second (or more) time in each search, 44, 27, 39, and 35 Web sites displayed by Yahoo Japan (Yahoo-J), Google Japan (Google-J), Yahoo United States (Yahoo-U), and Google United States (Google-U), respectively, were evaluated for further study.

Web Site Characteristics

Figure 1 summarizes the quality of the Web sites that satisfied the criteria of the JAMA benchmark. Authorship was displayed in more than 70% of the Web sites displayed by the three searches: 31 in Google-U (88.6%), 34 in Yahoo-J (70.3%, $p = 0.243$), and 19 in Google-J (88.6%, $p = 0.106$). Attribution of the content was found in 20 (57.1%) of the Web sites in Google-U, and 7 (15.9%, $p < 0.001$) and 6 (22.2%, $p = 0.009$) of the Web sites in Yahoo-J and Google-J, respectively. Twenty-eight (63.6%, $p = 0.001$) Web sites in Yahoo-J, 11 (40.7%, $p < 0.001$) in Google-J, and 33 (94.2%) in Google-U made the disclosure. Display of currency was found in 29 (82.9%) sites in Google-U, but in less than 50% of the Web sites in the Japanese searches; 11 (25.0%, $p < 0.001$) in Yahoo-J and 11 (40.7%, $p = 0.001$) in Google-J.

Quality of Description of the Treatment

Evaluation of the treatment description for advanced non-small cell lung cancer is summarized in Figure 2. The

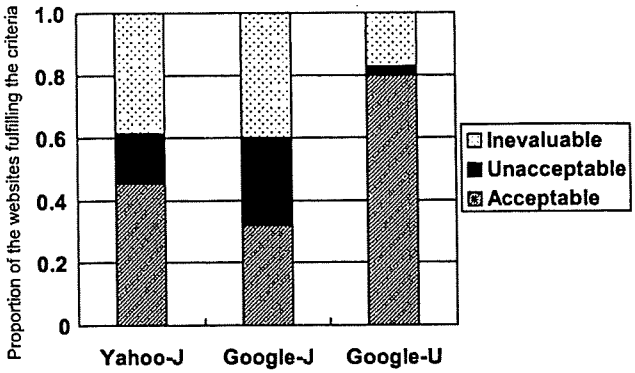

FIGURE 2. Evaluations of the treatment description in the Web sites: The treatment description is classified into three categories: acceptable (description of the systematic review such as guidelines from authorized facilities[17–20]; links to systematic reviews; abstracts of systematic reviews), unacceptable (recommendation of alternative medicine or a generally unapproved treatment), and invaluable (lack of description).

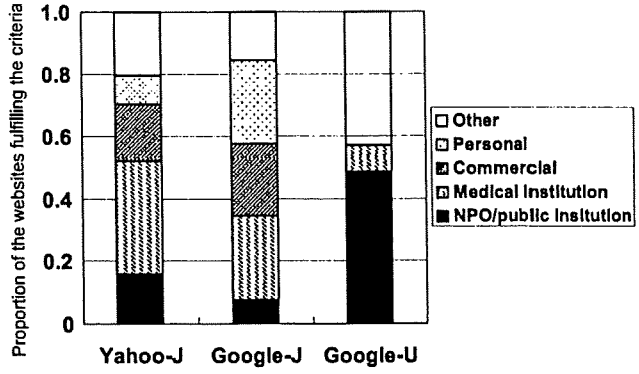

FIGURE 3. Administrators of the Web sites: Administrators were classified into five categories: NPO (nonprofit organization) or public institution, medical institution, commercial (for the specific treatments), personal (pages made by patients or their families), and others.

TABLE 1. Correlation of Sites Between the Top 50 Google and Yahoo, and the Rate of Reliable Sites in Each Engine

	United States	Japan
Correlation of titles in top 50 site of Google and Yahoo	11	10
Percentage of reliable sites in top 50 (%)		
Google	80.0	29.6
Yahoo	71.8[a]	45.5

Correlation of titles in both engines was almost the same in both countries. Proportions of reliable sites were comparable in countries but were not in search engines.

[a] Accessed and evaluated on January 21, 2009.

description was acceptable in 28 (80.0%) of the Web sites generated by Google-U, as these sites described chemotherapy as the standard treatment for advanced lung cancer. Only one site recommended alternative medicine. In Web sites ranked by Yahoo-J and Google-J, standard therapy was only described in 20 (45.5%, $p < 0.001$) and 10 (37.0%, $p < 0.001$) sites, respectively, whereas 7 (15.9%, $p = 0.070$) and 7 (25.9%, $p = 0.017$) sites, respectively, recommended alternative medicine. Table 1 summarizes the quality of the Web sites displayed in Yahoo and Google by both countries. Proportions of reliable sites were comparable in countries but were not in search engines.

Administrators of the Web sites

The administrators of the Web sites are shown in Figure 3. In Google-U, the administrators of 16 (45.7%) Web sites were NPO or public institution, whereas only 7 (15.9%, $p = 0.006$) and 2 (7.4%, $p = 0.001$), respectively, in Yahoo-J and Google-J were managed by them. Commercial site for specific treatments was not displayed in Google-U but was displayed in 8 (18.2%, $p = 0.007$) and 6 (22.2%, $p = 0.005$) Web sites in Yahoo-J and Google-J, respectively. Web sites administered personally by the patients themselves or their families were also not found among the Web site displayed in Google-U, whereas 4 (9.1%, $p = 0.125$) sites in Yahoo-J and 7 (25.9%, $p = 0.002$) sites in Google-J were personally managed.

Administrators and Quality of the Contents of the Web Sites

Table 2 shows the correlation between the Web site administrator and the quality of the contents of the sites. Ten sites generated by both Google-J and Yahoo-J were integrated. There was no site from NPO or public institution category, either Japanese or English, which provided misleading information. Most of the unacceptable sites were managed by commercial or personal sites, neither of which was found in the English-language sites.

DISCUSSION

By comparing the differences of quality of cancer information on the Internet between the different languages, we, for the first time, evaluated the correlation between the Web site administrator and the quality of the medical information in the Web sites. Furthermore, it is one of the few studies to evaluate the information on lung cancer available on the Internet.[15] We also showed that the Web sites displayed in the United States provide information of much higher quality than those displayed by Japanese Web sites, with regard to lung cancer treatment, and this may be related to the quality of the administrators of the displayed Web sites.

It is generally a difficult task to make people access reliable Web sites that would provide the precise information that they are looking for. Regulating access to only trustworthy Web sites that provide useful information is extremely difficult, because a global rule is a necessary step toward controlling the content of the worldwide Web sites. There are also no confirmed tools for weighting the information on the Internet in any field, including medicine. In this chaotic scenario, search engines such as Google and Yahoo have come up with a solution by developing an algorithm to rank the sites. Nowadays, their value is well established in the

TABLE 2. Correlation Between the Quality of the Web site Administrators and the Quality of the Information

	NPO Public Institution	Med Institution	Commercial	Personal	Other	Total
Japanese						
Acceptable	6	10	0	1	5	22
Unacceptable	0	0	10	7	2	19
Inevaluable	2	10	1	1	6	20
Total	8	20	11	9	13	61
English						
Acceptable	15	3	0	0	10	28
Unacceptable	0	0	0	0	1	1
Inevaluable	2	0	0	0	4	6
Total	17	3	0	0	15	35

Ten sites generated by both Google-J and Yahoo-J were integrated. No site from the NPO or public institution category provided misleading information in either the Japanese or the English search. Commercial administrators recommending specific treatments and personal sites accounted entirely for the sites providing unacceptable information.

Internet, and people are generally using this tool for searching medical and other information. Even though there is a concern that the order in which the sites are placed by these tools is not entirely appropriate for the field of medicine,[3,21,22] the high frequency at which these are used has made it meaningless to say that they pose a problem in one-particular field. Therefore, what we must consider now is how to provide reliable information using these tools.

Why is misleading and nonreliable information provided on the Internet? One key characteristic of the Internet is the interaction between the provider and the consumer (in the medical field, patient). Web sites that are not accessed frequently will be ranked lower in the search engine system. Therefore, when discussing the results of Web sites ranked by the search engine, we should consider it from both the standpoint of the provider and the consumer. People access the Internet by requesting the information they want. Many cancer patients suffer from an incurable disease and look for a ray of hope in the Internet. This situation is most advantageous to the information senders. They can promote their treatment as the treatment that would bring about the miraculous cure that the patients are seeking. In this study, most of the sources recommending alternative or unapproved drugs were from commercial and personal sites. Information on medical subjects should be correct and be of assistance to the users to help them better understand their disease. People should be protected from disruptive information. Creating confusion in the minds of people by providing misleading information for profit to the administrator is a vexing situation.

One of the interesting findings in this study was that the correlation between the quality of the Web site administrator and the quality of the contents of the site was seen not only for sites providing misleading information but also for those providing reliable information. At present, there are two major administrators providing reliable information, namely, medical institutions and specialized organizations for information administered by patient advocate NPO or public institution. However, the type of information provided differed between the two types of administrators. In general, each medical institution provides reliable messages but not review articles, whereas the patient advocate group NPO and public institution provide a path to the review articles. This is not surprising because the aims of providing information are different between the two types of administrators. For each medical institution, the goal is to display the treatment that they are interested in, and describing the entire medical consensus is outside their reach. Therefore, sites specialized in providing information are the ones that can be most expected to provide general information. Differences in the number of reliable sites between the languages in this study may be because of the difference in the number of such organizations between the countries. The number of public institution sites may depend on the countries in which each language is spoken in, and the growth in the number of patient advocate NPO may depend on the social system or the differences in culture. However, it is noteworthy that patient advocate NPO can play a major role in providing reliable health information.

There were several limitations in this study. One is that we evaluated sites only from Yahoo Japan and Google Japan, and Google United States. We chose Google United States as the reference, because most previous studies on the Internet have been conducted in the United States, and Google is the most popular search engine in the United States.[23] In Japan, Yahoo ranks first as the most frequently used search engine, followed next by Google,[24] which is the reason we selected these two as the representative search engines for our search of Web sites in Japanese. Although this approach may limit evaluation of the overall Internet situation in the two countries, we believe that this was the closest way to reproduce the way people browse the Internet. Another concern is the number of sites generated by these tools. The total number of Web sites displayed by our search using the keywords differs between the two languages and maybe attributable to the differences in the quality of the administrators. Google-U generated approximately seven times as many Web sites as Google-J. This discrepancy could be because of the difference in the number of people using the two languages. However, we only evaluated the top 50 sites, which is far short of the total number of sites displayed but may already

be too much for anyone seeking any type of information. Because the ranking system has prevailed, the quality of the highest ranked Web sites and not the total number of sites displayed is important to the user. Lastly, another important problem is whether people in the United States and Japan desire the same answers from the Internet. In general, search engines attempt to rank the Web sites sought by the users. If these differed between countries, the ranking would also reflect these differences. Differences in the social backgrounds of the populations in the two countries were confounding factors in this study. However, no studies evaluating the topic from this perspective have been conducted. These are topics of interest that need further investigation.

In this era of abundance of information, it is absolutely essential for people to make their choices based on the quality. As medical professionals, we have the responsibility of providing appropriate information to people who are unaware and anxious about their future. In the new era of the Internet technology, facilitating easy access to reliable information, and providing reliable information is important. This study may facilitate an understanding of the actual status of dispersal of information and pave the way for discussing methods to achieve better accessibility to high-quality health information.

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Circulating Endothelial Cells in Non-small Cell Lung Cancer Patients Treated with Carboplatin and Paclitaxel

Makoto Kawaishi, MD, Yutaka Fujiwara, MD,† Tomoya Fukui, MD,* Terufumi Kato, MD,* Kazuhiko Yamada, MD,† Yuichiro Ohe, MD, PhD,† Hideo Kunitoh, MD, PhD,† Ikuo Sekine, MD, PhD,† Noboru Yamamoto, MD, PhD,† Hiroshi Nokihara, MD, PhD,† Takeshi Watabe, PhD,‡ Yuji Shimoda, PhD,‡ Tokuzo Arao, MD, PhD,§ Kazuto Nishio, MD, PhD,§ Tomohide Tamura, MD† and Fumiaki Koizumi, MD, PhD**

Introduction: Circulating endothelial cells (CECs) increase in cancer patients and play an important role in tumor neovascularization.
Methods: This study was designed to investigate the role of CEC as a marker for predicting the effectiveness of a carboplatin plus paclitaxel based first line chemotherapy in advanced non-small cell lung cancer (NSCLC).
Results: The CEC count in 4 ml of peripheral blood before starting chemotherapy (baseline value) was significantly higher in NSCLC patients, ranging from 32 to 4501/4 ml (n = 31, mean ± SD = 595 ± 832), than in healthy volunteers (n = 53, 46.2 ± 86.3). We did not detect a significant correlation between the CEC count and estimated tumor volume. CECs were significantly decreased by chemotherapy as compared with pretreatment values (175.6 ± 24 and 173.0 ± 24, day +8, +22, respectively). We investigated the correlation between baseline CEC and the clinical effectiveness of chemotherapy. CEC values are significantly higher in patients with clinical benefit (partial response and stable disease, 516 ± 458, 870.8 ± 1215, respectively) than in progressive disease patients (211 ± 150). Furthermore, a statistically significant decrease in CECs, on day 22, was observed only in patients with partial response. Patients who had a baseline CEC count greater than 400/4 ml showed a longer progression-free survival (>400, 271 days [range: 181–361] versus <400, 34 [range: 81–186], p = 0.019).
Conclusion: CEC is suggested to be a promising predictive marker of the clinical efficacy of the CBDCA plus paclitaxel regimen in patients with NSCLC.

Key Words: Circulating endothelial cell, NSCLC, Chemotherapy.

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*Shien-Lab; †Medical Oncology, National Cancer Center Hospital, Chuo-ku, Tokyo, Japan; ‡Center for Molecular Biology and Cytogenetics, SRL Inc., Shinmachi, Hino-shi, Tokyo; and §Department of Genome Biology, Kinki University School of Medicine, Osaka-Sayama-shi, Osaka, Japan.
Disclosure: The authors declare no conflicts of interest.
Address for correspondence: Fumiaki Koizumi, MD, PhD, Shien-Lab, National Cancer Center Hospital, 5-1-1 Tsukiji, Chuo-ku, Tokyo, Japan. E-mail: fkoizumi@gan2.res.ncc.go.jp

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Angiogenesis plays a critical role in the growth and metastasis of solid tumors.[1] The clinical importance of angiogenesis in human tumors has been demonstrated by several reports indicating a positive relationship between the blood vessel density in the tumor mass and poor prognosis, i.e., survival, in patients with various types of cancers including non-small cell lung cancer (NSCLC).[2–6] Furthermore, Natsume et al.[7] reported the antitumor activities of anticancer agents to be less active against vascular endothelial growth factor-secreting cells (SBC-3/VEGF), in vivo as compared with its mock transfectant (SBC-3/Neo). In recent years, antiangiogenic agents have also been demonstrated to be active against a variety of malignancies, including lung, colorectal, and renal cancer.[8–10] Thus, angiogenesis is a promising target for cancer treatment and is related to the prognosis and efficacy of these drugs, though the tumor vessel biomarkers which predict the effectiveness of antiangiogenic agents and other anticancer agents are not always useful and have not become well-established.

Circulating endothelial cells (CECs) have been recognized as a useful biomarker for vascular damage. CECs are increased in cardiovascular disease, vasculitis, infectious disease, and various cancers.[11–14] Recently, CECs were found to be more numerous and viable in cancer patients than in healthy subjects.[14,15] Furthermore, elevated CECs in cancer patients were found to be nearly normalized when the tumor was removed surgically or with chemotherapy.[15] Therefore, most CECs are considered to be disseminated tissue endothelial cells in the tumors and the CEC number may reflect the extent of tumor angiogenesis. Indeed, the CEC level has been demonstrated to correlate with the plasma level of VEGF, one of the pivotal factors promoting tumor angiogenesis.[15] Mancuso et al. reported that CEC kinetics and viability are promising predictors of the response to chemotherapy with antiangiogenic activity in patients with advanced breast cancer.[16] Thus, CEC is likely to be a useful marker for predicting the effectiveness of chemotherapy as a noninvasive angiogenesis marker.

NSCLC is the leading cause of cancer-related death worldwide. NSCLC accounts for approximately 50% of patients presenting with unresectable advanced stage,[17] and platinum-based chemotherapy offers only a small improve-

ment in survival with advanced NSCLC.[18,19] Over the past decade, several new agents against NSCLC have become available, including the taxanes, gemcitabine, vinorelbine, and irinotecan. The combination of platinum and these new agents has resulted in a high response rate and prolonged survival compared with older chemotherapy regimens (e.g., vindesine, mitomycin, ifosfamide, with cisplatin). Therefore, these regimens are considered standard chemotherapy for advanced NSCLC.[20–26] Although new agents have different mechanisms of action, these combination regimens have not been administered based on the biologic characteristics of each tumor.

Paclitaxel inhibits several endothelial cell functions in vitro such as proliferation, migration, morphogenesis, and metalloprotease production.[27–29] These activities result in antiangiogenic activity in in vivo xenograft models.[27,30] Interestingly, human endothelial cells are more sensitive to paclitaxel than other cellular types.[29] We hypothesized that the CEC value is associated with tumor neovascularization, which is one of the targets of paclitaxel. In the present study, we investigated whether the CEC count at baseline is associated with the effectiveness of the CDDP plus paclitaxel regimen in patients with advanced-stage NSCLC.

MATERIALS AND METHODS

Patients

Patients with histologically or cytologically documented advanced NSCLC were eligible for this study. Each patient was required to meet the following criteria: (1) no prior treatment including chemotherapy, surgery, irradiation, or any fluid drainage; (2) no prior general anesthesia for diagnostic procedures including mediastinoscopy or thoracoscopy; (3) no concomitant diseases including ischemic heart diseases, systemic vasculitis, pulmonary hypertension, or serious complications including infectious disease or diabetes; (4) written informed consent. The trial document was approved by the institutional review board. The clinical characteristics of the patients are shown in Table 1.

TABLE 1. Baseline Characteristics of the Patients

Characteristic	$N = 31$ No. (%)
Gender	
Male	17 (55)
Female	14 (45)
Median age (yr)	60
Range	43–71
ECOG performance status	
0	18 (58)
1	13 (42)
Stage	
IIIA	2 (6)
IIIB	7 (23)
IV	22 (71)
Histology	
Adenocarcinoma	23 (74)
Squamous cell carcinoma	4 (13)
Others	4 (13)

Treatment Schedule and Response Evaluation

All patients were treated according to the following chemotherapeutic regimen: paclitaxel at 200 mg/m^2 over a 3-hour period followed by carboplatin at a dose with an area under the curve of 6 on day 1, repeated every 3 weeks. The treatment was repeated for three or more cycles unless the patients met the criteria for progressive disease (PD) or experienced unacceptable toxicity.

The major axis (a) and minor axis (b) of the tumor mass in each patient were measured with computed tomography. Estimated tumor volume (ETV) was calculated using the following formula; ETV = $4/3 \times \pi$ (a/2 × b/2) × (a/2 + b/2)/2. Computed tomography examinations were performed before treatment and with every one or two cycles of chemotherapy. Response was evaluated according to the RECIST, and tumor markers were excluded from the criteria.[31]

Assay for CEC

Blood samples from NSCLC patients and healthy volunteers were drawn into a 10-ml Cellsave Preservative Tube (Immunicon Corp. Huntingdon Valley, PA) for CEC enumeration. The CEC protocol used was approved by the Institutional Review Board and written informed consent was obtained from each subject. Samples from NSCLC were obtained before (baseline) and 8 and 22 days after starting chemotherapy. Samples were kept at room temperature and processed within 42 hours after collection. All evaluations were performed without knowledge of the clinical status of the patients. The CellTracks system (Immunicon Corp) which consists of CellTracks AutoPrep system and the CellSpotter Analyzer system was used for endothelial cell enumeration.[32,33] In this system, CD146+/DAPI+/CD105-PE+/CD45APC- cells are defined as CECs. Briefly, cells which express CD146 were immunomagnetically captured using ferrofluids coated with CD146 antibodies. The enriched cells were then labeled with the nuclear dye 4V,6-diamidino-2-phenylindole (DAPI), CD105 antibodies conjugated to phycoerythrin (CD105-PE), and the pan-leukocyte antibody CD45 conjugated to allophycocyanin (CD45-APC). In this system, the CD146-enriched, fluorescently labeled cells were identified as CECs when the cells exhibited the DAPI+/CD105+/CD45- phenotype. We performed CEC enumeration twice, using the same sample, and calculated the mean value.

Statistical Analyses

This study was carried out as exploratory research for detecting CECs from NSCLC patients. The number of enrolled patients was therefore not precalculated. Spearman's correlation analysis was performed to investigate the correlation between CEC count and ETV. Between-group comparisons were made using the t test. The association between CEC count and progression free survival (PFS) was estimated using the Kaplan-Meier method. The log-rank test was used to assess the survival difference between strata. Differences were considered statistically significant at $p < 0.05$.

RESULTS

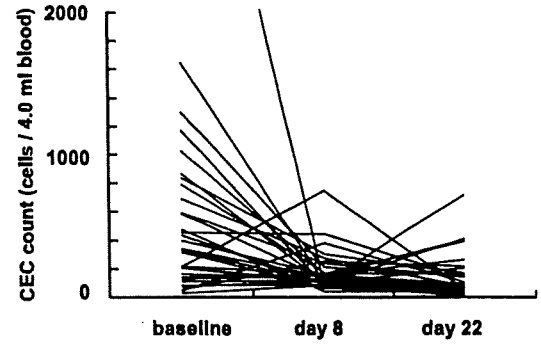
Patient Characteristics

A total of 32 patients were enrolled in the study between August 2005 and March 2006 (Table 1). One patient withdrew consent to participate. Table 1 summarizes the characteristics of the study population. The median age of the patients was 60 years (range, 43–71). The histologic and/or cytologic diagnosis was adenocarcinoma in 23 patients (74.2%), squamous cell carcinoma in 4 (12.9%), and unclassified NSCLC in 4 (12.9%). There were 17 males (54.8%). The clinical stage was IIIA in 2 patients (6.5%), IIIB in 7 (22.6%), and IV in 22 (71.0%).

Ninety-two CEC samples from 31 patients (three samples per patient) were obtained and analyzed. One sample, obtained 22 days after treatment, was not examined because of inadequate collection.

Quantification of CEC

In 31 advanced NSCLC patients, CECs ranged from 32 to 4501 cells/4.0 ml of blood, mean ± SD = 595 ± 832 at baseline. CEC counts were elevated in a large portion of patients with NSCLC as compared with healthy volunteers (n = 53, mean ± SD = 46.2 ± 86.3/4 ml). Case 21 had an exceptionally high CEC count (4501 at baseline). We did not detect a significant correlation between the CEC count and ETV in the 28 assessable patients (p = 0.84, Figure 1). The analysis of CECs during the first course of treatment showed CEC levels to be reduced by CBDCA plus paclitaxel chemotherapy as compared with pretreatment values (176 ± 141 at 8 days and 173 ± 189 at 22 days after treatment) (Figure 2). These reductions were significant (p = 0.011 on day 8 and p = 0.04 on day 22), but there was no significant difference between CEC amounts on day 8 versus day 22 (p = 0.476). There was no difference in the amount of CEC at baseline when patients were subgrouped according to characteristics, such as sex, smoking history, histologic type, and clinical stage. Furthermore, there was no correlation of CEC amounts with the blood examination data (e.g., number of white blood cells, neutrophils, lymphocytes, hemoglobin, platelets, albumin, LDH, CRP, CEA, CYFRA).



	baseline	day 8	day 22
mean ± SD	595± 832	176± 141 *	173 ± 189 *

FIGURE 2. Circulating endothelial cell (CEC) levels during the first course of CDDP plus paclitaxel chemotherapy. *p < 0.05 versus values at baseline.

CEC Amounts and Objective Tumor Response to Chemotherapy

Thirteen (41.9%) of the 31 patients who received carboplatin and paclitaxel therapy showed a partial response (PR) and 12 (38.7%) showed stable disease (SD). The other 6 patients (19.4%) showed PD. The amounts of CEC at baseline in the patients who showed PR and SD were 516 ± 458/4 ml and 871 ± 1215/4 ml, respectively, and these values were significantly higher than in PD patients (211 ± 150/4 ml, p = 0.023 and p = 0.044, respectively) (Figure 3*A*). Although CEC decrements during chemotherapy were observed in all three subgroups, the extent of the decrements tended to be greater in

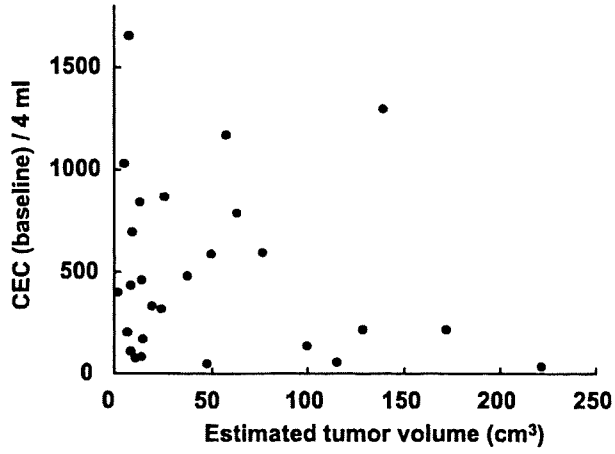


FIGURE 1. Scatter plot analysis to determine the correlation between the number of circulating endothelial cell (CEC) and estimated tumor volume (ETV). ETV is calculated with computed tomography (CT) examination. Case 21 is not included.

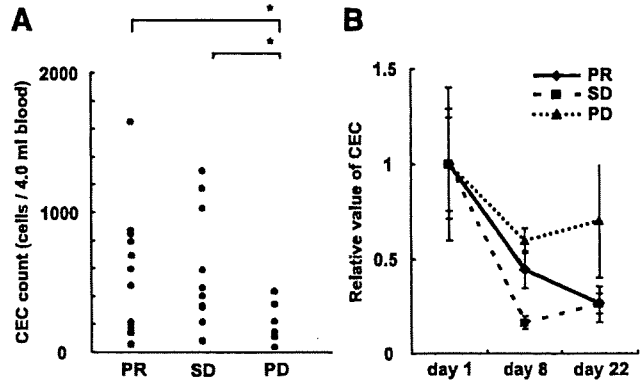


FIGURE 3. *A*, Comparison of circulating endothelial cell (CEC) amount at baseline in non-small cell lung cancer (NSCLC) patients with different clinical responses to CBDCA plus paclitaxel chemotherapy. *p < 0.05 versus values of patients with progressive disease (PD). Case 21 is not included. *B*, Relative change in CEC amount in patients with partial response (PR), stable disease (SD), and PD.

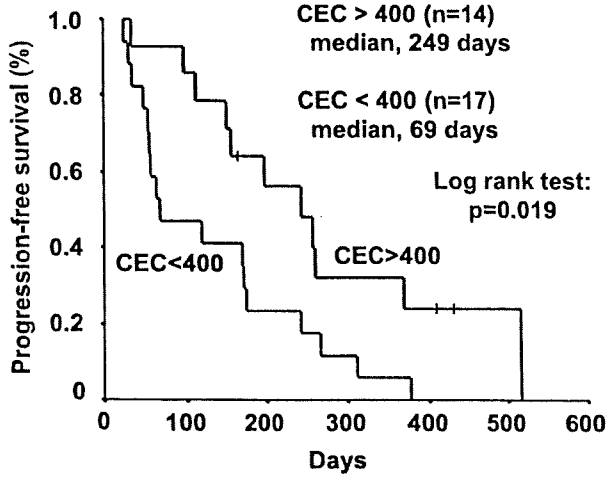


FIGURE 4. Progression-free-survival according to circulating endothelial cell (CEC) count at baseline. The median duration of progression-free survival was greater in patients whose CEC count exceeded 400 (median, 244 days) than in patients whose CEC count was less than 400 (69 days).

patients with PR and SD than in those with PD (Figure 3*B*). In the subgroup analysis, a significant decrease in CECs was observed on day 22 only in PR patients ($p = 0.018$).

CEC Amounts and PFS

For all 31 patients, the median PFS was 154 days (range, 81–361 days). Univariate analysis indicated that patients who had a CEC count of more than 400/4 ml at baseline showed a significantly improved PFS ($n = 14$, median; 244 days) (Log-rank test, $p = 0.019$, Figure 4). A CEC count below 400 at baseline was associated with a poorer PFS ($n = 17$, median; 69 days). The CEC count did not exceed the value of 400/4 ml in any of the healthy volunteers. When we compared the patients whose CEC counts exceeded 200 with those whose counts were less than 200, a consistent difference in PFS was observed between the two groups (>200; $n = 22$, median 227, <200; $n = 9$, median 116, $p < 0.039$).

DISCUSSION

In the present study, we investigated the number of CEC during the first course of CBDCA plus paclitaxel chemotherapy. To our knowledge, this is the first report of CEC in NSCLC patients before treatment. Our findings demonstrated CEC counts in advanced NSCLC at baseline level to be much higher than those in healthy subjects (595 ± 832/4.0 ml versus 32.6 ± 29.5/4.0 ml). Because the NSCLC patients had not yet received anticancer therapy, these increased CECs are likely to be mostly derived from the tumor site. In a previous study, it was found that the amounts of CECs correlate strongly with tumor volume in vivo in an animal model[34]. Nevertheless, we did not find a significant correlation between CECs and ETV. Because the number of CECs could be influenced by many factors related to tumor vasculature, neovascularization, and localization of the tumor, our failure to identify a strong correlation in this study is not surprising. We were also unable to detect a significant direct correlation between CEC amounts and various blood examination data including tumor markers such as CEA and CYFRA. It is unclear at present what biologic characteristics of the tumor or clinical features the CEC number most closely reflects as a biomarker. Mancuso et al. reported that CECs are strongly associated with plasma levels of VCAM-1 and VEGF in breast cancer and lymphoma patients.[15,31] Because VCAM-1 and VEGF are crucial factors for tumor angiogenesis, the variability in CEC values among NSCLC patients might indicate a difference in the neovascularization of each tumor.

We were further able to demonstrate that elevated CECs decreased dramatically after CBDCA plus paclitaxel treatment, but did not reach the level of healthy subjects. Decreased CEC values did not rise again during the first cycle of chemotherapy. Although myelosupression was observed on day 8 and recovered on day 22 in many patients (data not shown), CEC kinetics do not parallel those of WBC, indicating that CEC kinetics might not be influenced by myelopoiesis. Several clinical studies in the field measuring CEC found chemotherapy to be associated with either an increase or a decrease in CECs.[15–19] The different tumor types, stages, prior therapy or not, the anticancer drugs used, measuring points and quantification methods of CEC might have influenced the CEC results after treatment. In the present study, the pretreatment CEC value was much higher than that in lung cancer with metastasis (mean ± SD = 146 ± 270/4 ml), as reported elsewhere.[33] Although the details of the prior therapy in patients with metastatic carcinoma were not provided,[33] chemotherapy can eventually decrease the CEC count.

Schiller et al. compared four standard chemotherapy regimens, cisplatin plus paclitaxel, cisplatin plus gemcitabine, cisplatin plus docetaxel, and carboplatin plus paclitaxel and found no significant difference in survival.[25] Despite the different modes of action of each nonplatinum agent against tumors and different biologic characteristics of each tumor, we could not select the regimen based on these characteristics. In our small study, the patients with PR/SD and longer PFS had higher baseline CEC values. Therefore, it seems that the baseline CEC count is a promising predictor of clinical response to the CBDCA plus paclitaxel regimen and survival in advanced NSCLC. If CEC is a marker for angiogenesis and reflects tumor neovascularization, it is likely that a high CEC is associated with a poor prognosis and lower effectiveness of antiangiogenic therapy. Paclitaxel and docetaxel are categorized as mitotic spindle agents with potent antiangiogenic properties.[27–30] This is why a paclitaxel based regimen might be more effective against tumors with high CEC values. Nevertheless, CEC counts have also been reported to be increased in several clinical syndromes, such as cardiovascular diseases, infectious diseases, and vasculitides.[11–13] The CEC counts in patients with vasculitides have been reported to be dozens of fold higher than those in healthy subjects,[12] therefore, we have to consider the patient condition carefully while interpreting the CEC counts in individual patients, although there were no patients with vasculitis in the present study. Further clinical investigation, with a similar approach, including other nonplatinum anticancer agents, such as

CDDP plus gemcitabine, is essential for the clinical application of CEC for made-to-order chemotherapy in NSCLC.

Antiangiogenic therapy targeting the VEGF pathway such as bevacizumab and VEGFR inhibitors have shown promise in the treatment of solid tumors.[8,39] These agents inhibit endothelial cells through inhibition of the VEGF pathway. It was recently demonstrated that the addition of bevacizumab to CBDCA plus paclitaxel in advanced NSCLC patients produces a significant survival benefit as compared with chemotherapy alone.[40] Considering the outstanding clinical trial and our present study, it would be of great interest to investigate the role of CEC in this regimen.

In conclusion, CECs were measured in NSCLC patients before treatment. Our small clinical study indicates that the CEC count at baseline is a potential biomarker for predicting the response to chemotherapy and PFS, but further clinical evaluation is needed. In the near future, we will start a clinical investigation, using a similar approach, to examine other chemotherapeutic regimens.

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Clinical Studies

Characteristics and outcomes of patients with advanced non-small-cell lung cancer who declined to participate in randomised clinical chemotherapy trials

C Tanai*[,1], H Nokihara[1], S Yamamoto[2], H Kunitoh[1], N Yamamoto[1], I Sekine[1], Y Ohe[1] and T Tamura[1]

[1]Department of Medical Oncology, National Cancer Center Hospital, Tokyo, Japan; [2]Cancer Information Services and Surveillance Division, Center for Cancer Control and Information Services, National Cancer Center, Tokyo, Japan

There are inadequate data on the outcomes of patients who declined to participate in randomised clinical trials as compared with those of participants. We retrospectively reviewed the patient characteristics and treatment outcomes of both participants and non-participants in the two randomised trials for chemotherapy-naive advanced non-small-cell lung cancer. Trial 1 compared four platinum-based combination regimens. Trial 2 compared two sequences of carboplatin plus paclitaxel and gefitinib therapies. Nineteen of 119 (16%) and 153 (37%) patients declined to participate in Trials 1 and 2, respectively. Among the background patient characteristics, the only variable associated with trial participation or declining was the patients' attending physicians ($P<0.001$). Important differences were not observed in the clinical outcomes between participants and non-participants, for whom the response rates were 30.6 *vs* 34.2% and the median survival times were 489 *vs* 461 days, respectively. The hazard ratio for overall survival, adjusted for other confounding variables, was 0.965 (95% confidence interval: 0.73–1.28). In conclusion, there was no evidence to suggest any difference in the characteristics and clinical outcomes between participants and non-participants. Trial designs and the doctor–patient relationship may have an impact on the patient accrual to randomised trials.
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Randomised clinical trials (RCTs) are the definitive method for comparing the efficacy of treatments and a crucial step in the development of new cancer treatments. There has always been a big problem that their low accrual rates limit their progress (Lara *et al*, 2001; Corrie *et al*, 2003; Go *et al*, 2006).

A number of studies have examined the motivations of patients for accepting or declining entry to RCTs (Jenkins and Fallowfield, 2000; Madsen *et al*, 2000, 2002; Ellis *et al*, 2001; Wright *et al*, 2004; Ho *et al*, 2006; Albrecht *et al*, 2008). The results of questionnaire surveys administered to patients regarding clinical trials revealed that two of the most common reasons for entering the trial were the hope for personal benefit and the opportunity to contribute to the research knowledge thereby benefiting others in the future (Jenkins and Fallowfield, 2000; Madsen *et al*, 2000, 2002; Ellis *et al*, 2001; Wright *et al*, 2004; Albrecht *et al*, 2008). On the other hand, the common reasons for declining participation were worries about the process of randomisation, overestimation of the benefits of standard therapy and fear of the trial's experimental nature (Jenkins and Fallowfield, 2000; Ellis *et al*, 2001; Ho *et al*, 2006).

However, inadequate data are available on the actual outcomes of non-participants compared with those participating in RCTs (Schmoor *et al*, 1996; Braunholtz *et al*, 2001; Burgers *et al*, 2002; Peppercorn *et al*, 2004; West *et al*, 2005). Although several reports and their review (Braunholtz *et al*, 2001) have suggested the existence of a 'trial effect', in which participants enjoy favourable outcomes, others, especially those which attempted to exclude the confounding factors, have refuted this finding (Schmoor *et al*, 1996; Burgers *et al*, 2002; Peppercorn *et al*, 2004; West *et al*, 2005).

On the other hand, if participation in prospective trials is associated with certain clinical characteristics of the patients, generalisability of the conclusion from the data to the clinical practise, even in patients who meet the restrictive eligibility criteria, should be in question.

The purpose of this study was to analyse the characteristics and outcomes of the patients who met the eligibility criteria but declined to participate in RCTs, as compared with those who did participate, and to search for clues to improve patient accrual to clinical trials.

MATERIALS AND METHODS

Between October 2000 and October 2005, each of the 272 patients, who fulfilled the entry criteria of our top priority studies during the period, was informed of all aspects of RCTs on non-small-cell lung cancer (NSCLC) and was invited to participate in one of the two trials to be conducted at the National Cancer Center Hospital, Tokyo, Japan. We make it a rule for each patient with advanced

*Correspondence: Dr C Tanai, Department of Internal Medicine, National Cancer Center Hospital, 5-1-1 Tsukiji, Chuo-ku, Tokyo 104-0045, Japan; E-mail: ctanai@ncc.go.jp
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lung cancer to be hospitalised for the first-line chemotherapy. All patients are then checked for the eligibility criteria of clinical trials available at the time and recorded in our database, whether or not they are treated on trials.

Signed informed consent was obtained from the patients for future statistical analysis of their clinical courses and outcomes, even when they were treated outside clinical trials.

Trial 1 was conducted to compare the four platinum-based combination regimens (cisplatin–irinotecan, carboplatin–paclitaxel, cisplatin–gemcitabine and cisplatin–vinorelbine) in patients with untreated advanced NSCLC between October 2000 and June 2002 (Ohe *et al*, 2007). When patients declined to participate, cisplatin-based combination regimens, such as cisplatin–irinotecan, the reference arm of the trial, were recommended. The patients ultimately selected the treatment following discussions with their families and the physicians.

Trial 2 was conducted between June 2003 and October 2005 to compare the following two treatment arms; (A) four courses of carboplatin and paclitaxel (CP) followed by gefitinib, and (B) gefitinib until disease progression followed by CP, in patients with advanced NSCLC (Nokihara *et al*, 2008). When patients declined to participate, platinum-based combination regimens, such as CP, were recommended. The patients ultimately selected the treatment following discussions with their families and the physicians; treatment options included gefitinib as first-line chemotherapy, when the patients and their families wished to start with it.

Patients in each trial had to meet the following criteria: histologically and/or cytologically documented NSCLC; clinical stage IV or IIIB (including only patients with no indications for curative radiotherapy); no earlier systematic chemotherapy; at least one measurable lesion; age 20–74 years old; Eastern Cooperative Oncology Group Performance Status (PS) of 0 or 1; adequate haematological, hepatic and renal functions; and partial pressure of arterial oxygen of 60 torr or more. Each patient was required to submit a written informed consent before entry.

Four physicians (A, B, C and D) participated in Trial 1 and five physicians (A, B, C, D and E) in Trial 2. All were male. Physicians A, B, C and D had 16, 14, 11 and 9 years of experience, respectively, at the time of activation of Trial 1 (October 2000), and Physician E had 9 years of experience at the start of Trial 2 (June 2003). One of the five attending staff physicians and one to two residents or trainees attended each consultation. Which doctor actually offered the RCTs depended on each case and was not recorded, but the attending staff physician finally confirmed the decision by the patient.

Paper and/or electronic medical records from the initial visit to our centre to the end of the follow-up were retrospectively reviewed. Demographic data (age, gender, smoking history), medical information (tumour histology, clinical stage, performance status, therapy characteristics), and clinical outcomes (response rate, follow-up time, overall survival time, 1- and 2-year survival rates) were abstracted and analysed. The response was evaluated according to the Response Evaluation Criteria in Solid Tumours (RECIST) (Therasse *et al*, 2000) by the attending physicians. It is our policy to assess clinical responses with RECIST, even in routine practise. Follow-up time at our institution was defined as the period from the initiation of the first day of the initial therapy or decision of no therapy, to the last day at our institution (including death during follow-up). Survival data of the patients who left our institution could be collected by enquiry into official agency for family registry in Japan.

χ^2-tests and logistic regression analysis was used to assess associations between patient characteristics and the rate of declining to participate. Overall survival (OS) curves were produced using the Kaplan–Meier method and compared with the log rank test. All participants (those who agreed to be enroled into the RCT) and non-participants (those who declined to participate in the RCT) were included in the OS analysis. A Cox proportional hazards model was used to adjust for other potential confounding factors (age, gender, smoking history, clinical stage and PS) in comparing the OS of participants and non-participants. *P*-values <0.05 were considered statistically significant. The data collected were analysed using an SPSS II statistical package.

Japanese ethics guidelines for clinical and epidemiological studies, which took effect in August 2007, do not mandate institutional review board (IRB) approval for a single-institutional, retrospective data analysis from the medical charts, when the predesignated person of the institution so judges. This study was thus exempted from ethical review of IRB in due process, on the judgment of the responsible official, deputy director of National Cancer Center Hospital.

RESULTS

There were no significant differences in the outcomes between the arms of each trial. In Trial 1, no statistically significant differences in the response rate, progression-free survival and OS were observed between the four regimens. In Trial 2, there were no statistically significant differences in the median survival time (MST) (18.8 and 17.2 months) and the survival rate at 1 year between the two arms. Seventy-five patients declined to participate in those trials, and 1 of the 197 who initially accepted entry withdrew consent, refusing to continue the trial immediately after randomisation.

Table 1 shows the patient characteristics and rate of declining. 100 patients accepted and 19 patients (16%) declined entry to Trial 1, and 96 patients accepted and 57 patients (37%) declined entry to clinical Trial 2 (including the one patient already mentioned who withdrew consent after randomisation) ($P<0.001$). No significant influence on the rate of declining of patient gender, age,

Table 1 Patient characteristics and rate of declining

	Clinical trial 1			Clinical trial 2			Total		
	P	NP	ROD (%)	P	NP	ROD (%)	P	NP	ROD (%)
No.	100	19	16	96	57	37	196	76	28
Gender									
Male	64	12	16	55	34	38	119	46	28
Female	36	7	16	41	23	36	77	30	28
Age									
<60	46	9	16	37	29	44	83	38	31
⩾60	54	10	16	59	28	32	113	38	25
Smoking history									
+	69	9	12	55	33	38	124	43	26
–	31	10	24	41	24	37	72	33	31
Clinical stage									
III	24	6	20	21	19	48	45	25	36
IV	76	13	15	75	38	34	151	51	25
PS									
0	27	4	13	47	19	29	74	23	24
1	73	15	17	49	38	44	122	53	30
Physicians									
A	32	5	14	23	25	52	55	30	35
B	28	0	0	25	1	4	53	1	2
C	18	2	10	34	4	11	52	6	10
D	22	12	35	7	18	72	29	30	51
E	—	—	—	7	9	56	7	9	56

Abbreviations: NP = non-participants, P = participants; PS = performance status; ROD = rate of declining.

Table 2 Prediction of participation or declining to trials

	Univariate analysis[a]		Multivariate analysis[b]	
	Odds ratio (95% CI)	***P*-value**	**Odds ratio (95% CI)**	***P*-value**
Gender (male *vs* female)	1.008 (0.586–1.733)	0.977	0.646 (0.300–1.391)	0.264
Age (<60 *vs* ⩾60)	0.735 (0.432–1.250)	0.254	0.701 (0.376–1.310)	0.266
Smoking history (+ *vs* −)	1.394 (0.815–2.386)	0.225	2.538 (1.162–5.541)	0.019
Clinical stage (III *vs* IV)	0.608 (0.339–1.089)	0.093	0.681 (0.346–1.340)	0.266
PS (0 *vs* 1)	1.398 (0.792–2.467)	0.247	0.785 (0.396–1.554)	0.487
Physicians (A–E)		<0.001		<0.001

Abbreviations: NP = non-participant; P = participant; PS = performance status; ROD = rate of declining. [a]By Pearson's χ^2-test. [b]By logistic regression analysis.

Table 3 Number of courses of the first-line chemotherapy

	Clinical trial 1		Clinical trial 2		
	Participants	**Non-participants**	**Participants**	**Non-participants**	***P*-value**
	100	16	96	57	
First-line cycles					
1	10 (10%)	4 (25%)	6 (12%)	4 (9%)	0.418[a]
2	18 (18%)	4 (25%)	8 (16%)	12 (27%)	
3	37 (37%)	7 (44%)	5 (10%)	9 (20%)	
⩾4	35 (35%)	1 (6%)	30 (61%)	20 (44%)	
Gefitinib median duration (day)			73	99	0.118[b]
Range			13–752	34–1065	
IQR			29–204	38.5–512	

Abbreviation: IQR = interquartile range. [a]By Pearson's χ^2-test. [b]By log rank test.

smoking history, tumour histology, clinical stage or PS was observed (Table 2). There were, however, large differences in the rates of decline among the attending physicians who informed the patients about the trials and asked them to participate ($P<0.001$).

The treatment regimens for those who declined participation in the clinical trials were as follows. The majority of those who declined participation in Trial 1 selected one of the four platinum-based combination regimens presented in the trial: cisplatin–irinotecan 4, cisplatin–vinorelbine 3, cisplatin–gemcitabine 1, carboplatin–paclitaxel 4. Three patients in Trial 1 desired to have no more active treatments and opted for supportive care only, but later received active treatment at their referred hospitals. The detail of their therapy is unknown.

The majority of those who declined participation in Trial 2 selected carboplatin-based combination chemotherapy: carboplatin–paclitaxel 34 and carboplatin–gemcitabine 11, there by reflecting the shift to carboplatin for advanced NSCLC in Japan at the time of Trial 2, on the basis of the reports on the activity of the carboplatin-based regimens (Kelly *et al*, 2001; Schiller *et al*, 2002; Ohe *et al*, 2007). Twelve patients (21%) selected gefitinib as first-line chemotherapy.

Survival was analysed for all of the 196 participants and 76 of the non-participants. Post-therapy was analysed for all of the 196 participants and 73 of the non-participants, who were treated at our centre. There was one possible treatment-related death due to perforation of the colon during gefitinib treatment in Trial 2. No other toxic deaths were observed among either participants or non-participants. More participants of both the clinical trials were given four cycles or more of the first-line chemotherapy, probably reflecting protocol regulations (Table 3).

Table 4 summarises the treatment after the initial therapy. There were no significant differences between participants and non-participants in the number of chemotherapy regimens. Six (8%) of those who declined participation in the trial later participated in early-phase clinical trials of experimental therapies.

Table 4 Treatment after the first-line chemotherapy

	Participants 196 (%)	**Non-participants 73 (%)**	***P*-value[a]**
Chemotherapy regimen			
0[b]	26	40	0.108
1	38	26	
2	22	25	
3	9	8	
>4	5	1	
Radiotherapy	49	34	0.031
Pleural or pericardial drainage	10	5	0.227
Operation on metastatic brain tumors	1	3	0.122
Early-phase trials	13	8	0.300

[a]By Pearson's χ^2-test. [b]Patients received first-line chemotherapy only.

We have observed no clinically relevant differences in the clinical outcomes between participants and non-participants (Table 5). Clinical response to the initial therapy was analysed for all of the 196 participants and 73 of the non-participants, excluding three patients who were not treated at our institute. The response rate was 30.6% in participants and 34.2% in non-participants ($P=0.325$). The median follow-up time at our centre was 388 days for participants and 406 days for non-participants, which was not statistically different.

The OS was not different between participants and non-participants (Table 5 and Figure 1), with a hazard ratio of participants *vs* non-participants of 0.998 (95% confidence interval: 0.76–1.32). No significant difference in OS was observed either in Trial 1 (Figure 2) or in Trial 2 (Figure 3).

Table 5 Clinical outcomes

	Clinical trial 1		Clinical trial 2		Total		
	Participants	Non-participants	Participants	Non-participants	Participants	Non-participants	*P*-value
Response rate (%)[a]	29 (29/100)	12.5 (2/16)	32.3 (31/96)	40 (23/57)	30.6 (60/196)	34.2 (25/73)	0.569[b]
Median follow-up time (day)	329	339	493	444	388	406	0.846[c]
Range	45–2704	1–2176	36–2036	22–1688	36–2704	1–2176	
IQR	177–665	59–582	213–861	175–658	197–742	146–604	
Median survival time (day)	416	408	573	519	489	461	0.987[c]
Range	34–2704	53–2380	40–2036	35–1688	34–2704	35–2380	
IQR	264–815	140–698	251–938	276–1012	259–863	229–774	
1-year survival (%)	56.0	63.2	65.6	64.9	60.7	64.5	0.567[b]
2-year survival (%)	29.4	21.1	38.5	29.8	33.9	27.6	0.379[b]

Abbreviation: IQR = interquartile range. [a]Excluding three patients who did not receive active treatment at our center. [b]By Pearson's χ^2-test. [c]By log rank test.

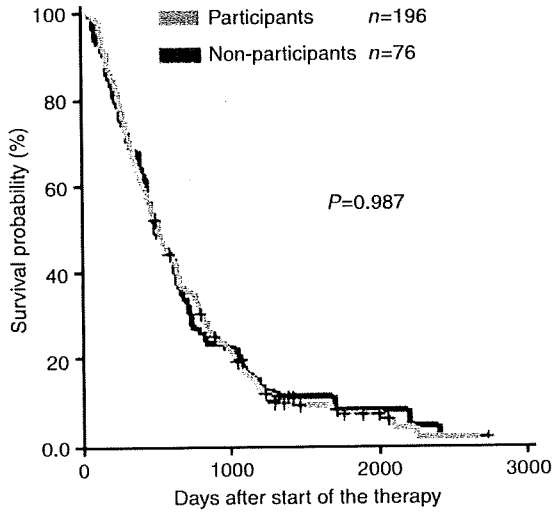

Figure 1 Overall survival of those who declined to participate in randomised trials (blue line, $n = 76$) as compared with the participants (pink line, $n = 196$). No significant difference can be observed.

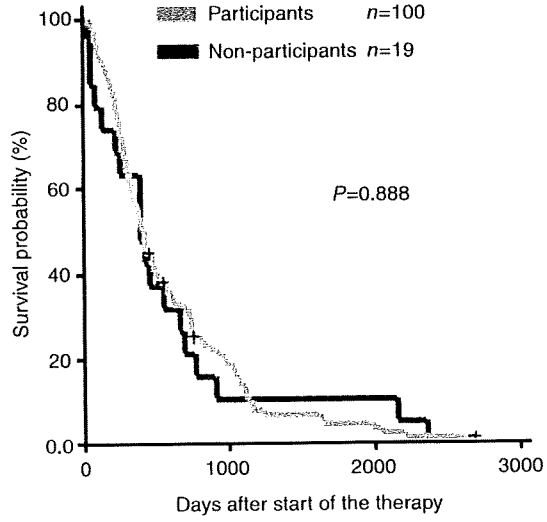

Figure 2 Overall survival of those who declined to participate in Trial 1 (blue line, $n = 19$) as compared with the participants (pink line, $n = 100$). No significant difference can be observed.

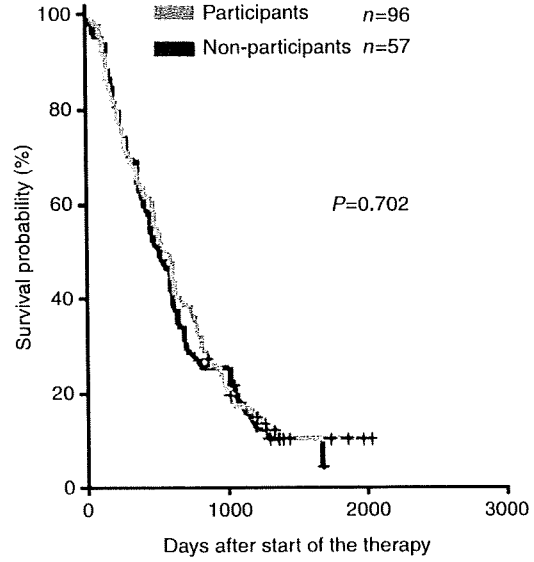

Figure 3 Overall survival of those who declined to participate in Trial 2 (blue line, $n = 57$) as compared with the participants (pink line, $n = 96$). No significant difference can be observed.

With the Cox proportional hazards model adjusted for gender, age, smoking history, clinical stage and PS, the hazard ratio of participants *vs* non-participants was 0.965 (95% confidence interval: 0.73–1.28, $P = 0.805$). Among the patient characteristics, PS was the only significant factor associated with OS in multivariate analysis ($P = 0.006$, by Cox proportional model).

DISCUSSION

It has been argued that trial participants have better outcomes than those who are not enroled in clinical trials. Several investigations have reported a favourable overall trend with trial entry (Braunholtz *et al*, 2001; Peppercorn *et al*, 2004; West *et al*, 2005). This 'trial effect' could derive from several factors, such as protocol effect (the way treatments are delivered), care effect (extra care related to data gathering), Hawthorne effect (changes in doctor or patient behaviour on the basis of the knowledge that they are under observation) or placebo effect (psychologically mediated benefits) (Braunholtz *et al*, 2001; Peppercorn *et al*, 2004).

In majority of the reports comparing outcomes between participants and non-participants of clinical trials, however, the