The Korean Journal of Pathology 2011; 45: 45-52
DOI: 10.4132/KoreanJPathol.2011.45.1.45

ERCC1 Predicts a Poorer Platinum-based Chemotherapy Outcome but a Better Outcome for Uracil-Tegafur in the Resected Stage I-II NSCLC

Han Suk Ryu[1] · Xianhua Xu[1]
Hyojin Kim[1] · Jong-Suk Lee[2]
Sanghoon Jheon[3] · Jin-Haeng Chung[1,4]

Departments of [1]Pathology and [2]Thoracic Surgery, [3]Division of Oncology, Department of Internal Medicine, [4]Tumor Immunity Medical Research Center, Seoul National University Bundang Hospital, Seoul National University College of Medicine, Seongnam, Korea

Received: October 25, 2010
Accepted: November 3, 2010

Corresponding Author
Jin-Haeng Chung, M.D.
Department of Pathology, Seoul National University Bundang Hospital, Tumor Immunity Medical Research Center, Seoul National University College of Medicine, 300 Gumi-dong, Bundang-gu, Seongnam 463-707, Korea
Tel: +82-31-787-7713
Fax: +82-31-787-4012
E-mail: chungjh@snu.ac.kr

Background: The role of excision repair cross-complementation group 1 (ERCC1) has been controversial in non-small cell lung cancer (NSCLC) patients who received adjuvant chemotherapy with a platinum agent. We investigated ERCC1 expression in stage I-II NSCLC to clarify its significance for adjuvant chemotherapy. **Methods:** The ERCC1 expression profile was evaluated by immunohistochemistry and compared according to adjuvant chemotherapeutic agents in 146 patients who underwent surgical resection for stage I-II NSCLC. The patients were divided into 3 groups; adjuvant chemotherapy with a platinum based agent (18.5%, 27/146); adjuvant chemotherapy with uracil-tegafur (UFT) (40.4%, 59/146); surgery-alone (41.1%, 60/146). **Results:** Nuclear ERCC1 expression was detected in 71.9% (105/146) of NSCLC and was significantly associated with a shortened survival period in the group 1 patients who received the platinum based regimen after surgery. The group 2 patients who received UFT showed the longest survival period, followed by the surgery-alone group (overall survival, $p=0.049$; disease-free survival [DFS], $p<0.001$). **Conclusions:** These results suggest that stage I-II NSCLC patients with ERCC1 expression experience a shorter DFS period with adjuvant chemotherapy with a platinum based regimen and may benefit from adjuvant chemotherapy with UFT, instead of platinum after surgery.

Key Words: Carcinoma, non-small-cell lung; ERCC1 protein, human; Chemotherapy, adjuvant

Lung cancer is the leading cause of cancer related death and its incidence is increasing.[1] Non-small-cell lung cancer (NSCLC) represents more than 80% of all lung cancers, and in its early stages surgery is the treatment of choice.[2] However, 30% to 70% of patients undergoing surgical resection develop recurrence and ultimately die of their disease.[3,4] Many clinical studies suggested that adjuvant cisplatin-based chemotherapy could yield an overall survival (OS) benefit.[5-7] Although evidence of the advantage of cisplatin-based adjuvant chemotherapy have been reported, there are still limitations in the therapeutic capacity of platinum-based adjuvant chemotherapy as well as serious side effects.[5,6]

Recent research on personalized treatment by selecting patients likely to respond to a particular chemotherapeutic regimen based upon biomarker expression may allow for improved treatment efficacy while avoiding unnecessary treatment side effects.[8] Translational research in some of the adjuvant trials has provided some information about biomarkers[9,10] and identification of the subsets of patients who might or might not benefit from adjuvant platinum-based chemotherapy.[11,12]

Excision repair cross complement group 1 (ERCC1) is an integral component of the nucleotide excision repair pathway that functions for repair of damaged DNA, which can be a cause of transformation of a normal cell to a cancer cell.[13] Therefore, *ERCC1* is considered as a keeper gene that prevents mutation through removal of damaged nucleotide segments and repair with a new one. On the other hand, since ERCC1 recognizes the platinum-DNA adduct and removes the damaged segment, it is regarded as a factor in reducing platinum-based chemotherapeutic effects on cancer cells of NSCLC, as well as that of other organs.[12,14]

Recently, a number of retrospective analyses confirmed that ERCC1 expression has adversely affected platinum-based chemotherapy in NSCLC,[11,15,16] although some negative reports highlighted the need for a more cautious interpretation due to the opposite result, namely that ERCC1 expression acts beneficially as a prognostic factor for patients who received a platinum agent as adjuvant chemotherapy.[17-19] In addition to this,

the use of adjuvant chemotherapy in early stages of NSCLC remains highly controversial.[20,21]

Uracil-tegafur (UFT) is an oral chemotherapeutic agent in which a pro-drug of 5-fluorouracil (5-FU), tegafur (FT), is combined with uracil as a dihydropyrimidine dehydrogenase (DPD)-inhibitor.[22] 5-FU is gradually released from FT and a certain 5-FU concentration can be maintained for a longer period as degradation of 5-FU is inhibited by a DPD inhibitor. Therefore, UFT is defined as an oral DPD-inhibitory fluoropyrimidine and has the clinical benefit of an anticancer effect but with a mild toxicity profile.

A number of Japanese adjuvant trials showed that a survival benefit was obtained with adjuvant chemotherapy using UFT in stage I NSCLC.[23,24] Based on these results, adjuvant chemotherapy might be useful beyond pathological stage I, which means that most patients with resected NSCLC might benefit from receiving adjuvant chemotherapy after surgery. These results raise the controversial issue of adjuvant chemotherapy in early stage NSCLC.[7]

To further examine these issues, we investigated the therapeutic benefit of adjuvant chemotherapy with UFT or a cisplatin-based agent in stage I-II NSCLC with ERCC1 expression.

MATERIALS AND METHODS

Patients and samples

A cohort of 146 consecutive patients who underwent surgical resection for stage I-II NSCLC from May 2003 to July 2006 was selected from the pathology archives of Seoul National University Bundang Hospital. The patients who received radiotherapy, neoadjuvant chemotherapy and who died related to other causes other than NSCLC were excluded. The remaining patients were divided into 3 groups: 1) adjuvant chemotherapy with platinum-doublet (cisplatin-gemcitabine regimen: gemcitabine 1,000 mg/m^2 on days 1 and 8; cisplatin 75 mg/m^2, Q 4 weeks) (18.5%, 27/146); 2) adjuvant chemotherapy with UFT (40.4%, 59/146) at a dose of 250 mg/m^2 twice a day during two years; and, 3) surgery-alone (41.1%, 60/146) (Table 1). There were 49 men and 97 women with a median age of 62.6 years (range, 36 to 81 years). Using the 2004 World Health Organization (WHO) classification, there were 90 adenocarcinomas (ADC, 61.6%) and 56 squamous cell carcinomas (SCC, 38.4%). Clinicopathologic information (including sex, age, smoking history, histologic subtype, tumor size, performance status, 7th tumor, node and metastasis [TNM] stage, and history of adjuvant chemotherapy) was retrieved by review of medical records. The median follow-up period for all patients was 47.4 months (range, 5 to 79 months). This study was approved by the Institutional Review Board of Seoul National University Bundang Hospital.

Immunohistochemical analysis

Four-micrometer sections from the formalin-fixed, paraffin-embedded tissue specimens were deparaffinized in xylene and rehydrated in graded ethanol. The succeeding steps were performed automatically at 37°C using the Benchmark XT Slide Staining System Specifications (Ventana Medical Systems, Tucson, AZ, USA). Antigen retrieval was performed by immersing slides in citrate buffer (pH 6.0) for 15 minutes and endogenous peroxidases were blocked with 1% H_2O_2 for 4 minutes. The sections were incubated with primary monoclonal antibody to ERCC1 (1 : 100, mouse, clone8F1, Neomarkers, Fremont, CA, USA) for 60 minutes at room temperature. To visualize the immunostaining, Ultravision LP kit (Lab Vision, Fremont, CA, USA) was used. The slides were stained using a diaminobenzidine detection kit and counterstained with haematoxylin. Positive controls included normal bronchiolar epithelial cells and stromal cells surrounding the tumor area for ERCC1 expression.

Evaluation of immunohistochemical analysis of ERCC1

The expression of ERCC1 was assessed semi-quantitatively by two pathologists (H.S.R and J.H.C) who were unaware of the clinicopathologic information of the patients. Semi-quantitative assessment was done by estimating the staining intensity and percentage of tumor cells with positive nuclear staining as previously described.[12] Briefly, each tumor cell was first scored from 0 to 3, which corresponded to negative, weak, moderate and strong intensities, respectively. Then, percentages of positively stained cells were counted (0-100%) and scores were as-

Table 1. Treatment characteristics of NSCLC patients (n = 146)

	Post-operative adjuvant treatment		
	Surgery-alone	Platinum doublet	UFT
7th TNM stage			
I (n = 98)	46 (46.9)	9 (9.2)	43 (43.9)
II (n = 48)	14 (29.7)	18 (38.3)	16 (34.0)
Total (n = 146)	60 (41.1)	27 (18.5)	59 (40.4)

Values are presented as number (%).
NSCLC, non-small cell lung cancer; TNM, tumor, node and metastasis; UFT, uracil-tegafur.

signed according to the proportion (0 if 0%, 0.1 if 1% to 9%, 0.5 if 10% to 49%, and 1.0 if 50% or more). The proportion score was multiplied by the staining intensity to obtain a histochemical score (H-score). Finally, each score was divided into ERCC1-positive groups and ERCC1-negative groups based on the median value of the H-score 1 (H-score ≥ 1 [positive] vs H-score < 1 [negative]) (Fig. 1).

Statistical analysis

The Pearson χ^2 test was performed to evaluate the association of clinicopathologic parameters with the status of ERCC1 expression. In order to evaluate how ERCC1 affects patients depending on modality of adjuvant chemotherapy, the median duration of disease-free survival (DFS) and OS were analyzed using the Kaplan-Meier method with the survival rates and using a stratified Cox-Mantel log-rank test for the status of ERCC1 expression. Categorical variables which proved to be significant prognostic factors in univariate analysis with Kaplan-Meier method became candidates for a multivariate logistic regression test to determine the interaction of multiple factors related to survival. Multivariate logistic regression analysis was carried out using a Cox proportional hazards model, depending on ERCC1 expression, and categorical variables were adjusted to identify significant prognostic factors for OS and DFS. Mean survival times and 95% confidence intervals (CI) are presented in both the univariate and multivariate logistic regression analysis. All reported p-values are two sided. p-values of less than 0.05 were considered significant. All analyses were performed using SPSS ver. 16.0 (SPSS Inc., Chicago, IL, USA).

RESULTS

ERCC1 immunoreactivity according to clinicopathologic parameters and survival analysis

ERCC1 expression was confined to the nuclei of tumor cells and to the bronchiolar epithelial cells. The median percentage of tumor cells that stained with monoclonal ERCC1 antibody was 26% (range, 0 to 100%). An H-score of 1.0 was used as a cut-off value. ERCC1 was positive in 71.9% (105/146) of the stage I-II NSCLC, specifically 68.8% (62/90) of ADC and 76.8% (43/56) of SCC (p = 0.302). According to the 7th TNM staging system, 67.2% (98/146) of NSCLC were stage I and 32.8% (48/146) stage II. Pearson's χ^2 test showed no significant correlation between the expression of ERCC1 and TNM stage, sex, age, smoking history, tumor size, pleural invasion, histologic type and performance status (Table 2).

Univariate survival analysis showed that some clinicopathologic parameters, including age, pleural invasion, TNM stage and performance status had significant prognostic implications

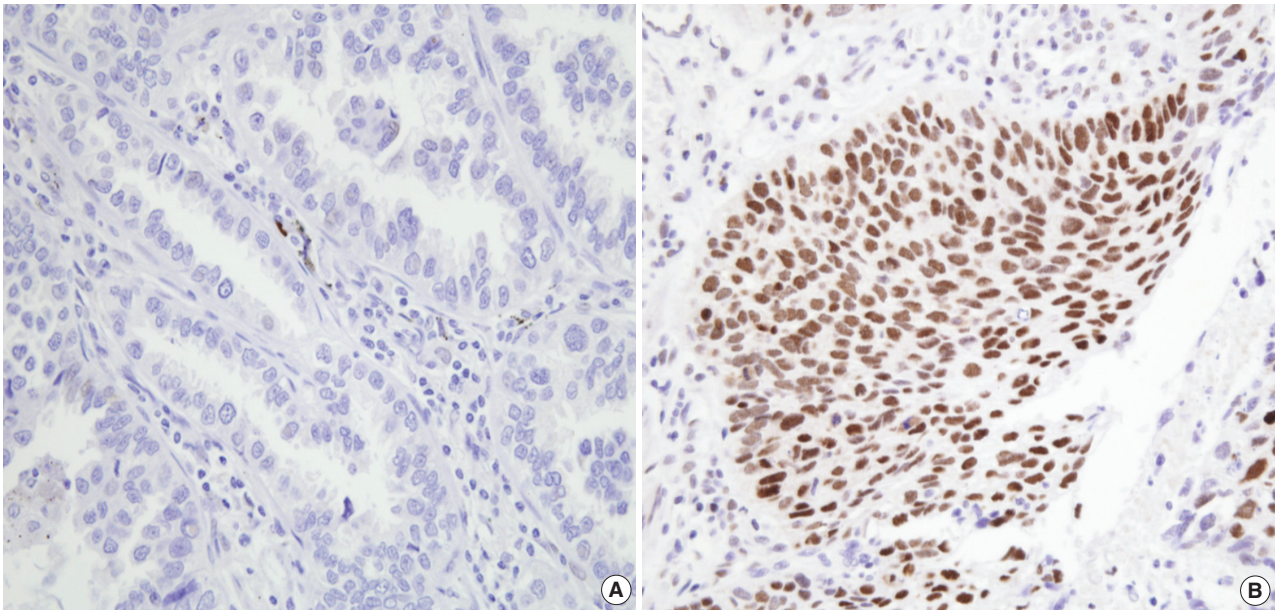

Fig. 1. Representative excision repair cross-complementation group 1 (ERCC1) expression (intranuclear staining) by immunohistochemistry. (A) An ERCC1 negative (histochemical score [H-score] = 0) non-small cell lung cancer (NSCLC, adenocarcinoma). (B) An ERCC1 positive (H-score = 1) NSCLC (squamous cell carcinoma).

for disease free or OS (Table 3).

DFS according to adjuvant chemotherapy use in stage I-II NSCLC

The mean DFS time for patients who received UFT, a platinum agent and surgery-alone was 63.6 months, 43.1 months and 57.5 months, respectively ($p<0.001$). Patients who received platinum-based chemotherapy had an increased hazard ratio (HR) for tumor recurrence compared to the other two groups who were treated with surgery alone or with the UFT regimen (adjusted HR, 1.242; 95% CI, 1.112 to 2.285; $p=0.026$). Patients who were treated with the UFT regimen demonstrated a lower risk rate for tumor recurrence than the other two groups treated with surgical resection only or platinum-based chemotherapy (adjusted HR, 0.330; 95% CI, 0.132 to 0.825; $p=0.018$). However, adjuvant chemotherapy was not identified as an independent prognostic factor for OS (Table 4).

Survival analyses according to adjuvant chemotherapy in the ERCC1 positive group

Among the patients who were ERCC1-positive NSCLC (n=105), group 2 patients who received UFT-based chemotherapy

Table 2. Characteristics of patients and tumors according to ERCC1 expression of the patients (n=146)

	Total (n=146)	Patients with ERCC1 expression		p-value
		Positive (n=105)	Negative (n=41)	
Sex				
Male	49	35 (33.3)	14 (34.1)	0.926
Female	97	70 (66.7)	27 (65.9)	
Age (yr)				
≤65	79	58 (55.2)	21 (51.2)	0.661
>65	67	47 (44.8)	20 (48.8)	
Smoking				
Yes	87	63 (60.0)	24 (58.5)	0.871
No	59	42 (40.0)	17 (41.5)	
Tumor size (cm)				
≤3	96	72 (68.6)	24 (58.5)	0.251
>3	50	33 (31.4)	17 (41.5)	
Pleural invasion				
No	106	79 (75.2)	27 (65.9)	0.463
Visceral pleura	38	25 (23.8)	13 (31.7)	
Parietal pleura	2	1 (1.0)	1 (2.4)	
7th TNM stage				
Stage I	98	73 (69.5)	25 (61.0)	0.478
Stage II	48	32 (30.5)	16 (39.0)	
Histologic type				
ADC	90	62 (59.0)	28 (68.3)	0.302
SCC	56	43 (41.0)	13 (31.7)	
Performance status				
0	99	72 (68.6)	27 (65.9)	0.263
1	37	28 (26.6)	9 (22.0)	
2	10	5 (4.8)	5 (12.2)	

Values are presented as number (%).
ERCC1, excision repair cross-complementation group 1; TNM, tumor, node and metastasis; ADC, adenocarcinomas; SCC, squamous cell carcinomas.

Table 3. Univariate analysis results of disease free survival and overall survival in 146 patients with NSCLC

	Disease free survival (p-value)	Overall survival (p-value)
Age (≤65 yr vs >65 yr)	0.091	**0.004**
Histologic type (ADC vs SCC)	0.373	0.777
Tumor size (≤3 cm vs >3 cm)	0.819	0.287
Pleural invasion (N, VP, PP)	**0.003**	**0.001**
7th TNM stage (I vs II)	**0.009**	0.417
ERCC1 expression (positive vs negative)	0.609	0.476
Chemotherapy (SA, platinum, UFT)	**<0.001**	0.253
Performance status (0, 1, 2)	**0.010**	**0.002**

NSCLC, non-small cell lung cancer; ADC, adenocarcinoma; SCC, squamous cell carcinoma; N, no pleural invasion; VP, visceral pleural invasion; PP, parietal pleural invasion, TNM, tumor, node and metastasis; ERCC1, excisional repair cross-complementation group 1; SA, surgery alone, UFT, uracil-tegafur.

Table 4. Disease free survival and overall survival in multivariate analysis (Cox regression, n=146)

	Disease free survival			Overall survival		
	HR	95% CI	p-value	HR	95% CI	p-value
Age (≤65 yr vs >65 yr)	1.373	0.796-2.368	0.254	6.671	2.305-21.703	**0.002**
Tumor size (≤3 cm vs >3 cm)	2.132	1.157-3.928	**0.015**	2.041	0.751-5.549	0.162
Pleural invasion (N, VP, PP)	1.698	1.115-2.588	**0.014**	2.327	1.262-4.290	**0.007**
7th TNM stage (I vs II)	2.142	1.601-2.866	**<0.001**	2.752	1.741-4.350	**<0.001**
ERCC 1 expression (0 vs 1)	0.851	0.499-1.452	0.554	0.456	0.198-1.052	0.066
Chemotherapy (NAC, platinum, UFT)			**0.010**			0.254
Surgery-alone vs platinum	1.242	1.112-2.285	**0.026**	1.202	0.855-2.039	0.110
Surgery-alone vs UFT	0.330	0.132-0.825	**0.018**	0.536	0.132-2.178	0.536
Performance status (0, 1, 2)	1.706	1.191-2.443	**0.004**	3.970	2.220-7.100	**<0.001**

HR, adjusted hazard ratio; CI, confidence interval; N, no pleural invasion; VP, visceral pleural invasion; PP, parietal pleural invasion; TNM, tumor, node and metastasis; ERCC1, excisional repair cross-complementation group 1; NAC, neoadjuvant chemotherapy; UFT, uracil-tegafur.

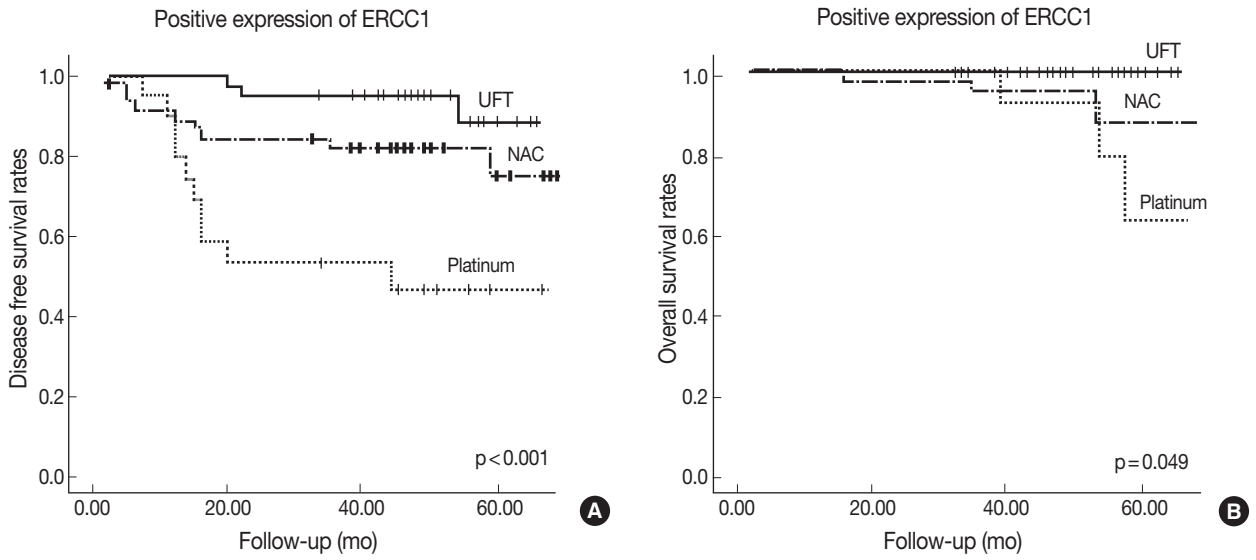

Fig. 2. Kaplan-Meier survival analysis according to adjuvant chemotherapeutic modality in excision repair cross-complementation group 1 (ERCC1) expressing non-small cell lung cancer. (A) Overall survival of patients in the positive-ERCC1 expression group (p=0.049). (B) Disease-free survival of patients in positive ERCC1-expression group (p<0.001).
UFT, uracil-tegafur; NAC, neoadjuvant chemotherapy.

Table 5. Univariate analysis results of disease free survival and overall survival in 146 patients with NSCLC (stratified Cox-Mantel log-rank tests for the status of ERCC1 expression)

	ERCC1 positive (p-value) (n=105)		ERCC1 negative (p-value) (n=41)	
	Disease free survival	Overall survival	Disease free survival	Overall survival
Age (≤65 yr vs >65 yr)	0.034	0.001	0.476	0.042
Histologic type (ADC vs SCC)	0.199	0.083	0.645	0.894
Tumor size (≤3 cm vs >3 cm)	<0.001	0.046	<0.001	0.002
Pleural invasion (N, VP, PP)	0.061	0.006	0.001	0.002
7th TNM stage (I vs II)	<0.001	<0.001	<0.001	<0.001
Chemotherapy (SA, platinum, UFT)	<0.001	0.049	0.075	0.719
Performance status (0, 1, 2)	0.035	0.028	0.129	<0.001

NSCLC, non-small cell lung cancer; ERCC1, excisional repair cross-complementation group 1; ADC, adenocarcinoma; SCC, squamous cell carcinoma; N, no pleural invasion; VP, visceral pleural invasion; PP, parietal pleural invasion, TNM, tumor, node and metastasis; SA: surgery alone, UFT, uracil-tegafur.

Table 6. Multivariate analysis in earlier-stage (TNM stage I & II) NSCLC patients with increased ERCC1 expression (n=105)

	Disease free survival			Overall survival		
	HR	95% CI	p-value	HR	95% CI	p-value
Age (≤65 yr vs >65 yr)	2.792	0.975-8.521	0.056	1.854	0.542-4.368	0.524
Tumor size (≤3 cm vs >3 cm)	1.777	0.558-5.653	0.330	1.565	0.345-3.327	0.371
Pleural invasion (N, VP, PP)	2.145	0.981-4.691	0.056	2.930	1.121-6.102	**0.030**
7th TNM stage (I vs II)	1.598	0.488-5.234	0.439	1.101	0.174-3.356	0.470
Chemotherapy (NAC, platinum, UFT)			**0.021**			0.329
Surgery-alone vs platinum	2.593	1.109-7.396	**0.008**	0.924	0.185-6.512	0.559
Surgery-alone vs UFT	0.324	0.085-0.924	**0.035**	2.331	0.930-5.217	0.091
Performance status (0, 1, 2)	1.093	0.513-2.327	0.818	0.210	0.081-3.041	0.420

TNM, tumor, node and metastasis; NSCLC, non-small cell lung cancer; ERCC1, excisional repair cross-complementation group 1; HR, adjusted hazard ratio; CI, confidence interval; N, no pleural invasion; VP, visceral pleural invasion; PP, parietal pleural invasion; NAC, neoadjuvant chemotherapy; UFT, uracil-tegafur.

demonstrated significant prolonged DFS compared to the other groups treated with platinum-based chemotherapy or surgery-alone (p<0.001) (Fig. 2A, Table 5). OS was also significantly longer in the UTF-treated group than the other two groups (p=

0.049) (Fig. 2B, Table 5). Furthermore, the Cox proportional analysis for DFS showed that UFT was a beneficial adjuvant chemotherapeutic agent (adjusted HR, 0.324; 95% CI, 0.085 to 0.924; p=0.035). Platinum-based chemotherapy had an adverse effect on DFS in stage I-II NSCLC with ERCC1 expression (adjusted HR, 2.593; 95% CI, 1.109 to 7.396; p=0.008) (Table 6). By contrast, OS analysis did not show adjuvant chemotherapeutic agents to be a significant prognostic variable (Table 6). Statistical significance also remained limited for DFS and OS in patients with ERCC1.

DISCUSSION

Our results demonstrate that increased expression of ERCC1 predicts a poorer platinum-based chemotherapy outcome in Korean patients with stage I-II NSCLC. On the other hand, a single compound of a UFT agent showed DFS benefits when used as postoperative adjuvant chemotherapy compared to surgery alone, and cisplatin use did not confer an advantage over surgery alone in ERCC1-positive tumors. These results suggest that UFT might be a candidate agent for adjuvant chemotherapy in stage I-II NSCLC patients with increased ERCC1 protein.

Currently, in order to improve survival in patients with resected NSCLC, numerous adjuvant therapeutic regimens have been examined in clinical trials. Pooled analysis of large trials of cisplatin-based adjuvant chemotherapy in patients with NSCLC conducted by the Lung Adjuvant Cisplatin Evaluation Collaborative Group showed that adjuvant cisplatin-based chemotherapy significantly reduced the risk of postoperative death with a HR of 0.89 (p<0.005).[21] The analysis also demonstrated that adjuvant cisplatin-based chemotherapy improved survival in patients with completely resected NSCLC, especially in stage II and III.[21] On the other hand, cisplatin-based chemotherapy showed either a reverse effect or no effect on earlier-stage patients (HR, 1.40 and 0.93). In addition, cisplatin-based chemotherapy in NSCLC has a reported mortality rate of therapy-related toxicity of 1.7% (3/840). This mortality rate caused by adjuvant platinum-based chemotherapy has been similarly reported in other major trials.[6,7] Although cisplatin is still being considered as a powerful tool in terms of survival benefit in patients with advanced NSCLC, attention should be paid to the ambiguous survival benefit and risk of drug toxicity on earlier-stage patients.

Adjuvant chemotherapy after complete tumor resection has been controversial, especially in early-stage NSCLC patients.[25] Over the past 10 years, the results of well-designed randomized controlled trials comparing adjuvant chemotherapy with lower anti-tumor effect and milder toxicity after surgery have been released, and UFT has been considered as a useful adjuvant chemotherapeutic agent following complete tumor resection, especially for earlier-stage NSCLC.[24,26] UFT, a 5-FU derived agent is phosphorylated into active metabolites and kills tumor cells through inhibition of thymidylate synthase, which is a key enzyme in *de novo* DNA synthesis.[22]

Since the first remarkable trial by the West Japan Study Group for Lung Cancer Surgery showing a significant survival improvement in the UFT-alone group as compared with that of surgery-alone group for resected p-stage I-III NSCLC was reported,[26] the efficacy of postoperative adjuvant UFT treatment for earlier-stage NSCLC patients has been confirmed in a series of analyses.[24,27] In a randomized trial of adjuvant chemotherapy with UFT for a total of 999 patients with resected p-stage I (T1 or T2) adenocarcinoma, the survival benefit of adjuvant UFT administration was not documented in stage 1A (T1) but was observed in stage IB (T2).[24] However, Tsuboi *et al*.[28] documented a significant survival benefit of adjuvant UFT chemotherapy even in stage IA patients when the tumor diameter was larger than 2 cm in their exploratory analysis. These results suggest that there is a survival benefit of UFT treatment in earlier stage NSCLC, and that UFT can be recommended for p-stage IB patient or IA patients with tumor size of 2 cm or greater.

When stratified to ERCC1 positive and ERCC1 negative tumors, we demonstrated a DFS benefit in the postoperative UFT chemotherapy group compared to the surgery-alone group in patients with ERCC1 positive tumors (HR, 0.324; 95% CI, 0.085 to 0.924; p=0.035). On the other hand, the cisplatin-based chemotherapy group had a shortened DFS period when compared with the surgery-alone group in stage I-II NSCLC with increased ERCC1 expression (HR, 2.593; 95% CI, 1.109 to 7.396; p=0.008). These findings suggest that cisplatin-based chemotherapy in ERCC1-positive NSCLC might lead to adverse effects on DFS, as demonstrated in several studies regarding the prognostic impact of ERCC1.[11,12]

Although our results showed a survival benefit of UFT treatment in patients with ERCC1 expression, the functional mechanism related to UFT in NSCLC with ERCC1 expression has not been unveiled. This is in contrast to the relationship between ERCC1 expression and the cisplatin-based chemotherapeutic effect that has been analyzed in the previous studies of NSCLC.[29] Shirota *et al.*[30] showed that thymidylate synthase (TS) and ERCC1 mRNA expression levels were independent predictive mar-

kers of survival when evaluating the efficacy of 5-FU and platinum-based combination chemotherapy in advanced colorectal cancer, and they suggested a certain relationship between the two markers as well as an interaction between 5-FU and platinum-based chemotherapeutic agents. In our opinion, UFT inhibits the action of TS, which is essential for *de novo* DNA synthesis in tumor cells, and ERCC1 is also considered to be involved in the process of DNA synthesis as a function of repair; thus, further evaluation of the relationship between these two proteins and chemotherapeutic agents is still needed in in studies of NSCLC.

In the present study, we investigated ERCC1 expression by immunohistochemistry. Although measurement of the mRNA level has been widely used for ERCC1 detection in NSCLC,[18-20] the results among the studies have been variable. As mRNA measurement from whole tissue includes its expression in the non-neoplastic tissue as well as the tumor tissue, ERCC1 mRNA levels may be limited for predicting the efficacy of platinum-based chemotherapy.[18] On the other hand, the morphologic discrimination of normal and tumor cells is feasible and normal tissue included in the test material could be excluded by immunohistochemistry. Further studies are required to evaluate treatment outcomes using various laboratory techniques for ERCC1 expression.

There are some limitations in our study. First, it was not possible to evaluate OS according to ERCC1 expression due to the small number of patients and relatively short follow-up period. For this reason, we used the mean-survival period duration instead of median-survival period. However, these limitations of our study could be resolved by ongoing long-term follow up, which will enable us to demonstrate more evidences for the relation between ERCC1 expression and the effect of adjuvant chemotherapy. Second, we could not demonstrate any significant differences between the 3 groups in ERCC1-negative NSCLC due to the small number of patients. A further large-scaled study is needed.

In conclusion, our study demonstrates that increased ERCC1 expression was associated with a shortened DFS period in patients treated with cisplatin-based chemotherapy in stage I-II NSCLC. However, patients receiving UFT treatment showed a DFS benefit after tumor resection. Based on these results, increased ERCC1 protein expression might be considered as a predictive marker for an improved outcome concerning the choice of adjuvant chemotherapy in stage I-II NSCLC.

ACKNOWLEDGMENTS

Supported by Korean Science & Engineering Foundation (KOSEF) through the Tumor Immunity Medical Research Center at Seoul National University College of Medicine and partly supported by a grant of the Korea Healthcare Technology R&D project, Ministry of Health & Welfare, Republic of Korea (A08-4578).

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