RESEARCH ARTICLE

Expression of ERCC1, MSH2 and PARP1 in Non-small Cell Lung Cancer and Prognostic Value in Patients Treated with Platinum-based Chemotherapy

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Abstract

Purpose: To evaluate the prognostic value of the expression of excision repair cross-complementation group l (ERCC1), MutS protein homolog 2 (MSH2) and poly ADP-ribose polymerase 1 (PARP1) in non-small-cell lung cancer patients receiving platinum-based postoperative adjuvant chemotherapy. Methods: Immunohistochemistry was applied to detect the expression of ERCC1, MSH2 and PARP1 in 111 cases of non-small cell lung cancer paraffin embedded surgical specimens. Through og-rank survival analysis, we evaluated the prognostic value of the ERCC1, MSH2, PARP1 and the related clinicopathological factors. COX regression analysis was used to determine whether ERCC1, MSH2 and PARP1 were independent prognostic factors. Results: In the enrolled 111 non-small cell lung cancer patients, the positive expression rate of ERCC1, MSH2 and RARP1 was 33.3%, 36.9% and 55.9%, respectively. ERCC1 (P<0.001) and PARP1 (P=0.033) were found to be correlated with the survival time while there was no correlation for MSH2 (P=0.298). Patients with both ERCC1 and PARP1 negative cancer had significantly longer survival time than those with ERCC1 (P=0.042) or PARP1 (P=0.027) positive alone. Similalry, the survival time of patients with both ERCC1 and PARP1 positive cancer was shorter than those with ERCC1 (P=0.048) or PARP1 (P=0.01) positive alone. Conclusion: Patients with ERCC1 or PARP1 negative non-small cell lung cancer appear to benefit from platinum-based postoperative adjuvant chemotherapy.

Keywords: DNA repair gene - ERCC1 - MSH2 - RARP1 - non-small cell lung cancer - prognosis - chemotherapy

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Introduction

Lung cancer is one of the most common malignances, of which the occurrence and mortality is increasing every year due to air pollution, environmental breakdown and cigarette abuse (Jemal et al., 2005; 2006). Non-small cell lung cancer (NSCLC) is the main type of lung cancer, accounting for about 80-85%, and is the leading reason of death caused by lung cancer (Parkin et al., 2005). Surgical treatment is generally considered curative for early-stage NSCLC. However, more than 60% patients suffer local or distant recurrence of NSCLC at early time after curative resection (Jassem et al., 2000). Platinum-based adjuvant chemotherapy is mostly used for these patients. Studies presented that the platinum-based chemotherapy can decrease mortality by 11% and improve five year survival by 5.4% (Pignon et al., 2008). However, patients respond differently to platinum drugs (Bahland Falk, 2001). After administration, the platinum drug binds to DNA in cancer cells and forms Pt-DNA complexes which result in crosslinking in a DNA strand or between strands, causing damages to DNA replication and thereby inhibiting growth of cancer cells. At the other hand, decreased drug accumulation in cells, increased resistance to Pt-DNA complexes and stronger capability of DNA repair can reduce the efficacy of platinum drugs, among which the DNA repair pathways contribute most (Rosell et al., 2002).

Repair of DNA damage is a protection system in cells to correct damages to DNA molecules involving multiple complexes. In theory, cells that have less efficient DNA repair mechanisms are most prone to undergo cell death, whereas those with efficient mechanisms would prove resistant to the agent of insult. Multiple DNA repair pathways have already been confirmed to be associated with tumor prognosis, drug efficacy or chemotherapeutic resistance, including base excision repair (BER), mismatch repair (MMR), translesion synthesis, nucleotide excision repair (NER) and homologous recombination (HR) (Scartozzi et al., 2006; Wang et al., 2009).

Excision Repair Cross-Complementation group l (ERCC1), an important factor in NER pathway, has been found to be related to the response to platinum- or cisplatin- based chemotherapy in NSCLCs (Olaussen et al., 2006; Zhang et al., 2012; Li et al., 2013). MutS protein

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homolog 2 (MSH2) in MMR pathway is related to DNA repair after platinum insult. A lower level or no expression of MSH2 is reported to be a predictor of better prognostic and a better predictive value can be achieved when combined with ERCC1 (Kamal et al., 2010; Pierceall et al., 2012). Poly ADP-ribose polymerase (PARP), involved in BER pathway, is also demonstrated to be associated with resistance to platinum drugs and prognosis in NSCLC patients (Kummar et al., 2012).

It is proposed that DNA repair pathways affect the resistance of NSCLC patients to platinum-based chemotherapy. However, proteins that can serve as biomarkers for prognosis after chemotherapy are yet to be established. Furthermore, there are few literatures focusing on the synergic effect of proteins involved in different DNA repair pathways. In our study, expression of ERCC1, MSH2 and PARP1 in tumor specimens from NSCLC patients were quantified by immunochistochemistry. We then assessed the relationships between the expression of ERCC1, MSH2 and PARP1 and clinical variables, including gender, age, smoking history, histologic type, TNM stage and tumor size, in NSCLC patients. Further, the prognostic values of these proteins were evaluated in NSCLC patients treated with platinum-based postoperative adjuvant therapy.

Materials and Methods

Subjects

NSCLC patients that received curative resection at People's Hospital in Shaoxing City, Zhejiang Province, China, from January 2008 to April 2010 were recruited for this study. Patients who had received radiotherapy or chemotherapy before resection or suffered from other kinds of tumor other than NSCLC were excluded. The histological classification was based on a WHO report. Clinical staging was determined by the current International Staging System based on an initial evaluation that comprised a clinical assessment, chest X-ray, computed tomography of the chest and abdomen, computed tomography or magnetic resonance imaging of the brain, and bone scintigraphy. A total of 111 patients with adequate cancer specimens collected before chemotherapy were enrolled in this study. The clinicopathological characteristics of all the patients are listed in Table 1. Their median age at diagnosis was 63 years (range, 43-81 years).

Immunohistochemistry

Immunohistochemical staining was performed on a series of 4 μm formalin-fixed, paraffin-embedded tissue sections. The slides were deparaffinized in xylene and dehydrated in a graded ethanol series. For antigen retrieval, the slides were immersed in 10 mm citric buffer solution (pH 6.0) and heated to 125˚C by exposed to autoclave irradiation for 2 min. The slides were then cooled at room temperature and washed in water and PBS. Nonspecific binding was blocked by preincubation with 2% BSA plus 0.1% NaN3 for 30 min. After draining off of the blocking solution, each slide was incubated overnight at 4˚C with 50 μl primary antibodies for ERCC1 (dilution 1:800), MSH2 (dilution 1:300) and PARP1 (dilution 1:300). All the primary antibodies were obtained from Biorbyt, England. Staining with PBS instead of primary antibodies was routinely performed as a negative control procedure. After washed twice with PBS, the slides were incubated with a second antibody (DAKO, Glostrup, Denmark) for 30 min. The 2% 3,3'-diaminobenzidine in 50 mM Tris buffer (pH 7.6) containing 0.3% hydrogen was used as chromogen. Slides were counterstained with hematoxylin. All of the slides were examined and scored according to the following rule independently by two observers without any knowledge of the clinical data of the patients. Staining intensity was scored as below: similar to background was defined as zero, pale yellow as 1, clay bank as 2 and sepia as 3. Percent of cells with positive staining per 400 tumor cells was scored as below: less than 10% was defined as zero, 0-10% as 1, 1-50% as 2, 51-75% as 3 and more than 75% as 4. This percentage score was multiplied by the score of staining intensity to obtain a final semiquantitative score. Slides with final score less than 4 was considered as negative expression while larger than 4 as positive.

Statistical analysis

The correlations between protein expression and the clinical parameters as well as the correlations between them and response to chemotherapy were evaluated by the χ^2 test or Fisher's exact test. Overall survival was measured from the start of chemotherapy to the date of death from any cause or the date the patient was last known to be alive. Survival curves were estimated by the Kaplan–Meier method. The Cox proportional hazards model was used for multivariate analysis. A p value <0.05 was considered significant. All the statistical analyses were performed in the software SPSS 20.0.

Results

Expression of ERCC1, MSH2 and PARP1 in NSCLC

Thirty-seven (33.3%) of the 111 NSCLC patients were ERCC1-positive, 41 (36.9%) were MSH2-positive and 62 (55.9%) were PARP1-positive (Table 1). Representative negative and positive staining is shown in Figure 1.

Table 1. Expression of ERCC1, MSH2 and PARP1 in NSCLC

Group	N	ERCC1		MSH2		PARP1	
		Negative	Positive	Negative	Positive	Negative	Positive
Squamous cell carcinoma	43	32	12 (27.9%)	23	20 (46.5%)	18	25 (58.1%)
Adenocarcinoma	56	34	22 (39.3%)	37	19 (33.9%)	26	30 (53.6%)
Other	12	9	3 (25.0%)	10	2 (16.7%)	5	7 (58.3%)
Total	111	74	37 (33.3%)	70	41 (36.9%)	49	62 (55.9%)

Correlation between the expression of ERCC1, MSH2 and PARP1 and the clinical variables in NSCLC patients

The relationships between clinical variables and expression of ERCC1, MSH2 and PARP1 were assessed in this study and shown in Table 2. No correlation was observed between gender, age, smoking history and expression of all the three proteins studied here. There were 56 cases of adenocarcinoma (ADC), 43 of squamous cell carcinoma (SCC) and 12 other histologic types among the NSCLC patients. Also, no correlation was detected between the histologic type and the expression of all the three proteins. Similarly, three was no correlation between expression of ERCC1, MSH2 and PARP1 and TNM stage. But, the expression of PARP1 was found to be associated with T staging (p=0.027, χ^2 test).

Correlation between the clinical variables and the outcomes of NSCLC patients after platinum-based chemotherapy

Among the 111 NSCLC patients, the survival data of 72 patients who received platinum-based chemotherapy were available. The relationships between the clinical variables and the outcomes in NSCLC patients are shown in Table 3. No correlation was observed between the median survival time and some clinical parameters, such as gender, age, smoking history and histologic type. In contrast, TNM stage (p=0.004), T stage (p=0.006), N

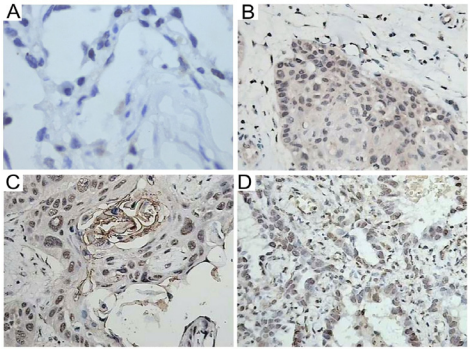

Figure 1. Representative Immunohistochemical Staining in NSCLC Specimen (40×). A) negative control; B) ERCC1; C) MSH2; D) PARP1

Table 3. Summary of Relationship between Clinical Variables and Survival

Variables		N	median survival time	chi-square	P value
Gender					
	Male	49	39	0.848	0.357
	Female	23	44		
Age					
	<60	23	45	2.024	0.155
	≥60	49	38		
Smoking History					
	<400	32	42	0.125	0.43
	≥400	40	39		
Type					
	adenocarcinoma	31	45	2.733	0.724
	squamous cell carcinoma	33	38		
	Other	8	31		
TNM Stage					
	I	25	50	10.833	0.004
	II	25	40		
	III+IV	22	28		
T Stage					
	T1	18	54	10.074	0.006
	T2	24	41		
	T3+T4	30	32		
N Stage					
	N0	41	47	10.939	0.012
	N1	18	32		
	N2	11	31		
	N3	2	15		
M Stage					
	M0	70	48	4.526	33
	M1	2	21		

Table 2. Relationship between Clinical Variables and Expression of ERCC1, MSH2 and PARP1

		N	ERCC1			MSH2			PARP1		
			Negative	Positive	P value	Negative	Positive	P value	Negative	Positive	P value
Gender	Male	73	50	23	0.573	46	27	0.988	31	42	0.622
	Female	38	20	14		24	14		18	20	
Age	<60	33	21	12	0.66	24	9	0.17	17	16	0.309
	≥60	78	53	25		46	32		32	46	
Smoking History	<400	57	40	17	0.43	38	19	0.438	23	34	0.408
	≥400	54	34	20		32	22		26	28	
Type	ADC	56	34	22	0.399	37	19	0.134	26	30	0.887
	SSC	43	31	12		23	20		18	25	
	Other	12	9	3		10	2		5	7	
TNM Stage	I	49	32	17	0.641	30	19	0.886	26	23	0.22
	II	30	22	8		20	10		12	18	
	III+IV	32	20	12		20	12		11	21	
T Stage	T1	42	29	13	0.722	28	14	0.678	25	17	0.027
	T2	25	15	10		14	11		7	18	
	T3+T4	44	30	14		28	16		17	27	
N Stage	N0	68	49	19	0.208	44	24	0.833	30	35	0.636
	N1	25	15	10		14	11		10	15	
	N2	14	9	5		9	5		5	9	
	N3	4	1	3		3	1		1	3	
M Stage	M0	109	73	36	0.614	68	41	0.275	51	58	0.928
	M1	2	1	1		2	0		1	1	

*ADC, adenocarcinoma; SCC, squamous cell carcinoma

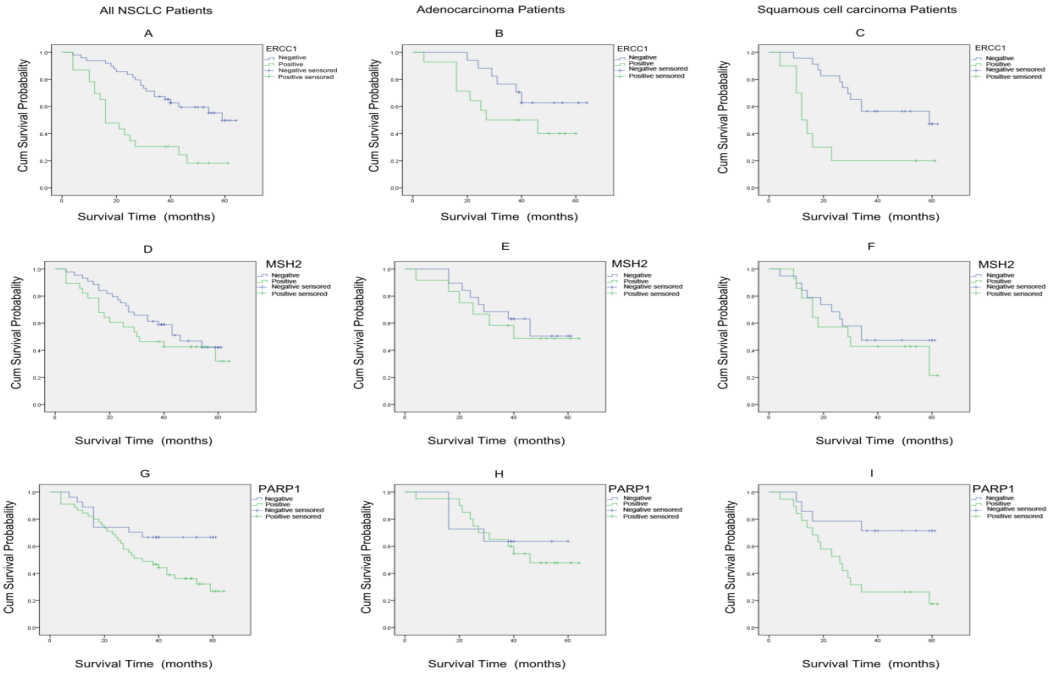

Figure 2. Relationship between the Expression of ERCC1, MSH2 and PARP1 and the Survival Time in NSCLC Patients. ERCC1 and PARP1 negative NSCLC patients have a significantly longer median survival time than ERCC1 positive (A, $p<0.001$) and PARP1 positive patients (G, $p=0.033$), respectively. The difference of the survival time between MSH2 positive and negative NSCLC patients is of no statistical significance (D, p 0.298). Considering specific histological type, expression of ERCC1 is correlated with the survival time of SCC patients (C, $p=0.156$) but not with ADC patients (B, $p=0.004$).Expression of MSH2 is neither correlated with the survival time ADC patients (E, $p=0.701$) nor with SCC patients (F, $p=0.461$). Expression of PARP1 is correlated with the survival SCC patients (I, $p=0.009$) but not ADC (H, $p=0.703$)

Table 4. Multivariate Analysis for Overall Survival of NSCLC Patients after Chemotherapy

Variables	HR	P value	95% CI	
			Upper	Lower
TNM staging	3.803	0.001	1.241	9.62
T staging	0.647	0.44	0.209	2
N staging	1.365	0.276	0.78	2.387
M staging	1.091	0.361	0.429	10.018
ERCC1	3.295	0.029	1.73	11.278
PARP1	2.292	0.036	1.082	4.854

HR, hazard ratio; CI, Confidence interval

Table 5. Multivariate Analysis for Overall Survival of ADCs after Chemotherapy

Variables	RR	P	95% CI	
			Upper	Lower
TNM staging	4.962	0.002	1.22	20.175
T staging	2.768	0.107	0.659	11.133
N staging	0.882	0.776	0.37	2.101
M staging	1.191	0.87	0.147	9.667
ERCC1	6.106	0.009	1.685	22.127
PARP1	2.292	0.028	2.159	32.01

RR, relative risk; CI, Confidence interval

stage ($p=0.012$) and M stage ($p=0.033$) appeared to affect the survival of NSCLC patients after platinum-based chemotherapy.

Expression of ERCC1, MSH2 and PARP1 and clinical outcomes of NSCLC patients after platinum-based chemotherapy

The prognostic values of ERCC1, MSH2 and PARP1 in NSCLC patients after platinum-based chemotherapy were also evaluated. No significant associations were found between the expression of MSH2 and the survival of NSCLC patients ($p=0.298$, Figure 2D). In contrast, the expression of ERCC1 ($p<0.001$, Figure 2A) and RARP1 ($p=0.033$, Figure 2G) were significantly associated with the survival of patients. Multivariate analysis was performed by using the Cox proportional hazards model to find independent prognostic variables for NSCLC patients after chemotherapy. The results showed that ERCC1, PARP1 and TNM staging were the independent prognostic factors for NSCLC patients (Table 4).

Expression of ERCC1, MSH2 and PARP1 was also analyzed in specific histologic type of NSCLC. It was found that expression of ERCC1, MSH2 and PARP1 has no correlation with prognosis of ADCs (Figure 2B, 2E and 2H). But for SCCs, ERCC1 and PARP1 appeared to have predictive ability for the survival of patients with p value of 0.004 (Figure 2C) and 0.009 (Figure 2I), respectively. Cox regression analysis revealed that ERCC1 and PARP1 were independent prognostic factors for SCCs (Table 5).

Combined prognostic values of ERCC1 and PARP1

The 72 NSCLC patients that received chemotherapy after curative resection were subdivided into 4 groups according to the expression of ERCC1 and PARP1: both positive (N=17), ERCC1 positive (PARP1 negative, N=10), PARP1 positive (ERCC1 negative, N=30) and both negative (N=15). The median survival times of the four groups were 22, 37, 42 and 57 months, respectively. The both positive group had significantly shorter survival time than ERCC1 positive group (Figure 3A, $p=0.048$) and

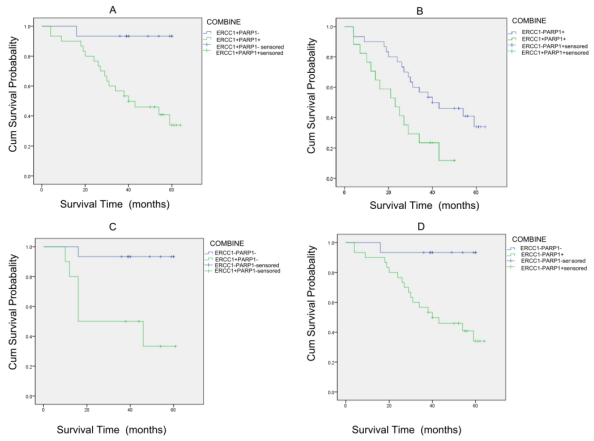

Figure 3. Relationship between the Combined Expression Pattern of ERCC1 and PARP1 and the Survival Time in NSCLC Patients. Patients with both positive expression of ERCC1 and PARP1 had significantly shorter survival time than ERCC1 positive alone (A, p=0.048) and PARP1 positive singly (p=0.01).Patients with both negative expression of ERCC1 and PARP1 had significantly longer survival time than ERCC1 positive singly (C, p=0.004) and PARP1 positive alone (D, p=0.027)

PARP1 positive group (Figuer 3B, p=0.01) while the both negative group had significantly longer survival time than ERCC1 positive group (Figure 3C, p=0.004) and PARP1 positive group (Figure 3D, p=0.027).

Discussion

Platinum-based adjuvant chemotherapy is commonly used for NSCLC treatment. It was reported that platinum-based adjuvant chemotherapy increased the five year survival rate by 4.1% in the 1867 NSCLC patients from International Adjuvant Lung Cancer Trail (IALT) (Arriagada et al., 2004). Platinum drugs bind to DNA and the resulting complex induce lesion of DNA molecules, leading to cell death (OlaussenDunant et al., 2006). Therefore, proteins in DNA repair pathways can affect response to platinum drugs and influence the survival of patients (Parsons et al., 2005; OlaussenDunant et al., 2006; Rosell et al., 2007; KamalSoria et al., 2010; Michels et al., 2013).

In the 111 NSCLC patients enrolled in our study, the positive rate of ERCC1, MSH2 and PARP1 expression is 33.3%, 36.9% and 55.9%, respectively. Additionally, no correlation was observed between the expression of ERCC1, MSH2 and PARP1 and clinical variables including gender, age, smoking history, histologic type, TNM staging.

ERCC1 is an important factor in NER pathway which plays a crucial role in stabilizing the genome, correcting replication errors, avoiding mutations and inhibiting cancer genesis. Activity of ERCC1 is considered to be the representative of the function level of NER pathway (Friedberg, 2001). Zhang et al. (2012) found that ERCC1 118 T/T genotypes might be association with lower survival of NSCLC patients after cisplatin-based chemotherapy. A meta-analysis pooled 11 studies and found that a higher expression of ERCC1 was correlated with worse prognosis in NSCLC patients (Rothand Carlson, 2011). Similarly, in present study, expression of ERCC1 was a prognostic factor in all NSCLC patients and SCCs. However, the most recent study failed to validate the prognostic value of ERCC1 in a cohort of 494 NSCLC patients (Friboulet et al., 2013). Ozdemir et al. (2013) reported that 60.2% NSCLC patients displayed ERCC1 positive expression, higher than reported by current study, but there was no association between ERCC1 expression and the clinical outcomes of NSCLC patients after platinum-based chemotherapy. It should be noted that in the study of conducted by Ozdemir et al., only patients with advanced NSCLC (stage 3B and 4) were included which may be the reason of the different results presented in their and our studies. Due to the inconsistent results, we are not sure to apply ERCC1 to guide customized therapy; instead, we found combination of ERCC1 and PARP1 showed relatively reliable prognostic value. Besides, we didn't detect the association between ERCC1 expression and the clinical outcomes of ADC patients in spite of the prognostic value of it in NSCLC and SCC patients. It is implied that the prognostic value of ERCC1 may be limited to histological type.

MMR is the most important way of correcting replication errors in cell proliferation, keeping the accuracy of genetic information. MSH1 and MSH2 are regulatory factors in MMR pathway which identify and correct mismatched bases. MSH2 is a component of complex MutSot and MutSI3 (Hays et al., 2005). Lower mRNA (Vageli et al., 2012) or protein (PierceallOlaussen et al., 2012) level of MSH2 has been found correlated with better efficacy of chemotherapy to SCCs but not to ADCs. Our study failed to find correlation between MSH2 expression and prognosis in both SCCs and ADCs, which may be attributed to the limited cases of patients.

BER is the most important pathway to deal with DNA damages induced by platinum drugs. PARP1 is recruited after DNA breakdown and binds to the breaking site, initiating BER DNA repair pathway (Rouleau et al., 2010; KummarChen et al., 2012). A higher positive rate of PARP1 expression was found in ADCs compared with SCCs (Olaussen et al., 2013). However, in another study, more SCC patients were PARP1 positive compared to ADC patients (PierceallOlaussen et al., 2012). In our study, lower level of PARP1 was found significantly correlated with longer survival time for SCCs. More cases are needed to confirm this result.

We hypothesize that tumor involves systemic changes and a single gene or protein has limited predictive value while a combination of genes or proteins can offer satisfactory predictive value. Therefore, based on the results above, we analyzed the combined effect of ERCC1 and PARP1. Interestingly, patients with both negative expression of ERCC1 and PARP1 had significantly longer survival time than those with ERCC1 negative or PARP1 negative alone. We proposed that patients with both lower expression of ERCC1 and PARP1 are benefit from platinum-base chemotherapy while those with both higher expression are recommended to consider other ways to help cure NSCLC.

However, quantification by immunochemistry staining

is in part subjective. In summary, more data are needed to testify the use of ERCC1 and PARP1 in prediction of NSCLC prognosis.

In conclusion, TNM staging is used to predict the clinical outcomes of NSCLC patients. However, TNM staging is not sufficient for personalized therapy and objective biomarkers including genes and proteins are in need. Our study explored the predictive values of ERCC1, MSH2 and PARP1 proteins for prognosis in NSCLC patients with platinum-based adjuvant chemotherapy and found that ERCC1 and PARP1 were potential predictors for the survival of NSCLC patients.

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References

Arriagada R, Bergman B, Dunant A, et al (2004). Cisplatin-based adjuvant chemotherapy in patients with completely resected non-small-cell lung cancer. *N Engl J Med*, **350**, 351-60.

Bahl A, Falk S (2001). Meta-analysis of single agents in the chemotherapy of NSCLC: what do we want to know? *Br J Cancer*, **84**, 1143-5.

Friboulet L, Olaussen KA, Pignon JP, et al (2013). ERCC1 isoform expression and DNA repair in non-small-cell lung cancer. *N Engl J Med*, **368**, 1101-10.

Friedberg EC (2001). How nucleotide excision repair protects against cancer. Nature reviews. *Cancer*, **1**, 22-33.

Hays JB, Hoffman PD, Wang H (2005). Discrimination and versatility in mismatch repair. *DNA repair*, **4**, 1463-74.

Jassem J, Skokowski J, Dziadziuszko R, et al (2000). Results of surgical treatment of non-small cell lung cancer: validation of the new postoperative pathologic TNM classification. *J Thorac Cardiovasc Surg*, **119**, 1141-6.

Jemal A, Murray T, Ward E, et al (2005). Cancer statistics, 2005. *CA Cancer J Clin*, **55**, 10-30.

Jemal A, Siegel R, Ward E, et al (2006). Cancer statistics, 2006. *CA Cancer J Clin*, **56**, 106-30.

Kamal NS, Soria JC, Mendiboure J, et al (2010). MutS homologue 2 and the long-term benefit of adjuvant chemotherapy in lung cancer. *Clin Cancer Res*, **16**, 1206-15.

Kummar S, Chen A, Parchment RE, et al (2012). Advances in using PARP inhibitors to treat cancer. *BMC medicine*, **10**, 25.

Li X-D, Han J-C, Zhang Y-J, et al (2013). Common variations of DNA repair genes are associated with response to platinum-based chemotherapy in NSCLCs. *Asian Pac J Cancer Prev*, **14**, 145-8.

Michels J, Vitale I, Galluzzi L, et al (2013). Cisplatin resistance associated with PARP hyperactivation. *Cancer Res*, **73**, 2271-80.

Olaussen K A, Adam J, Vanhecke E, et al (2013). PARP1 impact on DNA repair of platinum adducts: preclinical and clinical read-outs. *Lung Cancer*, **80**, 216-22.

Olaussen KA, Dunant A, Fouret P, et al (2006). DNA repair by ERCC1 in non-small-cell lung cancer and cisplatin-based adjuvant chemotherapy. *N Engl J Med*, **355**, 983-91.

Ozdemir O, Ozdemir P, Veral A, et al (2013). ERCC1 expression does not predict survival and treatment response in advanced stage non-small cell lung cancer cases treated with platinum based chemotherapy. *Asian Pac J Cancer Prev*, **14**, 4679-83.

Parkin DM, Bray F, Ferlay J, Pisani P (2005). Global cancer statistics, 2002. *CA Cancer J Clin*, **55**, 74-108.

Parsons JL, Dianova, II, Allinson SL, Dianov GL (2005). Poly(ADP-ribose) polymerase-1 protects excessive DNA strand breaks from deterioration during repair in human cell extracts. *FEBS J*, **272**, 2012-21.

Pierceall WE, Olaussen KA, Rousseau V, et al (2012). Cisplatin benefit is predicted by immunohistochemical analysis of DNA repair proteins in squamous cell carcinoma but not adenocarcinoma: theranostic modeling by NSCLC constituent histological subclasses. *Ann Oncol*, **23**, 2245-52.

Pignon JP, Tribodet H, Scagliotti GV, et al (2008). Lung adjuvant cisplatin evaluation: a pooled analysis by the LACE Collaborative Group. *J Clin Oncol*, **26**, 3552-9.

Rosell R, Lord RV, Taron M, Reguart N (2002). DNA repair and cisplatin resistance in non-small-cell lung cancer. *Lung cancer*, **38**, 217-27.

Rosell R, Skrzypski M, Jassem E, et al (2007). BRCA1: a novel prognostic factor in resected non-small-cell lung cancer. *PloS one*, **2**, e1129.

Roth JA, Carlson JJ (2011). Prognostic role of ERCC1 in advanced non-small-cell lung cancer: a systematic review and meta-analysis. *Clin Lung Cancer*, **12**, 393-401.

Rouleau M, Patel A, Hendzel MJ, et al (2010). PARP inhibition: PARP1 and beyond. *Nat Rev Cancer*, **10**, 293-301.

Scartozzi M, Franciosi V, Campanini N, et al (2006). Mismatch repair system (MMR) status correlates with response and survival in non-small cell lung cancer (NSCLC) patients. *Lung Cancer*, **53**, 103-9.

Vageli DP, Zaravinos A, Daniil Z, et al (2012). hMSH2 and hMLH1 gene expression patterns differ between lung adenocarcinoma and squamous cell carcinoma: correlation with patient survival and response to adjuvant chemotherapy treatment. *Int J Biol Markers*, **27**, e400-4.

Wang D, Xiang DB, Yang XQ, et al (2009). APE1 overexpression is associated with cisplatin resistance in non-small cell lung cancer and targeted inhibition of APE1 enhances the activity of cisplatin in A549 cells. *Lung Cancer*, **66**, 298-304.

Zhang Z-Y, Tian X, Wu R, et al (2012). Predictive role of ERCC1 and XPD genetic polymorphisms in survival of Chinese non-small cell lung cancer patients receiving chemotherapy. *Asian Pac J Cancer Prev*, **13**, 2583-6.