pISSN 1598-2998, eISSN 2005-9256

Cancer Res Treat. 2016;48(1):71-79

http://dx.doi.org/10.4143/crt.2015.018

Original Article

Open Access

The Prognostic Significance of *FGFR4* Gly388 Polymorphism in Esophageal Squamous Cell Carcinoma after Concurrent Chemoradiotherapy

Hyun-Jeong Shim, MD, PhD[1]
Min-Ho Shin, MD, PhD[2,3]
Hee-Nam Kim, MD, PhD[3]
Jo-Heon Kim, MD, PhD[4]
Jun-Eul Hwang, MD, PhD[1]
Woo-Kyun Bae, MD, PhD[1]
Ik-Joo Chung, MD, PhD[1]
Sang-Hee Cho, MD, PhD[1]

Departments of [1]Hemato-Oncology and [2]Preventive Medicine, [3]Jeonnam Regional Cancer Center, [4]Department of Pathology, Chonnam National University Medical School, Hwasun, Korea

Correspondence: Sang-Hee Cho, MD, PhD
Department of Hemato-Oncology
Chonnam National University Hwasun Hospital,
322 Seoyang-ro, Hwasun 58128, Korea
Tel: 82-61-379-7633
Fax: 82-61-379-7628
E-mail: shcho@jnu.ac.kr

Received January 12, 2015
Accepted April 6, 2015
Published Online May 14, 2015

Purpose

The purpose of this study is to investigate the role of fibroblast growth factor receptor 4 (*FGFR4*) polymorphism in esophageal cancer after chemoradiotherapy (CRT).

Materials and Methods

Peripheral blood samples from 244 patients treated with CRT for esophageal squamous cell carcinoma were assessed for the role of *FGFR4* genotype on treatment response and survival.

Results

A total of 94 patients were homozygous for the Gly388 allele, and 110 were heterozygous and 40 homozygous for the Arg388 allele. No significant association was found between the *FGFR4* genotype and clinicopathological parameters. However, patients carrying the Gly388 allele showed a better overall response rate than Arg388 carriers (p=0.038). In addition, Gly388 allele patients at an earlier stage showed better overall survival (OS) and progression-free survival than Arg388 carriers. Among these, the Gly388 allele showed significantly improved OS compared to Arg388 carriers in the lymph node (LN) metastasis group (p=0.042) compared to the no LN metastasis group (p=0.125). However, similar survival outcomes were observed for advanced-stage disease regardless of genotype.

Conclusion

This result suggests that the role of *FGFR4* Gly388 in treatment outcomes differs according to esophageal cancer stage. It showed a predictive role in the response of esophageal cancer patients to CRT with a better trend for OS in Gly388 than Arg388 carriers in the early stages. In particular, LN-positive early-stage patients carrying the Gly388 allele showed improved OS compared to those carrying Arg388.

Key words
Biological markers, Chemoradiotherapy, Esophageal neoplasms, Fibroblast growth factor receptor 4

Introduction

Treatments for esophageal cancer have been studied intensively in recent decades. However, the clinical outcomes remain unsatisfactory, with 5-year survival rates of 49.3% for localized disease and 2.8% for metastatic disease [1]. Until recently, the standard treatment for early or locally advanced esophageal cancer was surgery. However, definitive chemoradiotherapy (CRT) is another possibility in cases where surgery is not an option due to advanced-stage disease or the presence of comorbidities [2]. To enhance the efficacy of CRT, clinical trials of various drugs have been conducted, including trials for cisplatin and 5-fluorouracil (5-FU), as well as docetaxel [3], paclitaxel [4], and cetuximab [5].

In addition, many recent studies have focused on identification of new biomarkers to help in prediction of the outcome after CRT and to find additional therapeutic targets. Although the underlying pathogenic mechanisms of lung cancer, colorectal cancer, and breast cancer have been inves-

tigated extensively and targeted therapies have since been developed, relatively little is known about esophageal cancer.

Esophageal cancer can metastasize readily, even at early disease stages, due to the absence of a serosa and a rich lymphatic system adjacent to the major organ, resulting in a tendency for metastasis and poor outcomes. Thus, to control the development of esophageal cancer, it is important to not only kill the tumor cells but also to inhibit invasion and metastasis. Recently, exploiting the relationship between tumor cells and the stroma was highlighted as a possible way to prevent the initiation and progression of tumors.

Fibroblasts play an important role in the tumor microenvironment. Carcinoma-associated fibroblasts in the tumor microenvironment influence tumor cell survival, growth, angiogenesis, invasion, inflammation, migration, and metastasis [6]. Fibroblast growth factors (FGFs) are divided into four groups (FGF1-4), and dysregulation of their receptors (FGFRs) plays an important role in tumorigenesis. Our understanding of the biological properties of FGFRs is increasing, and several reports have described their roles in cancer progression and drug sensitivity [7,8]. Among them, FGFR4 is expressed in myofibroblasts during regeneration following injury, but not in mature skeletal muscle. An *FGFR4* Gly388Arg polymorphism (rs351855), which causes the substitution of arginine for glycine (*FGFR4* Arg388) in the transmembrane domain of the receptor, was reported to increase cancer risk [9], aggressiveness, metastasis [10], and drug resistance [11]. Ansell et al. [12] recently reported a significantly increased risk of head and neck squamous cell carcinoma (HNSCC) with the *FGFR4* Gly388 allele, whereas Arg388 resulted in cisplatin sensitivity (p=0.141) in an *in vitro* assay. In addition, Arg388 carriers showed significantly poor survival outcome in head and neck or lung cancer patients [10,13]. Considering the similar causal factors such as alcohol or smoking and treatment modality of head and neck, lung or esophageal cancer, *FGFR4* polymorphism could be a targetable prognostic marker in esophageal cancer, for which there has been no reliable biomarker until now.

Thus, we investigated the role of *FGFR4* polymorphisms in esophageal squamous cell carcinoma patients who were treated with CRT.

Materials and Methods

1. Patients and treatment

This retrospective study was conducted to assess the relationship between *FGFR4* polymorphisms and treatment outcomes in patients receiving CRT for esophageal squamous cell carcinoma. The inclusion criteria were (1) a diagnosis of squamous esophageal carcinoma, (2) treatment with concurrent CRT as a preoperative or curative-aim first-line therapy with at least 4,000 cGy of radiation, (3) evaluation of the response after CRT, (4) a Karnofsky performance status ≥ 70 at diagnosis, (5) preserved organ function sufficient to receive CRT, and (6) a blood sample available for DNA analysis. Patients were excluded for (1) other confirmed or suspected malignancies, (2) types of cancer other than squamous cell carcinoma (e.g., adenocarcinoma), (3) gastroesophageal junction carcinoma, and (4) postoperative CRT.

During radiotherapy, the patients received combination chemotherapy with 5-FU (1,000 mg/m^2 on days 1-4) and cisplatin (75 mg/m^2 on day 1) every 4 weeks or weekly docetaxel (30 mg/m^2 on days 1 and 8) and cisplatin (30 mg/m^2 on days 1 and 8) every 3 weeks. Blood samples for genotyping were drawn before the start of CRT.

Evaluation of the response was based on the Response Evaluation Criteria in Solid Tumors (RECIST ver. 1.1), and was assessed 8 weeks after completion of CRT. After CRT, surgery was performed for the preoperative CRT cases. Additional chemotherapy was performed in cases where any disease remained without progression, or if surgery was not possible. Second-line chemotherapy was administered in cases where the disease had progressed.

Blood samples for polymorphism study were provided by the Chonnam National University Hwasun Hospital National Biobank of Korea, a member of the National Biobank of Korea, which is supported by the Ministry of Health, Welfare and Family Affairs. The study protocol was approved by the Institutional Review Board of Chonnam National University Hwasun Hospital (CNUHH-2014-016).

2. Genotyping

Genomic DNA was extracted from peripheral blood using a QIAamp DNA Blood Mini Kit (Qiagen, Valencia, CA), according to the manufacturer's protocol. *FGFR4* Gly388Arg genotyping was performed using high-resolution melting (HRM) analysis with a Rotor Gene 6000 (Corbett Research, Sydney, Australia). The primers were as follows: forward, 5'-ggagagcttctgcacagtgg-3'; and reverse, 5'-cttggctgtgctcctgct-3'. The reaction mixture for HRM included 200 nM polymerase chain reaction primers, 1 μM SYTO 9 fluorescent dye (Invitrogen, Carlsbad, CA), 0.5 U of f-Taq polymerase, and 40 ng of genomic DNA in 10-μL reaction volumes. The cycling conditions included an initial 5-minute hold at 95°C, followed by 40 cycles at 95°C for 5 seconds, 65°C for 30 seconds, and 72°C for 20 seconds, with an increase in the melting temperature from 78°C to 92°C at 0.1°C/sec. The genotyping results were validated using direct sequencing

Table 1. Patient clinicopathologic characteristics

	Total	G/G	G/A	A/A	p-value	G/A or AA	p-value
No. (%)	244	94 (39)	110 (45)	40 (16)		150 (61)	
Median age (range, yr)	66 (41-87)	64 (41-79)	66 (47-87)	66 (49-81)			
≤ 60	75 (31)	32 (34)	31 (28)	12 (30)	0.66	43 (29)	0.228
> 60	169 (69)	62 (66)	79 (72)	28 (70)		107 (71)	
Sex							
Male	237 (97)	91 (97)	106 (96)	40 (100)	0.485	146 (97)	0.549
Female	7 (7)	3 (3)	4 (4)	0		4 (3)	
Differentiation							
Well	102 (42)	37 (39)	46 (42)	19 (47)	0.95	65 (43)	0.75
Moderate	85 (35)	32 (34)	40 (36)	13 (33)		53 (35)	
Poorly	32 (13)	15 (16)	13 (12)	4 (10)		17 (12)	
Unknown	25 (10)	10 (11)	11 (10)	4 (10)		15 (10)	
Tumor location							
Upper	63 (26)	27 (29)	21 (19)	15 (38)	0.123	36 (24)	0.712
Mid	133 (54)	49 (52)	68 (62)	16 (40)		84 (56)	
Lower	48 (20)	18 (19)	21 (19)	9 (23)		30 (20)	
TNM stage							
I	11 (5)	3 (3)	7 (6)	1 (3)	0.859	8 (5)	0.839
II	54 (22)	21 (21)	23 (21)	11 (27)		34 (23)	
III	101 (41)	39 (42)	45 (41)	17 (43)		62 (41)	
IV	78 (32)	32 (34)	35 (32)	1 (27)		46 (31)	
Chemotherapy							
FP	204 (84)	76 (81)	93 (85)	36 (87)	0.596	128 (85)	0.228
DP	40 (16)	18 (19)	17 (15)	5 (13)		22 (15)	
Operation							
Yes	58 (24)	73 (78)	82 (74)	32 (80)	0.661	113 (75)	0.399
No	186 (76)	21 (22)	29 (26)	8 (20)		37 (25)	
Second-line chemotherapy							
Yes	88 (36)	58 (62)	71 (65)	27 (67)	0.802	98 (65)	0.33
No	156 (64)	36 (38)	39 (35)	13 (33)		52 (35)	

Values are presented as number (%) unless otherwise indicated. FP, fluoropyrimidine and cisplatin; DP, docetaxel and cisplatin.

(ABI PRISM 3100 Genetic Analyzer, Life Technologies, Carlsbad, CA) of 16 subjects (6%), and the results showed 100% concordant. Appropriate positive/negative and internal controls were included.

3. Statistical analyses

The progression-free survival (PFS) time was calculated from the start of CRT to the appearance of disease progression or recurrence after surgery. The overall survival (OS) time was calculated from the start of CRT to death from any cause; patients who were alive at the last follow-up were recorded at that time. Chi-square tests were used to evaluate associations between genotypes and clinicopathological characteristics and chemotherapy responses. The Cox proportional hazards model was used to evaluate the effect of variables on OS and PFS, and to produce survival curves. The proportional hazards assumption was assessed by graphical evaluation of log-log plots. All statistical analyses were performed using SPSS ver. 17.0 (SPSS Inc., Chicago, IL). A p-value of < 0.05 was considered to indicate statistical significance.

Results

1. Study population

Between May 2004 and December 2012, 264 patients were treated for esophageal cancer using CRT, and 244 patients

Table 2. Response after chemoradiotherapy according to *FGFR4* Gly388Arg polymorphism

Genotype	Responder (CR+PR, n=210)	Non-responder (SD+PD, n=34)	p-value
G/G (n=94)	86 (91.5)	8 (8.5)	0.090
G/A (n=110)	89 (80.9)	21 (19.1)	
A/A (n=40)	35 (87.5)	5 (12.5)	
G/G (n=94)	86 (91.5)	8 (8.5)	0.038
G/A+A/A (n=150)	124 (82.7)	26 (17.3)	

Values are presented as number (% of each gene group). *FGFR4*, fibroblast growth factor receptor 4; CR, complete response; PR, partial response; SD, stable disease; PD, progressive disease.

suitable for inclusion were analyzed in this study. The median dose of radiation was 5,400 cGy (range, 4,000 to 6,600 cGy); 187 patients (77%) received radiation doses ≥ 5,000 cGy, and 57 (23%) received doses < 5,000 cGy due to preoperative aims or poor general conditions.

The chemotherapy regimens during radiation treatment were as follows: 5-FU and cisplatin in 204 (84%) and docetaxel and cisplatin in 40 patients (16%). After CRT, 58 patients (24%) underwent surgery with a curative aim. In total, 143 patients (58.6%) showed disease progression after CRT or surgery, and 88 patients (36%) received second-line chemotherapy (Table 1).

2. Chemotherapy responses and survival

Fifty-two patients (21%; 95% confidence interval [CI], 16 to 27) showed a complete response, and 158 patients (65%; 95% CI, 58 to 70) showed a partial response after CRT.

Patients at an earlier stage of disease showed a significantly better response. The chemotherapy regimen during radiation therapy (5-FU+cisplatin vs. docetaxel+cisplatin) had no effect on the overall response rate (p=0.307). The median OS and PFS for stage I disease were not reached in this analysis. The median OS for stages II, III, and IV were 44.0 months (95% CI, 22.6 to 46.2), 25.6 months (95% CI, 14.0 to 32.6), and 21.9 months (95% CI, 11.3 to 27.3), respectively, and statistically significant differences were observed among the stages (p=0.001). The median PFS rates of stages II, III, and IV were also significantly different at 42.0 months (95% CI, 0.8 to 54.8), 12.7 months (95% CI, 10.0 to 15.6), and 9.3 months (95% CI, 12.1 to 16.9), respectively (p < 0.001). These results were similar after grouping as early-stage (stages I and II) versus advanced-stage (stages III and IV) disease in PFS (p < 0.001) and OS (p=0.001). Although 58 patients (23.7%) underwent surgery after CRT, no significant difference in survival was observed between those who underwent surgery (PFS, 17.3 months; 95% CI, 5.4 to 29.2 and OS, 30.2 months; 95% CI, 15.5 to 44.9) and those who did not (PFS, 13.6 months; 95% CI, 10.6 to 16.6; p=0.293 and OS, 24.3 months; 95% CI, 18.5 to 30.1; p=0.434).

3. *FGFR4* genotype and treatment outcome

The frequencies of the *FGFR4* Gly388Arg polymorphic genotypes were as follows: 94 patients (38.5%) had the Gly388 allele (G/G), 110 (45.4%) were heterozygous for G/A, and 40 (16.4%) had the Arg388 allele (A/A). Patients with the G/G allele (91.5%; 95% CI, 82.9 to 95.6) showed significantly better responses to CRT than Arg carriers (82.7%; 95% CI, 75.4 to 88.0; p=0.038) (Table 2). However, no significant association was found between the *FGFR4* genotype and any clinicopathological parameter examined, including tumor stage, differentiation, and tumor location (Table 1). No difference between heterozygous and homozygous Arg388 carriers was observed in this study.

In survival analyses, no significant difference in OS or PFS was observed among genotypes. For comprehensive evaluation of the role of genotype in treatment outcome, the patients were divided into early (stages I and II; n=65) and advanced stage groups (stages III and IV; n=179). In the early stages, patients with the Gly388 allele tended to have somewhat better PFS (unreached median OS [mOS] in G/G, 38 months in Arg388 carriers; 95% CI, 11.9 to 63.9; p=0.506) and OS rates (unreached mOS in G/G, 45 months; 95% CI, 26.1 to 63.5; p=0.773) (Fig. 1A and C) than the Arg388 carriers. However, patients with advanced stage disease showed similar patterns of PFS (9.6 months in G/G; 95% CI, 6.1 to 13 and 12.6 months in Arg388 carriers; 95% CI, 10.2 to 14.9; p=0.678) and OS (24.7 months in G/G; 95% CI, 19.5 to 29.90 and 22.4 months in Arg388 carriers, 95% CI, 15.3 to 29.5; p=0.668) (Fig. 1B and D) regardless of genotype. In particular, patients with the Gly388 allele showed a significantly better OS rate than Arg388 carriers in the lymph node (LN) metastasis group (p=0.042) compared to the no LN metastasis group (p=0.125) (Fig. 2) in the early stages.

4. Univariate and multivariate analyses

To explore the prognostic factors for esophageal cancer patients, a univariate analysis was performed with clinicopathologic variables and genotypes. A multivariate analysis

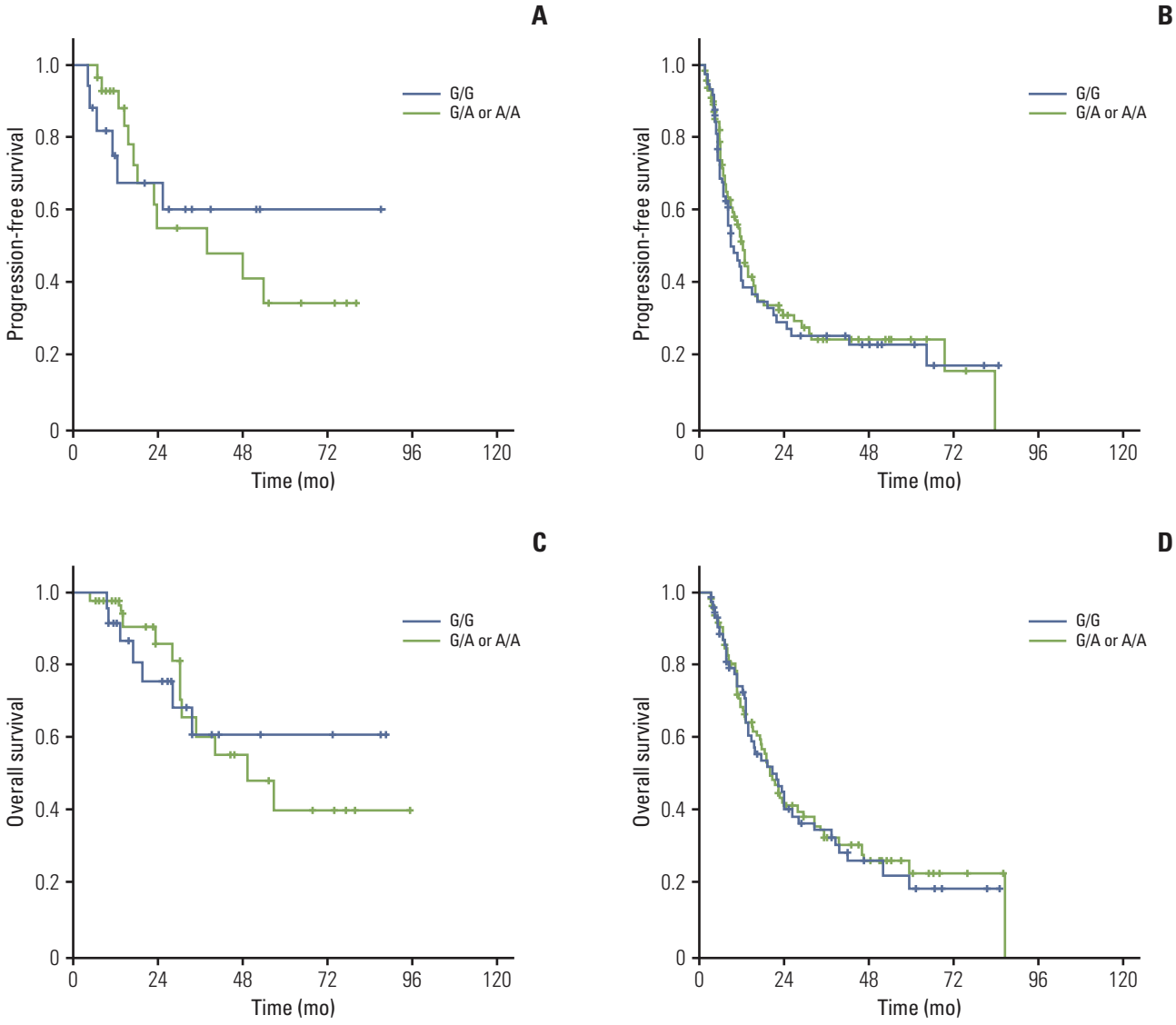

Fig. 1. Association between genotype and survival outcome. In early stage of esophageal cancer, *FGFR4* Gly388 allele (G/G) patients show better trends of progression-free survival (A) and overall survival (C) than *FGFR4* Arg388 carriers (G/A or A/A) without statistical significance. However, the progression-free survival (B) and overall survival (D) in advanced stage of esophageal cancer patients show comparable outcomes regardless of genotypes.

using the Cox proportional hazards model was used for evaluation of the statistically significant variables found in the univariate analysis. As shown in Table 3, age > 60 years, early-stage disease, and positive responder status (showing a complete or partial response) were significantly favorable prognostic factors for PFS, as were female sex, early-stage disease, and positive responder status for OS in the univariate analysis. Among these factors, stage and responder status were significant prognostic factors for survival in the multivariate analysis.

Discussion

Treatment outcomes for various cancers have improved over recent decades. This can be attributed largely to the use of new chemotherapeutic agents, as well as enhanced technologies for surgery and radiation therapy. However, despite development of more drugs that target tumor cells, many obstacles to successful treatment remain. Accurate control of tumor cells and the tumor microenvironment is important in

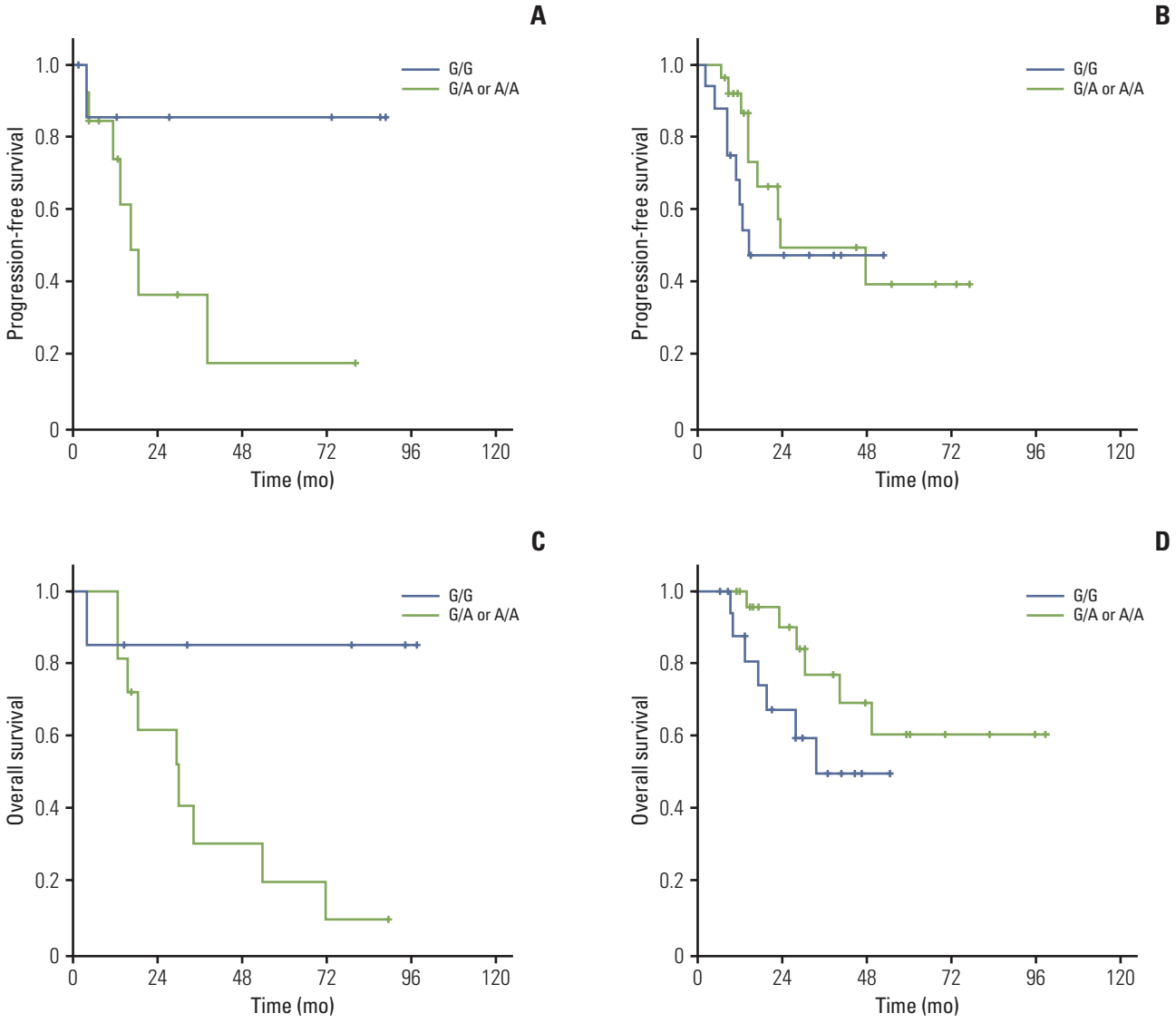

Fig. 2. Survival according to genotype in early esophageal cancer. In early esophageal cancer, *FGFR4* Gly388 allele patients showed a better trend of progression-free survival (A) (p=0.079) and significantly better overall survival (C) (p=0.042) than *FGFR4* Arg388 carriers in lymph node (LN)-positive patients. However, there was no difference in LN-negative patients according to genotypes (B, D).

eradicating cancer. Based on this rationale, FGFRs have been identified as potential therapeutic targets known to be associated with drug resistance. Although FGFR1-3 has been extensively investigated [14-16], the role of FGFR4 and its potential as a therapeutic target is less understood. FGFR4 is overexpressed in human prostate, breast, colon, rhabdomyosarcoma, gastric, and hepatocellular cancers, where it may be associated with tumor progression and invasion via a variety of pathways, and with a poor prognosis [17,18]. Recent studies found that *FGFR4* promotes the epithelial-mesenchymal transition (EMT) in cancer cells [19,20].

Studies of *FGFR* polymorphisms have reported association of Arg388 with increased cancer incidence, tumor size [17,21], and recurrence after adjuvant treatment [11,12,22]. In addition, FGFR affects radiation therapy outcomes. The current study was conducted in order to assess the role of *FGFR4* Arg388 in patients treated with CRT for esophageal cancer, which is treated primarily by CRT in inoperable cases. We found that patients with the Gly388 allele showed a significantly better response to CRT than Arg388 carriers,

Table 3. Univariate analysis for survival

	PFS (median, mo)	p-value	OS (median, mo)	p-value
Age (yr)				
≤ 60	9.9±2.12	0.003	24.4±3.15	0.139
> 60	21.9±4.03			
Sex				
Male	8.1±7.10	0.472	16.3±6.10	0.044
Female	15.4±1.69		33.3±6.55	
Differentiation				
Well	20.9±5.47	0.055	34.5±11.12	0.318
Moderate+poorly	12.8±1.30		23.7±4.26	
Tumor location				
Upper	18.3±3.64	0.28	38.3±7.94	0.279
Middle	14.3±3.38		34.5±7.03	
Lower	12.3±2.55		19.0±1.82	
Clinical stage				
I-II	NR	< 0.001	NR	0.001
III-IV	11.6±1.13		23.5±2.29	
CRT response				
Responder (CR+PR)	20.9±2.93	< 0.001	38.3±4.52	< 0.001
Non responder (SD+PD)	6.8±1.11		11.8±1.56	
Operation				
Yes	21.7±5.97	0.239	56.7±15.49	0.14
No	14.3±1.43		29.8±4.79	
RT dose				
Stage I-II				
≥ 5,000 cGy	58.9±7.52	0.353	66.3±7.19	0.318
< 5,000 cGy	63.7±13.63		75.8±13.29	
Stage III-IV				
≥ 5,000 cGy	26.8±2.81	0.426	48.3±6.66	0.415
< 5,000 cGy	38.2±6.38		40.2±3.37	
Chemoregimen				
FP	15.0±1.66	0.637	32.4±4.57	0.664
DP	18.0±6.86		34.5±9.70	
Second-line chemotherapy				
Yes	8.8±0.82	0.382	21.6±1.99	0.692
No	9.0±1.14		17.8±2.64	
Genotype				
Stage I-II				
G/G	NR	0.506	NR	0.773
G/A+A/A	44.8±9.54		37.9±13.25	
Stage III-IV				
G/G	9.6±1.75	0.678	24.7±2.65	0.668
G/A+A/A	12.5±1.22		22.4±3.63	

PFS, progression-free survival; OS, overall survival; NR, not reached until this analysis; CRT, chemoradiotherapy; CR, complete response; PR, partial response; SD, stable disease; PD, progressive disease, RT, radiotherapy; FP, 5-fluorouracil+cisplatin; DP, docetaxel+cisplatin.

regardless of the cancer stage. Interestingly, the PFS and OS rates in early esophageal cancer (stages I and II) showed improved survival trends with the Gly388 versus the Arg388 allele. However, comparable patterns of PFS and OS in advanced esophageal cancer (stages III and IV) were observed between the groups.

In the study reported by Thussbas et al. [11] in breast cancer patients, disease-free survival with the Gly388 allele was prolonged significantly compared with the Arg388 allele when patients received adjuvant chemotherapy; however, no difference was observed among patients who underwent adjuvant endocrine therapy. Thus, FGFR4 could be involved in efficacy of chemotherapy, but not endocrine therapy. The current study suggests that FGFR4 affects the response to chemotherapy and adjuvant therapy outcomes. A possible mechanism that would fit this result is the EMT, an important step in disease recurrence after surgery [23]. In a previous report, FGFR4 was shown to participate in a complex with membrane-type 1 matrix metalloproteinase (MT1-MMP), decreasing MT1-MMP lysosomal degradation and leading to increased invasion. In the current study, we extended the focus by assessing the functional role of FGFR4 according to stage. Although the difference was not statistically significant, the pattern of survival differed according to stage, showing a tendency to be prolonged in Gly388 versus Arg388 carriers. In addition, in early esophageal cancer patients with LN invasion, significantly improved OS was observed in patients with the Gly388 allele compared to Arg388 carriers. However, this result was not observed in patients receiving palliative care for advanced-stage disease. Progression in advanced tumors is more complex than in early stage tumors through the activation of mesenchymal and epithelial signaling pathways. Thus the function of a single gene can be difficult to assess during treatment in advanced cancer. Taken together, the current data suggest that targeted therapy for FGFR4 could be more effective at earlier rather than more advanced stages of esophageal cancer, and that another treatment approach is needed for patients carrying Arg388 in order to improve the treatment outcome. Unexpectedly, a better trend of OS was observed in Gly388 with LN invasion patients compared to Gly388 without LN invasion patients. Further study with a large number of patients will be needed in order to provide additional evidence to support this result.

FGFs play important roles in the tumor microenvironment, where there is a protective niche for therapeutic escape, through the secretion of various growth factors [6]. The molecular mechanism by which the *FGFR4* Arg388 polymorphism leads to a more aggressive clinical phenotype is not yet fully understood. In a colorectal cancer model, Heinzle et al. [21] showed that the *FGFR4* Arg388 polymorphism induced overexpression of FGFR and that this could attenuate the response to chemotherapy [22]. A similar result was reported in a gastric cancer model showing that an FGFR4 inhibitor and 5-FU reduced proliferation and promoted apoptosis [24]. For HNSCC, the association between CRT and FGFR4 was evaluated *in vitro*, showing increased sensitivity of cells containing the Arg388 allele to cisplatin, which differs from our observations [12].

In contrast to various results primarily from *in vitro* studies, there is a lack of clear evidence for any relationship between the *FGFR4* Arg388 genotype and protein expression or clinicopathological parameters in cancer patients [11,18] However, the *FGFR4* Gly388 allele has been reported as a prognostic marker for survival in breast and gastric cancer patients. The lack of correlation between genotype and phenotype for FGFR4 may be explained in part by the lack of elevated tyrosine phosphorylation of *FGFR4* Arg388 compared with *FGFR4* Gly388, the use of different intracellular signal transduction pathway(s) or interactions with different cell surface protein(s), or linkage disequilibrium with other genetic changes that also contribute to a poor prognosis [11]. To the best of our knowledge, this is the first report describing a predictive role for *FGFR4* Arg388 after CRT. The biochemical effects of *FGFR4* Arg388 with regard to chemotherapy should be evaluated in future studies including other biomarkers.

Based on our results, clinical factors including early-stage disease and chemotherapy responders were favorable prognostic factors for PFS and OS in a multivariate analysis. Interestingly, patients > 60 years of age had longer PFS than younger patients. The proportions of TNM stages were similar between the two groups (p=0.324); thus, understanding the biology during aging may be another approach to improving treatment outcomes.

Conclusion

Although the *FGFR4* polymorphism examined in this study is not a significant prognostic factor in esophageal cancer treated with CRT, esophageal cancer patients carrying *FGFR4* Gly388 showed a good response after CRT. Especially in early esophageal cancer, the Gly388 allele resulted in increased OS and PFS, and select patients showing LN invasion showed significant prolonged OS compared to Arg388 carriers. These findings support the idea that the effect of *FGFR4* on treatment outcomes after CRT differs according to stage and further prospective study is needed in order to validate these findings.

Conflicts of Interest

Conflict of interest relevant to this article was not reported.

Acknowledgments

This research was sponsored by a grant (HCRI14027-21) from the Chonnam National University Hwasun Hospital Institute for Biomedical Science.

References

1. Siegel R, Naishadham D, Jemal A. Cancer statistics, 2012. CA Cancer J Clin. 2012;62:10-29.
2. Kim DE, Kim UJ, Choi WY, Kim MY, Kim SH, Kim MJ, et al. Clinical prognostic factors for locally advanced esophageal squamous carcinoma treated after definitive chemoradiotherapy. Cancer Res Treat. 2013;45:276-84.
3. Shim HJ, Kim DE, Hwang JE, Bae WK, Nam TK, Na KJ, et al. A phase II study of concurrent chemoradiotherapy with weekly docetaxel and cisplatin in advanced oesophageal cancer. Cancer Chemother Pharmacol. 2012;70:683-90.
4. Kelsey CR, Chino JP, Willett CG, Clough RW, Hurwitz HI, Morse MA, et al. Paclitaxel-based chemoradiotherapy in the treatment of patients with operable esophageal cancer. Int J Radiat Oncol Biol Phys. 2007;69:770-6.
5. Hong TS, Wo JY, Kwak EL. Targeted therapies with chemoradiation in esophageal cancer: development and future directions. Semin Radiat Oncol. 2013;23:31-7.
6. Paraiso KH, Smalley KS. Fibroblast-mediated drug resistance in cancer. Biochem Pharmacol. 2013;85:1033-41.
7. Yang F, Zhang Y, Ressler SJ, Ittmann MM, Ayala GE, Dang TD, et al. FGFR1 is essential for prostate cancer progression and metastasis. Cancer Res. 2013;73:3716-24.
8. Tran TN, Selinger CI, Kohonen-Corish MR, McCaughan BC, Kennedy CW, O'Toole SA, et al. Fibroblast growth factor receptor 1 (FGFR1) copy number is an independent prognostic factor in non-small cell lung cancer. Lung Cancer. 2013;81:462-7.
9. Ma Z, Tsuchiya N, Yuasa T, Inoue T, Kumazawa T, Narita S, et al. Polymorphisms of fibroblast growth factor receptor 4 have association with the development of prostate cancer and benign prostatic hyperplasia and the progression of prostate cancer in a Japanese population. Int J Cancer. 2008;123:2574-9.
10. Spinola M, Leoni V, Pignatiello C, Conti B, Ravagnani F, Pastorino U, et al. Functional FGFR4 Gly388Arg polymorphism predicts prognosis in lung adenocarcinoma patients. J Clin Oncol. 2005;23:7307-11.
11. Thussbas C, Nahrig J, Streit S, Bange J, Kriner M, Kates R, et al. FGFR4 Arg388 allele is associated with resistance to adjuvant therapy in primary breast cancer. J Clin Oncol. 2006;24:3747-55.
12. Ansell A, Farnebo L, Grenman R, Roberg K, Thunell LK. Polymorphism of FGFR4 in cancer development and sensitivity to cisplatin and radiation in head and neck cancer. Oral Oncol. 2009;45:23-9.
13. Dutra RL, de Carvalho MB, Dos Santos M, Mercante AM, Gazito D, de Cicco R, et al. FGFR4 profile as a prognostic marker in squamous cell carcinoma of the mouth and oropharynx. PLoS One. 2012;7:e50747.
14. Wesche J, Haglund K, Haugsten EM. Fibroblast growth factors and their receptors in cancer. Biochem J. 2011;437:199-213.
15. Greulich H, Pollock PM. Targeting mutant fibroblast growth factor receptors in cancer. Trends Mol Med. 2011;17:283-92.
16. Katoh M, Nakagama H. FGF receptors: cancer biology and therapeutics. Med Res Rev. 2014;34:280-300.
17. Taylor JG 6th, Cheuk AT, Tsang PS, Chung JY, Song YK, Desai K, et al. Identification of FGFR4-activating mutations in human rhabdomyosarcomas that promote metastasis in xenotransplanted models. J Clin Invest. 2009;119:3395-407.
18. Ye Y, Shi Y, Zhou Y, Du C, Wang C, Zhan H, et al. The fibroblast growth factor receptor-4 Arg388 allele is associated with gastric cancer progression. Ann Surg Oncol. 2010;17:3354-61.
19. Liu R, Li J, Xie K, Zhang T, Lei Y, Chen Y, et al. FGFR4 promotes stroma-induced epithelial-to-mesenchymal transition in colorectal cancer. Cancer Res. 2013;73:5926-35.
20. Pelaez-Garcia A, Barderas R, Torres S, Hernandez-Varas P, Teixido J, Bonilla F, et al. FGFR4 role in epithelial-mesenchymal transition and its therapeutic value in colorectal cancer. PLoS One. 2013;8:e63695.
21. Heinzle C, Gsur A, Hunjadi M, Erdem Z, Gauglhofer C, Stattner S, et al. Differential effects of polymorphic alleles of FGF receptor 4 on colon cancer growth and metastasis. Cancer Res. 2012;72:5767-77.
22. Gordon MA, Gil J, Lu B, Zhang W, Yang D, Yun J, et al. Genomic profiling associated with recurrence in patients with rectal cancer treated with chemoradiation. Pharmacogenomics. 2006;7:67-88.
23. Sugiyama N, Varjosalo M, Meller P, Lohi J, Chan KM, Zhou Z, et al. FGF receptor-4 (FGFR4) polymorphism acts as an activity switch of a membrane type 1 matrix metalloproteinase-FGFR4 complex. Proc Natl Acad Sci U S A. 2010;107:15786-91.
24. Ye YW, Hu S, Shi YQ, Zhang XF, Zhou Y, Zhao CL, et al. Combination of the FGFR4 inhibitor PD173074 and 5-fluorouracil reduces proliferation and promotes apoptosis in gastric cancer. Oncol Rep. 2013;30:2777-84.