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ORIGINAL RESEARCH

Influence of TS (rs34743033) and RUNX1 (rs2014300) gene polymorphisms on survival outcomes of fluorouracil-based chemotherapy in Chinese advanced gastric cancer patients

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Background: This study aimed to explore the clinical correlation of single-nucleotide polymorphisms of thymidylate synthase (TS) and runt-related transcription factor 1 (RUNX1) in patients with postoperative stage II and III gastric cancer (GC).

Patients and methods: Samples were obtained from 661 patients with postoperative stage II and III GC. TS (rs34743033) and RUNX1 (rs2014300) were genotyped in 261 patients who received postoperative basic platinum and fluorouracil chemotherapy regimens and 400 patients who did not accept chemotherapy.

Results: TS (rs34743033) variant genotypes significantly prolonged the median overall survival (OS) time compared to the patients who only received adjuvant chemotherapy (HR 1.604, 95% CI 1.068–2.410, p=0.021). Moreover, 3R/3R variant genotypes were demonstrated to have a positive effect on the OS of patients who received chemotherapy based on cisplatin (HR 1.754, 95% CI 1.041–2.954, p=0.031) compared to oxaliplatin. A stratification analysis indicated that 2R/3R and 2R/2R variant genotypes were associated with inferior survival in GC patients with intestinal-type tumors, tumor less than 5 cm in size, and poorly differentiated tumors (p<0.05). However, RUNX1 (rs2014300) AA genotypes markedly increased the risk of death in GC patients compared with the GG/GA genotypes (p=0.007), but no significant difference was observed between chemotherapy based on platinum. The stratification analysis showed that the GA/AA genotype was significantly associated with inferior survival in well to moderately differentiated tumors (HR 2.001, 95% CI 1.082–3.703, p=0.023).

Conclusion: These preliminary results indicated that the two polymorphisms had a significant effect on postoperative adjuvant chemotherapy. TS (rs34743033) and RUNX1 (rs2014300) may be used as biomarkers to predict prognosis and select chemotherapy regimens in GC patients.

Keywords: thymidylate synthase, runt-related transcription factor 1, single-nucleotide polymorphisms, adjuvant chemotherapy, gastric cancer

Introduction

Gastric cancer (GC) is the fifth most common human malignant tumor worldwide. Generally, East Asia has the highest incidence, especially Korea, Japan, Mongolia, and People's Republic of China.[1] The incidence and mortality of stomach cancer in People's Republic of China are the second highest among malignant cancers.[2] Surgery and perioperative chemotherapy can improve patient outcomes. In general, the 5-year survival rate increases from 49.6% to 55.3% with the addition of adjuvant chemo-

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therapy to simple surgery.[3] To date, adjuvant chemotherapy for platinum and fluorouracil (PF) has been regarded as an effective first-line therapy for advanced GC.[4,5] However, the effectiveness of fluorouracil is less than 50%, with different results manifesting in different patients.[6] A major reason for this is gene polymorphism, which could lead to different curative effects.[7]

Thymidylate synthase (TS) catalyzes intracellular deoxyuridine monophosphate to deoxypyrimidine monophosphate and is the only source of new thymine in cells. TS plays a key role in the metabolism of folate and deoxythymidine for DNA repair and synthesis.[8,9] The TS gene single-nucleotide polymorphism (SNP) rs34743003 is a type of tandem repeater at the 5-UTR and contains double (2R) or triple (3R) repeats of 28 bp sequences. The polymorphisms may regulate the generation of TS and determine the reaction and toxicity of (5-FU) chemotherapy. Previous studies have shown that high levels of TS expression can lead to poor clinical performance in different cancers.[10–12] Additionally, the active metabolite of 5-FU and 5-fluoro-2-deoxyuridine 5-monophosphate was combined with TS to form a stable structure to inhibit the activity of the TS enzyme. Therefore, TS and 5-FU and its derivatives are considered to be targets in the treatment of malignant tumors.[13] To date, the efficacy of TS rs34743003, specifically to what extent 3R/3R, 2R/3R, and 2R/3R genotypes have a superior overall survival (OS) rate, is controversial.[14,15] However, most samples used in past research are small in number. To further clarify the TS rs34743003 genotype in chemotherapy, we recruited a total of 661 patients for further study.

Runt-related transcription factor 1 (RUNX1), also called acute myeloid leukemia 1 protein, is located on chromosome 21 (21q22.12) and contains 138 amino acid runt homologous zones. RUNX1 is a member of the runt-related transcription factor family and can regulate hematopoietic cell differentiation, apoptosis, and self-renewal.[16] Over time, researchers have found that RUNX1 is not only associated with blood system tumors but also plays an important role in solid tumors.[17,18] The RUNX1 variant rs2014300 was also observed to have a significant association with several cancers.[19,20] However, the association of rs2014300 with GC is unclear.

In the present study, we studied the clinical significance of the functional polymorphisms of TS rs34743003 and RUNX1 rs2014300 in patients who received postoperative adjuvant chemotherapy for GC in People's Republic of China (661 cases, stage II or III GC) and compared them to patients treated without adjuvant chemotherapy. Additionally, we investigated the relationship between the prognosis of the SNPs and some subtypes of stomach cancer.

Material and methods

Study patients

In this retrospective analysis, all patients received a gastrectomy to confirm pathological II–III tumor stages and were recruited from Yixing People's Hospital from 1999 to 2006 (Yixing, People's Republic of China). No patient had received perioperative chemoradiation or preoperation radiotherapy. A total of 261 patients received PF-based adjuvant chemotherapy within 1 month after gastrectomy, but another 400 patients did not accept the abovementioned treatment for various reasons. The study end point was OS, and the survival time was calculated from the date of surgery until death or the end of follow-up in March 2009. Dates of death were confirmed by follow-up telephone calls or patient admission records. Clinical pathologic variables included sex, age, tumor site, tumor size, depth of invasion, lymph node metastasis, Lauren classification, differentiation, drinking, and smoking. The American Joint Committee on Cancer tumor, node and metastasis classification was applied, and Lauren's criteria were used to divide the tumors into intestinal and diffuse types. All detected genotype samples were classified by two independent pathologists.

Treatment plan

For patients with stage II–III disease, at least four cycles of PF-based regimens, including fluorouracil combined with cisplatin or oxaliplatin, were used. Patients who received adjuvant chemotherapy were required to have a neutrophil count of 1.5×10^9/L, a platelet count of 100×10^9/L, a hemoglobin level of ≥8 g/dL, and no signs of organ toxicity.

Genotyping

Genomic DNA was extracted from tumor specimens by proteinase K digestion, isopropanol extraction, and ethanol precipitation. The TS (rs34743033) and RUNX1 (rs2014300) SNPs were examined by multiplex SNaPshot technology using an ABI fluorescence-based assay allelic discrimination method (Applied Biosystems, Foster City, CA, USA) as described previously. The primers of TS for multiplexed PCR were F-primer (CGGAAGGGGTCCTGCCACC) and R-primer (GAGCCGGCCACAGGCATGG). The primers of RUNX1 for multiplexed PCR were F-primer (TCTCAGCCCAYGGTGAGCATTA) and R-primer (TGGAGTAGGGGTCAGTATAGGGATTG). Using the ABI 3130xl genetic analyzer (Applied Biosystems), we verified the

SNaPshot products. The genotypes were clarified using GeneMapper 4.0 software (Applied Biosystems). Approximately 10% of the samples were randomly selected for verification, and all results were 100% concordant.

Statistical analysis

All statistical analyses were performed using the SPSS statistical software package (version 19.0; IBM, Armonk, NY, USA). The correlations between TS (rs34743033)/RUNX1 (rs2014300) SNPs and clinicopathologic parameters were confirmed using Pearson's chi-squared test for categorical variables. Kaplan–Meier survival curves and the log-rank test were used to estimate the affiliation of TS (rs34743033) and RUNX1 (rs2014300) SNPs with the prognosis of GC. A univariate or multivariate Cox regression analysis was applied to confirm HRs and the independence of effects. All tests were two-sided, and $p<0.05$ was considered statistically significant.

Ethics statement

All participants provided their written informed consent as per the ethics protocol approved by the Institutional Review Board of Nanjing Medical University (Nanjing, People's Republic of China).

Results

Study patients' characteristics

In total, 661 samples were recruited in the present study after curative surgery. Among them, 261 patients received PF-based adjuvant chemotherapy and 400 patients had not received chemotherapy. Clinical and pathological factors in the two tranches were not observed to have any significant differences ($p>0.05$). Patients' characteristics and clinical information are summarized in Table 1. Approximately 119 patients and 17 patients were excluded from TS and RUNX1 analysis due to missing genotype information. The TS (rs34743033) genotype frequency distribution in all patients was 49.6% (328 patients) for the 3R/3R variant, 28.6% (189 patients) for the 2R/3R variant, and 3.8% (25 patients) for the 2R/2R variant. For RUNX1 (rs2014300), the genotype frequency distribution was 75.2% (498 patients) for the GG variant, 20.6% (136 patients) for the GA variant, and 1.7% (11 patients) for the GG variant. The genotype distributions of the two SNPs confirmed no distinction between the two cohorts. The median OS time was 54 months (95% CI 41.6–66.4) in all patients. However, stage III was related to a weaker median OS of 41 months than stage II (mean OS of 88 months, $p<0.001$). The OS time of the samples that received adjuvant chemotherapy was 62 months, but this was not a statistically significant difference compared to patients who had not received adjuvant chemotherapy (50 months, $p=0.322$). In the adjuvant chemotherapy group, 163 patients

Table 1 Characteristics of the two cohorts of the gastric cancer patients

Clinicopathologic features	Chemotherapy (n=261)	No chemotherapy (n=400)	*p*
Age (years)	58.61±9.501	61.86±10.401	0.134
Sex			
Male	212	305	0.123
Female	49	95	
Tumor size[a]			
<5 cm	143	212	0.357
≥5 cm	118	188	
Tumor site[b]			
Noncardia	98	144	0.426
Cardia	163	256	
Depth of invasion[c]			
T1	5	6	0.897
T2	36	54	
T3	215	315	
T4	5	25	
Lymph node metastasis[c]			
N0	55	90	0.702
N1	143	223	
N2	60	79	
N3	3	8	
Lauren classification[b]			
Intestinal type	75	122	0.292
Diffuse type	186	278	
Differentiation			
Well to moderate	132	212	0.139
Poorly	69	117	
Mucinous/signet-ring cell	60	71	
Drinking			
Nondrinker	247	372	0.091
Drinker	14	28	
Smoking			
Nonsmoker	239	370	0.388
Smoker	22	30	
rs34743033			
3R/3R	135	193	0.494
2R/3R	73	116	
2R/2R	15	10	
rs2014300			
G/G	208	289	0.077
G/A	47	89	
A/A	3	8	

Notes: [a]Tumor size was measured by the length of the tumor. [b]Partial data were not available and statistics were based on available data. [c]Data were defined according to the TNM classification (AJCC 8th edition of the American Joint Commission on Cancer Staging Manual)[42] for gastric cancer.
Abbreviations: A, adenine; G, guanine; T, thymine; TNM, tumor, node and metastasis.

received a fluorouracil (CF) and cisplatin regimen and 103 patients received fluorouracil and oxaliplatin. The median OS of fluorouracil and oxaliplatin regimen-treated patients was not statistically significant compared to CF-treated patients (p=0.06).

TS and RUNX1 polymorphisms predicted OS in GC patients receiving PF-based adjuvant chemotherapy

To confirm the role of the two gene polymorphisms in predicting clinical prognosis, the TS (rs34743033) and RUNX1 (rs2014300) genotypes with OS in the two cohorts were determined using Cox regression statistics (Table 2). For the without chemotherapy group, the two genotypes were not associated with the OS of patients with GC. However, in the adjuvant chemotherapy cohort, TS (rs34743033) 2R/3R and 2R/2R variant genotypes increased the risk of death (HR 1.604, 95% CI 1.068–2.410, p=0.021) compared with the 3R/3R genotype (Table 2). Patients who carried TS (rs34743033) 2R/3R and 2R/2R variant genotypes were observed to have only 39 months in median survival time; (MST), whereas patients who carried TS (rs34743033) 3R/3R variant genotypes had 67.4 months in MST (Figure 1). Therefore, the TS (rs34743033) 2R/3R and 2R/2R variant genotypes were significantly associated with OS time in PF-based adjuvant chemotherapy patients. Meanwhile, RUNX1 (rs2014300) genotypes were associated with OS time in the codominant mode (p=0.007), where the RUNX1 (rs2014300) GA and AA variant genotypes enhanced the risk of death (HR 1.309; 4.551; 95% CI 0.845–2.028, 1.421–14.568) compared with the GG genotype. Interestingly, the AA variant genotypes were observed to have a higher risk. In the recessive model, the AA genotypes markedly increased the risk of death in GC patient (HR 4.820, 95% CI 1.513–15.353, p=0.003) compared with the GG and GA genotypes (Table 2). The RUNX1 (rs2014300) genotypes GG, GA, and AA produced a median survival time of 62, 37, and 12 months, respectively (Figure 1).

TS and RUNX1 polymorphisms were associated with the OS of GC among the PF-based chemotherapy regimen subgroup

The TS and RUNX1 gene polymorphisms were associated with the survival of GC patients treated with chemotherapy as already mentioned. The patients were then stratified by chemotherapy regimens (based on cisplatin and oxaliplatin)

Table 2 Associations of TS (rs34743033) and RUNX1 (rs2014300) with gastric cancer-specific overall survival in both cohorts

Genetic models	Genotype	Adjuvant chemotherapy					No chemotherapy				
		Patients	Deaths	MST	*p*	HR (95% CI)[a]	Patients	Deaths	MST (month)	*p*	HR (95% CI)[a]
TS											
	3R/3R	135	48	67.4[b]	p=0.021	1.000	193	99	59.0	0.516	1.000
	2R/3R+2R/2R	88	45	39.0		1.604 (1.068–2.410)	126	60	70.0		0.900 (0.653–1.240)
RUNX1											
Codominant model	G/G	208	88	62.0[b]	p=0.007	1.000	289	158	48.0	0.509	1.000
	G/A	47	26	37.0		1.309 (0.845–2.028)	89	45	53.0		0.914 (0.656-1.273)
	A/A	3	3	12.0		4.551 (1.421–14.568)	8	3	98.0		0.533 (0.170–1.677)
Dominant model	G/G	208	88	62.0[b]	p=0.127	1.000	289	158	65.0	0.418	1.000
	G/A+A/A	50	29	36.0		1.381 (0.907–2.102)	97	48	76.0		0.876 (0.634–1.210)
Recessive model	G/G+G/C	255	114	62.0	p=0.003	1.000	378	203	48.0	0.306	1.000
	A/A	3	3	12.0		4.820 (1.513–15.353)	8	3	98.0		0.558 (0.178–1.747)

Notes: [a]Calculated in Cox regression and adjusted for age, sex, and tumor stage. [b]Mean survival time was presented when the median survival time could not be measured
Abbreviations: A, adenine; C, cytosine; G, guanine; MST, median survival time; RUNX1, runt-related transcription factor 1; TS, thymidylate synthase.

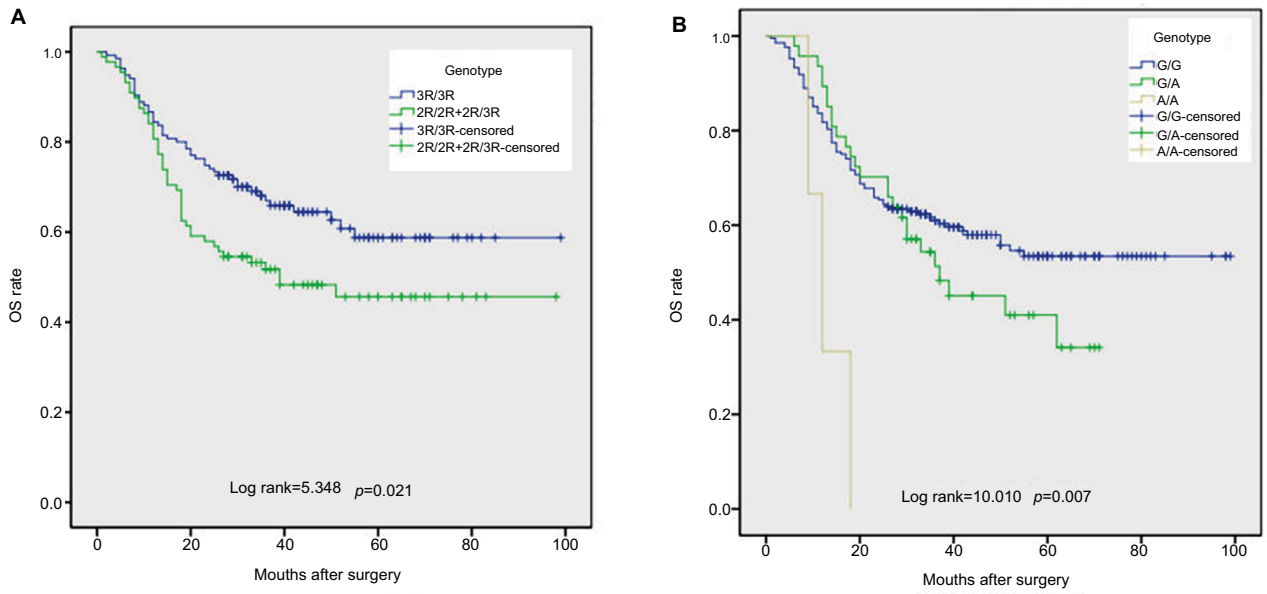

Figure 1 Kaplan–Meier survival curves of TS (rs34743033) and RUNX1 (rs2014300) for OS in GC patients who received adjuvant chemotherapy. (**A**) TS (rs34743033) associated with poor OS in GC patients. (**B**) RUNX1 (rs2014300) associated with poor OS in GC patients.
Abbreviations: GC, gastric cancer; OS, overall survival; RUNX1, runt-related transcription factor 1; TS, thymidylate synthase.

Table 3 Association between the dominant model of TS/RUNX1 and overall survival of gastric cancer among chemotherapy regimen subgroup.

Chemotherapy based on CDDP					
Genotype	**Patients**	**Deaths**	**MST (months)**	**log-rank *p***	**HR (95% CI)[a]**
3R/3R	66	25	66.9[b]	*p*=0.031	1.000
2R/3R+2R/2R	57	33	27.0		1.754 (1.041–2.954)
G/G	117	54	59.6[b]	*p*=0.146	1.000
G/A+A/A	29	20	33.0		1.455 (0.870–2.432)
Chemotherapy based on L-OHP					
Genotype	**Patients**	**Deaths**	**MST (months)**	**log-rank *p***	**HR (95% CI)[a]**
3R/3R	63	18	53.4	*p*=0.248	1.000
2R/3R+2R/2R	30	12	47.5		1.531 (0.736–3.183)
G/G	80	27	51.5	*p*=0.507	1.000
G/A+A/A	18	5	55.8		0.725 (0.279–1.887)

Notes: [a]HR adjusted for sex, age, TNM stage. [b]Mean the median survival time could not be measured.
Abbreviations: HR, hazard ratio; MST, median survival time; CI, confidence interval; RUNX1, runt-related transcription factor 1; TNM, tumor, node and metastasis; TS, thymidylate synthase; L-OHP, oxaliplatin; CDDP, cisplatin.

using the Kaplan–Meier survival curves, Cox regression, and log-rank tests to confirm the association of rs34743033 and rs2014300 genotypes in stratified patient OS times. We found that the TS (rs34743033) 2R/3R and 2R/2R variant genotypes had a negative effect on the OS of patients who received chemotherapy based on cisplatin (HR 1.754, 95% CI 1.041–2.954, *p*=0.031, Table 3). However, no similar results were observed in the subgroup with chemotherapy regimens based on oxaliplatin (L-OHP). The Kaplan–Meier survival curves are shown in Figure 2. However, the RUNX1 (rs2014300) genotypes were not significantly associated with the OS of patients who received chemotherapy based on cisplatin or oxaliplatin in the dominant models. Therefore, these results demonstrated that 2R/3R and 2R/2R were hazards for patients' prognosis involving adjuvant chemotherapy based on cisplatin (CDDP).

TS and RUNX1 polymorphisms were associated with specific subtypes of GC

The association between TS (rs34743033), RUNX1 (rs2014300), and OS in the adjuvant chemotherapy patients was further researched by a stratified analysis of tumors,

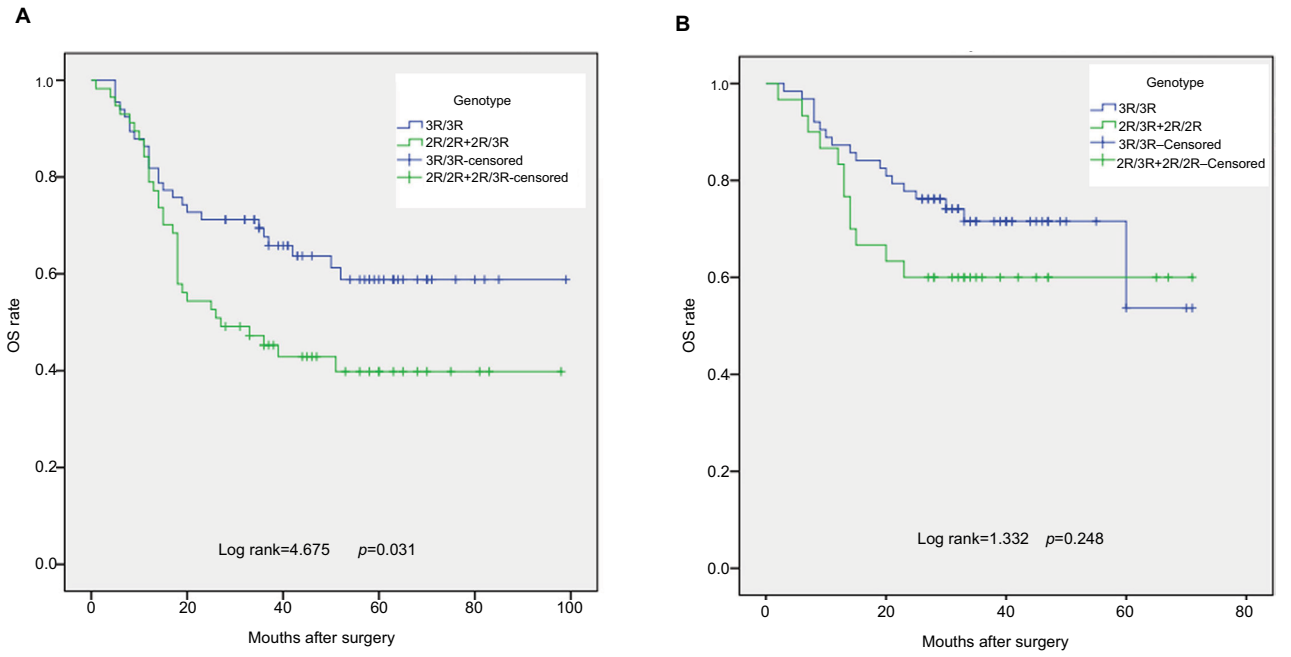

Figure 2 OS curve associated with TS (rs34743033) polymorphism in gastric cancer patients who received adjuvant chemotherapy. (**A**) TS (rs34743033) 2R/3R and 2R/2R variant genotypes had negative effect on OS of patients receiving chemotherapy based on cisplatin (p=0.031). (**B**) TS (rs34743033) 2R/3R and 2R/2R variant genotypes had no difference on OS in oxaliplatin-based chemotherapy (p=0.248).
Abbreviations: OS, overall survival; TS, thymidylate synthase.

Table 4 Stratified analysis of two polymorphisms with gastric cancer overall survival in patients who received adjuvant chemotherapy (n=261)

Variables	rs34743033				rs2014300			
	Deaths/patients				Deaths/patients			
Clinicopathologic features	3R/3R	2R/3R+2R/2R	p	HR (95% CI)[a]	GG	GA/AA	p	HR (95% CI)[a]
Tumor size								
<5 cm	27/79	26/48	0.033	1.777 (1.035–3.050)	44/144	17/28	0.081	1.635 (0.932–2.868)
≥5 cm	21/56	19/40	0.23	1.454 (0.781–2.705)	44/94	12/22	0.775	1.097 (0.579–2.079)
Tumor site								
Noncardia	29/83	26/53	0.079	1.597 (0.940–2.714)	60/136	15/25	0.367	1.294 (0.734–2.228)
Cardia	19/52	19/35	0.128	1.627 (0.861–3.077)	28/72	14/25	0.151	1.589(0.836–3.020)
Lauren classification[b]								
Intestinal type	14/40	20/28	0.006	2.520(1.267–5.014)	27/58	10/17	0.999	1.000 (0.483–2.068)
Diffuse type	34/95	25/60	0.425	1.232 (0.753–2.066)	61/150	19/33	0.057	1.637 (0.977–2.744)
Differentiation								
Well to moderate	21/64	16/45	0.702	1.183 (0.617–2.267)	38/106	14/23	0.023	2.001 (1.082–3.703)
Poorly	13/36	20/27	0.008	2.501 (1.238–5.054)	26/54	9/15	0.973	0.987 (0.462–2.109)
Mucinous/signet-ring cell	14/35	9/16	0.252	1.621 (0.700–3.755)	24/48	6/12	0.819	0.910 (0.366–2.217)

Notes: [a]Calculated in multivariate Cox regression and adjusted by age, sex, and tumor stage. [b]Partial data were not available and statistics were based on available data.
Abbreviations: A, adenine; G, guanine.

including clinical features and demographic characteristics (Table 4). Compared to the 3R/3R genotypes, the 2R/3R and 2R/2R variant genotypes were significantly related to poor survival in GC patients with intestinal-type tumors (HR 2.520, 95% CI 1.267–5.014, p=0.006), a tumor size less than 5 cm (HR 1.777, 95% CI 1.035–3.050, p=0.033), and poor differentiation (HR 2.501, 95% CI 1.238–5.054, p=0.008). A stratification analysis of RUNX1 (rs2014300) showed that the GA/AA genotype had a significant association with inferior survival in well to moderately differentiated tumors (HR 2.001, 95% CI 1.082–3.703, p=0.023).

Discussion

The present research showed that combination chemotherapy increases OS compared to single-agent therapy in GC.[21] However, no accurate biomarker exists to guide the selection

of chemotherapy regimens or determine the prognosis of patients with advanced GC. In the present study, we confirmed the clinical prognostic significance of the genetic polymorphisms of TS (rs34743033) and RUNX1 (rs2014300) in Chinese GC patients. The TS (rs34743033) 2R/3R, 2R/2R, and RUNX1 (rs2014300) A/A genotypes were associated with poorer OS only in patients who had received PF-based adjuvant chemotherapy. Furthermore, we demonstrated for the first time that TS (rs34743033) 2R/3R and 2R/2R genotypes generated poorer OS of GC in patients who had received chemotherapy based on CDDP, compared with the 3R/3R genotype. This result has not been reported in other studies. However, in oxaliplatin therapy, the TS (rs34743033) 2R/3R, 2R/2R, and RUNX1 (rs2014300) A/A genotypes had no association with GC patients' OS.

For the past few years, numerous studies have confirmed that the polymorphisms of TS affect the sensitivity of 5-fluorouracil; however, these results remain controversial. In 221 colorectal cancer patients, Iacopetta et al discovered that the TS 2R/2R or 2R/3R genotype showed significant long-term survival from 5-FU-based chemotherapy treatment compared with the other genotypes.[13] However, another study by Martinez-Balibrea et al found that the TS 2R/2R or 2R/3R genotype was associated with poor survival in 71 advanced colorectal cancer patients.[22] Pullarkat et al confirmed a higher response rate in 2R/2R patients compared with 50 2R/3R or 3R/3R colorectal cancer patients.[23] In a study involving 25 non-small cell lung cancer patients, Arévalo et al confirmed that the 3R/3R polymorphism is associated with a superior OS.[14] For GC patients receiving 5-FU chemotherapy regimens, Ishida et al showed that the 2R/2R and 2R/3R genotypes were associated with a long OS compared with the 3R/3R genotype in 115 GC patients who had received 5-FU-based chemotherapy.[15] Ott et al also showed that 3R/3R was a risk factor for GC patient survival.[24] However, Jakobsen et al confirmed that the 3R/3R genotype responded better to 5-FU and was associated with a longer survival time in 150 GC patients.[25] Accordingly, the TS polymorphism in 5-FU-based chemotherapy treatments is strongly contraindicated. A possible explanation for this may involve tumor type, the number of cases included, genetic background, and genetic correlation. In our study, we found that the 3R/3R genotype was associated with a superior OS in GC patients who had received 5-FU chemotherapy regimens compared with 2R/3R and 2R/2R. The results in the present study were similar to Jakobsen et al's findings (HR 1.604, 95% CI, 1.068–2.410, p=0.021, Table 2). The 3R allele could enhance TS protein expression but not mRNA expression when compared with the 2R allele in colorectal cancer patients.[26] Higher TS protein expression can improve the formation of the ternary complex, resulting in increased 5-FU sensitivity.

In addition to these results, we also found that patients with the 3R/3R genotype who had received CDDP-based adjuvant chemotherapy after surgery had a better prognosis (HR 1.754, 95% CI 1.041–2.954, p=0.031, Table 3). However, no similar results were observed in chemotherapy regimens based on oxaliplatin (L-OHP). It is already known that the basic mechanism of platinum drug anti-tumor effects also involves binding to the N7 reactive center on purine residues and can later cause DNA damage in cancer cells, block cell division, and may lead to apoptotic cell death.[27] Although oxaliplatin and cisplatin are the standard platinum chemotherapy drugs, the mechanism of drug resistance is different. This difference may explain why the TS (rs34743033) 3R/3R genotype can forecast responses to cisplatin rather than oxaliplatin. It has been reported that many factors cause cisplatin resistance, including intracellular platinum compound accumulation,[28] enhanced DNA damage repair,[29] the inactivation of apoptosis,[30] the activation of the epithelial–mesenchymal transition,[31] and the modification in DNA methylation.[32] Certainly, some classic cell signaling pathways are also associated with cisplatin resistance and involve the activation of STAT3,[33] Wnt signaling pathway,[34] and downregulated Myc expression.[35] In our research, we confirmed that the TS (rs34743033) 3R/3R genotype was significantly associated with the OS of GC patients who received cisplatin and 5-FU chemotherapy regimens. Therefore, we hypothesize that rs34743033 variants may be related to cisplatin resistance; however, the underlying mechanism requires further investigation. In addition, the TS (rs34743033) 2R/3R and 2R/2R variant genotypes were significantly associated with poorer survival in GC patients with poorly differentiated intestinal tumors less than 5 cm in size.

The RUNX (runt-related transcription factor) family of genes encodes the DNA-binding α-chain partners of the heterodimeric core-binding factor complex and participate in cell-cycl exit, continued proliferation, differentiation, and stage-specific self-renewal. RUNX1 belongs to the runt-related transcription factor family and can regulate hematopoietic cell differentiation, apoptosis, and self-renewal.[16] RUNX1 functions as an oncogene and is commonly found in several cancers, such as breast, uterus, gastrointestinal tract, and T-ALL,[36–40] but it also acts as a tumor suppressor in the gastrointestinal tract.[41] Bye et al showed that the RUNX1 variant rs2014300 reduced the risk of oesophageal squamous

cell carcinoma in Chinese populations but increased the risk of OSCC in mixed ancestry populations.[19] However, Yuan et al demonstrated that rs2014300 identified by the genome-wide association study of esophageal squamous cell carcinoma was not associated with HNC risk.[20]

The present study was first to report that the rs2014300 A/A genotype was significantly associated with the OS of GC patients who received adjuvant chemotherapy after surgery (HR 4.820, 95% CI 1.513–15.353, p=0.003). However, this genotype had no significant association with the OS of GC patients in the nonchemotherapy groups. Therefore, the A allele may be a risk factor in GC patient prognosis. Incidentally, the rs2014300 GA/GG genotypes are related to patients with tumorigenesis in well to moderately differentiated tumors.

Conclusion

Our results demonstrated that TS (rs34743033) polymorphisms can predict the prognosis of postoperative stage II or III GC patients who do and do not benefit from adjuvant chemotherapy. The 3R/3R genotype is a protective factor only in GC patients based on cisplatin. RUNX1 (rs2014300) polymorphisms have been shown to be significantly related to survival time in patients undergoing postoperative adjuvant chemotherapy, and the A allele is probably a risk factor for the prognosis of these patients. Moreover, TS (rs34743033) and RUNX1 (rs2014300) showed concrete effects on postoperative adjuvant chemotherapy in a specific subtype of GC patients. These results provide a new perspective on the selection of a plan for postoperative adjuvant chemotherapy in GC patients.

Disclosure

The authors report no conflicts of interest in this work.

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