

***ITGA1* polymorphisms and haplotypes are associated with gastric cancer risk in a Korean population**

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polymorphisms and haplotypes of the *ITGA1* gene and the risk of gastric cancer.

METHODS: The study subjects were 477 age- and sex-matched case-control pairs. Genotyping was performed for 15 single nucleotide polymorphisms (SNPs) in *ITGA1*. The associations between gastric cancer and these SNPs and haplotypes were analyzed with multivariate conditional logistic regression models. Multiple testing corrections were carried out following methodology for controlling the false discovery rate. Gene-based association tests were performed using the versatile gene-based association study (VEGAS) method.

RESULTS: In the codominant model, the ORs for SNPs *rs2432143* (1.517; 95%CI: 1.144-2.011) and *rs2447867* (1.258; 95%CI: 1.051-1.505) were statistically significant. In the dominant model, polymorphisms of *rs1862610* and *rs2447867* were found to be significant risk factors, with ORs of 1.337 (95%CI: 1.029-1.737) and 1.412 (95%CI: 1.061-1.881), respectively. In the recessive model, only the *rs2432143* polymorphism was significant (OR = 1.559, 95%CI: 1.150-2.114). The C-C type of *ITGA1* haplotype block 2 was a significant protective factor against gastric cancer in the both codominant model (OR = 0.602, 95%CI: 0.212-0.709, *P* = 0.021) and the dominant model (OR = 0.653, 95%CI: 0.483-0.884). The *ITGA1* gene showed a significant gene-based association with gastric cancer in the VEGAS test. In the dominant model, the A-T type of *ITGA1* haplotype block 2 was a significant risk factor (OR = 1.341, 95%CI: 1.034-1.741). SNP *rs2447867* might be related to the severity of gastric epithelial injury due to inflammation and, thus, to the risk of developing gastric cancer.

CONCLUSION: *ITGA1* gene SNPs *rs1862610*, *rs2432143*, and *rs2447867* and the *ITGA1* haplotype block that includes SNPs *rs1862610* and *rs2432143* were significantly associated with gastric cancer.

Abstract

AIM: To evaluate the association between the genetic

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Key words: Integrin; *ITGA1*; Gastric cancer; Polymorphism; Haplotype

Core tip: There are few studies addressing the role of the integrin $\alpha 1$ subunit in the development of gastric cancer. To the best of our knowledge, this study is the first to show that *ITGA1* gene single nucleotide polymorphisms and haplotypes are associated with gastric cancer risk.

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INTRODUCTION

Gastric cancer is the second most common cancer in South Korea and represents the second leading cause of cancer death for both men and women worldwide^[1,2]. Approximately one million new cases of stomach cancer are estimated to have occurred (989000 cases, 7.8% of the total), currently making it the fourth most common malignancy in the world, following cancers of the lung, breast and colo-rectum^[2]. Epidemiological studies have provided evidence that a high intake of salt and nitrite-rich foods and *Helicobacter pylori* (*H. pylori*) infection are associated with a high incidence of gastric cancer in South Korea^[3-7].

The risk of developing gastric cancer is estimated to be increased 2-6 fold in patients with *H. pylori* infection^[8]. The risk of gastric cancer among individuals infected with *H. pylori* is influenced by bacterial virulence. The most widely studied *H. pylori* virulence factors are the *cag* (cytotoxin-associated gene) antigens^[9]. Compared to individuals infected with *cagA*-negative *H. pylori* strains, those infected with *cagA*-positive *H. pylori* strains show a higher risk of developing gastric cancer^[10]. To introduce *cagA* into host cells, the *cagL* protein of *H. pylori* binds to integrins on the basolateral surface of gastric epithelial cells^[11,12].

Integrins are members of a family of heterodimeric cell-surface proteins that mediate cell-matrix and cell-cell interactions. The 18 integrin α -subunits and 8 β -subunits together form at least 25 different integrins^[13]. Integrins mediate signaling events that are essential for stable cell adhesion, spreading, migration, survival, proliferation and differentiation. Several integrins, including $\alpha 1 \beta 1$, bind to extracellular matrix proteins present in the basal membrane of mature vessels^[14,15]. The tumor progression and metastasis of various cancers are associated with integrins^[16,17].

The *ITGA1* gene, located on chromosome 5q11.2,

encodes the integrin $\alpha 1$ subunit, which is involved in the adhesion of gastric cancer cells to the peritoneum. The adhesion of integrin $\alpha 1$ -positive gastric cancer cells to the extracellular matrix is a critical process in peritoneal dissemination^[18,19]. There are few studies addressing the roles of integrins in the development of gastric cancer. An association with an increased risk of gastric cancer has only been reported for the *ITGA2* C807T polymorphism in a Chinese population^[20]. As the level of integrin $\alpha 1 \beta 1$ is up-regulated in association with inflammation of the gastrointestinal tract mucosa, which is the first step in gastric carcinogenesis^[21], it is possible that the integrin $\alpha 1$ subunit plays an important role in gastric cancer development.

The purpose of this study was to evaluate the association between the genetic polymorphisms and haplotypes of the *ITGA1* gene and the risk of gastric cancer.

MATERIALS AND METHODS

Study subjects

This subjects included in this study consisted of 477 newly diagnosed gastric cancer patients and an equal number of age- (within 3 years) and sex-matched controls. The diagnoses of the gastric cancer patients were all histologically confirmed at Chungbuk National University Hospital and Eulji University Hospital, which are located in a geographically central region of South Korea. Controls were selected from individuals receiving routine medical examinations in these hospitals, and individuals with a previous diagnosis of any type of cancer were excluded. Trained interviewers used a structured questionnaire including questions about demographic factors, smoking habits, alcohol consumption and dietary habits to interview all subjects who provided written informed consent. Peripheral blood samples were collected from all subjects. This study was approved by the institutional review boards of Chungbuk National University Hospital, South Korea (IRB No. 2011-09-071).

Selection of single nucleotide polymorphisms in *ITGA1*

At the International HapMap Project website (<http://hapmap.ncbi.nlm.nih.gov/>), tag SNPs were selected using a cut-off minimum minor allele frequency in the JPT population of 0.05 and pairwise tagging ($r^2 = 1-0.8$). SNPs that significantly deviated from Hardy-Weinberg equilibrium were discarded.

Genomic DNA was extracted from whole blood using the QuickGene-810 nucleic acid isolation system (Fujifilm, Tokyo, Japan) and the QuickGene DNA Whole Blood Kit (Kurabo, Osaka, Japan), in accordance with the manufacturer's instructions. DNA was stored at 4 °C until use. SNP genotyping was performed using a GoldenGate Genotyping Assay with VeraCode technology (Illumina, San Diego, CA, United States). A custom GoldenGate assay was designed for the analysis of the selected SNPs in the *ITGA1* gene. Those SNPs were then assessed for suitability for the GoldenGate genotyping platform, and

Table 1 Characteristics of the study subjects *n* (%)

| Variables | Controls (<i>n</i> = 477) | Cases (<i>n</i> = 477) | OR (95%CI) |
|-----------------------|----------------------------|-------------------------|------------------|
| Age (yr) | 57.8 ± 10.2 | 58.7 ± 9.9 | |
| mean ± SD | | | |
| Sex | | | |
| Males | 301 (63.1) | 301 (63.1) | |
| Females | 176 (36.9) | 176 (36.9) | |
| Smoking status | | | |
| Non-smokers | 225 (47.6) | 194 (41.0) | 1.00 (reference) |
| Smokers | 248 (52.4) | 279 (59.0) | 1.64 (0.95-2.84) |
| Alcohol intake status | | | |
| Non-drinkers | 194 (40.7) | 189 (39.6) | 1.00 (reference) |
| Drinkers | 283 (59.3) | 288 (60.4) | 1.18 (0.71-1.76) |

the analysis was carried out on the validated SNPs. The average call rate was 99.2%. Genotyping was carried out by MacroGen (Seoul, South Korea).

Statistical analysis

The study power was calculated using the “case-control for discrete traits” mode in the Genetic Power Calculator^[22]. The following parameters were applied: risk allele frequency -0.4, alpha error -0.01, and disease prevalence -0.1%. The power of a codominant model was 0.7768 when the heterozygous OR was set to 1.5. For a dominant model, when the OR for a genotype with one or 2 risk allele(s) was taken as 2, the power was 0.8821. When a value of 2 was input for the OR for a genotype with 2 risk allele(s), the power of a recessive model was 0.8182.

Testing for deviation from the HWP was performed for each SNP in both cases and in controls using Pearson's χ^2 test. *D* values were measured using Lewontin's method for all combinations of biallelic loci^[23,24], and linkage disequilibrium blocks were structured using Haploview version 4.2 (Daly Lab at the Broad Institute Cambridge, MA, United States). Haplotype blocks were constructed and statistically compared between cases and controls with SNP Analyzer version 2.0 (ISTEC Inc., Goyang, South Korea).

Student's *t* test was used to compare continuous variables between patients and control subjects. Associations between gastric cancer and the investigated SNPs and haplotypes were estimated *via* the OR and their corresponding 95%CI derived from multivariate conditional logistic regression models, after adjusting for potential confounding factors such as age, sex, smoking history, and alcohol intake. The genotypes of major homozygotes, heterozygotes and minor homozygotes were coded as 0, 1, and 2 in the codominant model, 0, 1 and 1 in the dominant model, and 0, 0 and 1 in the recessive model, respectively. Multiple testing corrections were carried out using Benjaminin and Hochberg's methods for controlling the false discovery rate (FDR)^[25]. A two-sided adjusted *P* value of < 0.05 was considered statistically significant. FDR *Q* values were calculated separately for the SNPs and haplotypes based on these numbers. Gene-based association tests were performed using the versatile gene-based association study (VEGAS) method^[26]. For these statistical analyses, SAS version 9.2 (SAS Institute,

Cary, NC, United States) was employed.

RESULTS

Patient characteristics are summarized in Table 1. No significant difference was observed between the distributions of the age, sex, and smoking and drinking habits of the cases and controls.

Table 2 lists and provides the frequencies of the 15 selected SNPs in the study subjects. None of the polymorphisms were significantly deviated from Hardy-Weinberg equilibrium. All the minor allele frequencies of the cases and controls were greater than 10%.

The haplotype linkage disequilibrium blocks and haplotype frequencies for *ITGA1* are shown in Figure 1. *D* values were measured using Lewontin's method. Four block haplotypes were constructed using Haploview version 4.2. The common haplotypes (frequency > 10%) in each block accounted for 84.2%, 99.8%, 91.6% and 99.9% for the cases and 85.7%, 99.8%, 91.2% and 99.9% for the controls.

The observed associations between the genetic polymorphisms in the *ITGA1* gene and the risk of gastric cancer are shown in Table 3. In the codominant model, the OR of 1.517 obtained for SNP *rs2432143* (95%CI: 1.144-2.011; *P* = 0.003; FDR *Q* = 0.045) was statistically significant, even after controlling the FDR, and that for *rs2447867*, of 1.258 (95%CI: 1.051-1.505; *P* = 0.012; FDR *Q* = 0.090), was marginally significant. In the dominant model, the *rs1862610* and *rs2447867* polymorphisms were not statistically significant risk factors for gastric cancer, displaying ORs of 1.337 (95%CI: 1.029-1.737; *P* = 0.029; FDR *Q* = 0.217) and 1.412 (95%CI: 1.061-1.881; *P* = 0.018; FDR *Q* = 0.217), respectively. Only the *rs2432143* polymorphism was marginally significant in the recessive model, exhibiting an OR of 1.559 (95%CI: 1.150-2.114; *P* = 0.004; FDR *Q* = 0.060).

When the *P* values for the minor alleles of the codominant, dominant and recessive models were subjected to the VEGAS test, no significant gene-based associations were found. However, when the lower *P* value generated by the dominant and recessive models was input for every SNP, the value of the test statistic was 29.622, which was statistically significant (*P* = 0.037).

Four haplotype blocks were constructed using SNP Analyzer version 2.0. These blocks were evaluated for an association with the risk of gastric cancer (Table 4). The C-C type of *ITGA1* haplotype block 2 was marginally significant in the codominant model (OR = 0.602, 95%CI: 0.212-0.709; *P* = 0.021; FDR *Q* = 0.063) and was a significant protective factor against gastric cancer in the dominant model (OR = 0.653, 95%CI: 0.483-0.884; *P* = 0.006; FDR *Q* = 0.018). In the dominant model, the A-T type of *ITGA1* haplotype block 2 was a significant risk factor (OR = 1.341, 95%CI: 1.034-1.741; *P* = 0.027; FDR *Q* = 0.045). No haplotype block was found to be significant in the recessive model.

Table 2 Frequency of *ITGA1* polymorphisms in cases and controls

| SNP | Chromosomal position | Amino acid change | Genotype case/control | | | | Case | | Control | |
|-------------------|----------------------|-------------------|-----------------------|---------|---------|---------|-----------|------------------|-----------|------------------|
| | | | | | | | Frequency | HWE ¹ | Frequency | HWE ¹ |
| <i>rs13188662</i> | 2686006 | - | AA | AG | GG | N | 0.280 | 0.573 | 0.276 | 0.597 |
| | | | 249/253 | 186/186 | 40/38 | 475/477 | | | | |
| <i>rs11740785</i> | 2707341 | - | AA | AC | CC | N | 0.241 | 0.866 | 0.229 | 0.259 |
| | | | 279/290 | 166/156 | 32/31 | 477/477 | | | | |
| <i>rs1820167</i> | 2713715 | - | AA | AG | GG | N | 0.435 | 0.806 | 0.420 | 0.904 |
| | | | 151/162 | 237/229 | 89/83 | 477/477 | | | | |
| <i>rs1862610</i> | 2722239 | - | CC | AC | AA | N | 0.369 | 0.861 | 0.387 | 0.484 |
| | | | 172/205 | 223/192 | 82/80 | 477/477 | | | | |
| <i>rs2432143</i> | 2725674 | - | TT | TC | CC | N | 0.104 | 0.671 | 0.146 | 0.658 |
| | | | 382/346 | 87/121 | 8/10 | 477/477 | | | | |
| <i>rs2447867</i> | 2751733 | C/C | CC | TC | TT | N | 0.490 | 0.742 | 0.430 | 0.769 |
| | | | 123/155 | 241/229 | 113/89 | 477/473 | | | | |
| <i>rs4865745</i> | 2770258 | - | TT | TC | CC | N | 0.270 | 0.892 | 0.268 | 0.124 |
| | | | 253/247 | 186/198 | 35/28 | 474/473 | | | | |
| <i>rs13163497</i> | 2773367 | - | GG | AG | AA | N | 0.110 | 0.409 | 0.108 | 0.515 |
| | | | 375/381 | 97/89 | 4/7 | 476/477 | | | | |
| <i>rs1904163</i> | 2780355 | - | CC | TC | TT | N | 0.298 | 0.196 | 0.272 | 0.698 |
| | | | 238/245 | 184/187 | 48/33 | 470/465 | | | | |
| <i>rs1466445</i> | 2789486 | - | CC | TC | TT | N | 0.460 | 0.783 | 0.455 | 0.696 |
| | | | 139/142 | 233/229 | 101/100 | 473/471 | | | | |
| <i>rs16880453</i> | 2789866 | - | GG | GC | CC | N | 0.466 | 0.914 | 0.465 | 0.424 |
| | | | 133/130 | 235/243 | 100/98 | 468/471 | | | | |
| <i>rs2452864</i> | 2796757 | - | TT | TC | CC | N | 0.367 | 0.874 | 0.369 | 0.368 |
| | | | 190/183 | 224/230 | 63/59 | 477/472 | | | | |
| <i>rs1275659</i> | 2828018 | - | AA | AG | GG | N | 0.257 | 0.185 | 0.278 | 0.864 |
| | | | 256/247 | 192/189 | 26/37 | 474/473 | | | | |
| <i>rs1871186</i> | 2828974 | - | TT | TC | CC | N | 0.221 | 0.723 | 0.213 | 0.674 |
| | | | 287/296 | 166/157 | 22/23 | 475/476 | | | | |
| <i>rs988574</i> | 2835169 | E/G | TT | TC | CC | N | 0.180 | 0.723 | 0.183 | 0.674 |
| | | | 319/309 | 141/155 | 15/9 | 475/473 | | | | |

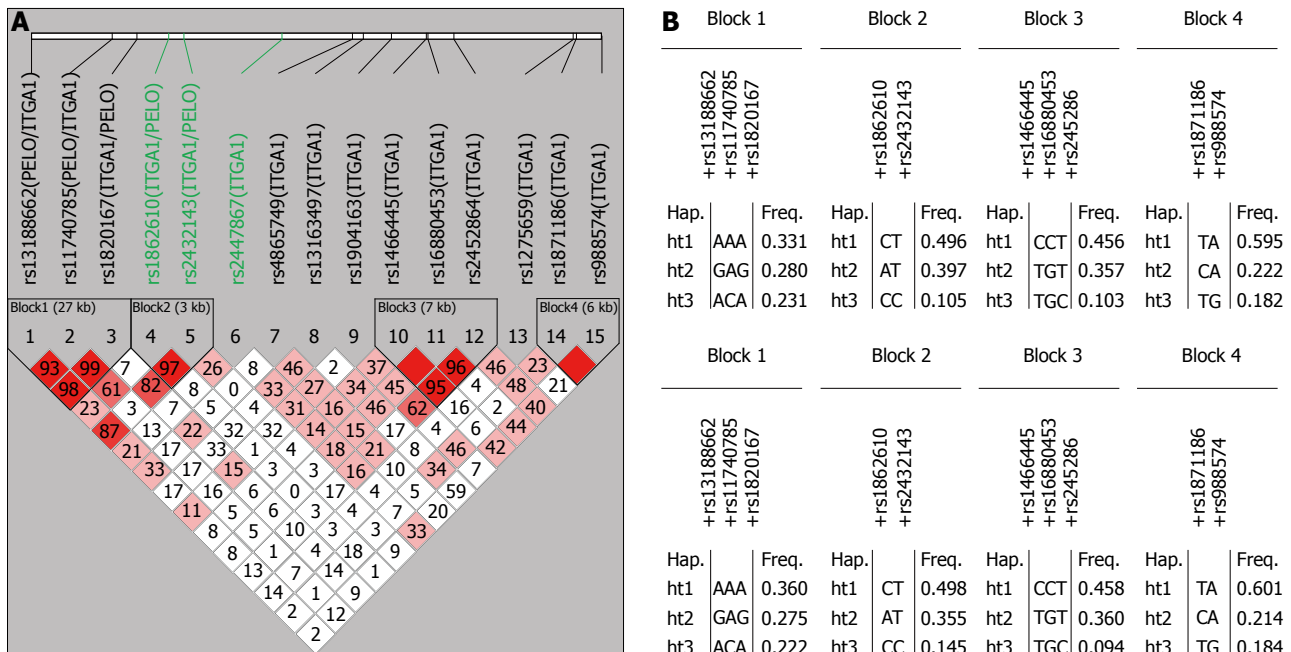
¹P value for deviation from Hardy-Weinberg Equilibrium (HWE). SNP: Single nucleotide polymorphism.**Figure 1** Haplotype linkage disequilibrium blocks and haplotype frequencies for *ITGA1*. A: Linkage disequilibrium (LD) blocks among *ITGA1* polymorphisms. Black squares indicate a statistically significant allelic association between a pair of single nucleotide polymorphisms, as measured by the *D* statistic; darker gray indicate higher values of *D*; B: Haplotype frequencies of *ITGA1* polymorphisms in cases (top) and controls (bottom).

Table 3 Association between *ITGA1* polymorphisms and gastric cancer in a case-control study of a Korean population

| SNP | Chromosomal position | Codominant | | | Dominant | | | Recessive | | |
|----------------------|----------------------|---------------------|----------------------|----------------|---------------------|----------------------|----------------|---------------------|----------------------|----------------|
| | | OR (95%CI) | P value ¹ | Q ² | OR (95%CI) | P value ¹ | Q ² | OR (95%CI) | P value ¹ | Q ² |
| rs13188662 | 2686006 | 1.040 (0.840-1.281) | 0.161 | 0.483 | 1.060 (0.811-1.379) | 0.689 | 0.866 | 1.060 (0.660-1.690) | 0.811 | 0.963 |
| rs11740785 | 2707341 | 1.069 (0.869-1.313) | 0.528 | 0.965 | 1.106 (0.848-1.442) | 0.457 | 0.866 | 1.032 (0.630-1.692) | 0.899 | 0.963 |
| rs1820167 | 2713715 | 1.066 (0.884-1.286) | 0.503 | 0.964 | 1.115 (0.846-1.468) | 0.440 | 0.866 | 1.043 (0.751-1.447) | 0.801 | 0.963 |
| rs1862610 | 2722239 | 1.151 (0.965-1.372) | 0.118 | 0.483 | 1.337 (1.029-1.737) | 0.029 | 0.217 | 1.029 (0.740-1.429) | 0.866 | 0.963 |
| rs2432143 | 2725674 | 1.517 (1.144-2.011) | 0.003 | 0.045 | 1.800 (0.603-5.371) | 0.292 | 0.883 | 1.559 (1.150-2.114) | 0.004 | 0.060 |
| rs2447867 | 2751733 | 1.258 (1.051-1.505) | 0.012 | 0.090 | 1.412 (1.061-1.881) | 0.018 | 0.217 | 1.303 (0.966-1.756) | 0.083 | 0.415 |
| rs4865745 | 2770258 | 1.016 (0.829-1.246) | 0.875 | 0.965 | 0.967 (0.750-1.247) | 0.795 | 0.863 | 1.269 (0.759-2.122) | 0.363 | 0.927 |
| rs13163497 | 2773367 | 1.021 (0.768-1.357) | 0.884 | 0.965 | 1.064 (0.781-1.449) | 0.693 | 0.866 | 0.571 (0.167-1.952) | 0.371 | 0.927 |
| rs1904163 | 2780355 | 1.157 (0.943-1.420) | 0.161 | 0.483 | 1.104 (0.849-1.436) | 0.461 | 0.866 | 1.593 (0.984-2.577) | 0.058 | 0.415 |
| rs1466445 | 2789486 | 1.013 (0.845-1.213) | 0.890 | 0.965 | 1.032 (0.778-1.368) | 0.829 | 0.883 | 1.000 (0.736-1.358) | 1.000 | 1.000 |
| rs16880453 | 2789866 | 1.000 (0.832-1.201) | 1.000 | 1.000 | 0.979 (0.734-1.305) | 0.883 | 0.883 | 1.025 (0.752-1.398) | 0.874 | 9.632 |
| rs2452864 | 2796757 | 0.986 (0.816-1.191) | 0.885 | 0.965 | 0.947 (0.728-1.233) | 0.687 | 0.883 | 1.056 (0.728-1.532) | 0.775 | 9.632 |
| rs1275659 | 2828018 | 1.136 (0.919-1.404) | 0.237 | 0.592 | 1.522 (0.899-2.575) | 0.117 | 0.585 | 1.095 (0.841-1.427) | 0.500 | 9.632 |
| rs1871186 | 2828974 | 1.043 (0.841-1.293) | 0.701 | 0.965 | 1.072 (0.828-1.388) | 0.597 | 0.866 | 0.957 (0.533-1.716) | 0.881 | 9.632 |
| rs988574 | 2835169 | 0.985 (0.772-1.256) | 0.901 | 0.965 | 0.927 (0.707-1.215) | 0.581 | 0.866 | 1.667 (0.729-3.808) | 0.225 | 0.843 |
| VEGAS statistics (P) | | 23.986 (0.105) | | | 16.823 (0.364) | | | 18.732 (0.260) | | |

¹P values for logistic analysis of three alternative models (codominant, dominant and recessive); ²False discovery rate Q value. When the lower P value generated by the dominant and recessive models was applied for every single nucleotide polymorphism (SNP), the value of the versatile gene-based association study (VEGAS) statistic was 29.622 (P = 0.037).

Table 4 Association between *ITGA1* haplotypes and gastric cancer

| Haplotypes | | Codominant | | | Dominant | | | Recessive | | |
|--------------|-----|---------------------|----------------------|----------------|---------------------|----------------------|----------------|---------------------|----------------------|----------------|
| | | OR (95%CI) | P value ¹ | Q ² | OR (95%CI) | P value ¹ | Q ² | OR (95%CI) | P value ¹ | Q ² |
| <i>ITGA1</i> | AAA | 0.771 (0.510-1.165) | 0.414 | 0.973 | 0.860 (0.666-1.112) | 0.250 | 0.750 | 0.819 (0.555-1.210) | 0.316 | 0.913 |
| Haplotype | GAG | 1.039 (0.643-1.678) | 0.973 | 0.973 | 1.030 (0.799-1.328) | 0.819 | 0.819 | 1.026 (0.644-1.636) | 0.913 | 0.913 |
| block 1 | ACA | 0.992 (0.559-1.760) | 0.768 | 0.973 | 1.088 (0.839-1.410) | 0.525 | 0.787 | 0.957 (0.544-1.683) | 0.879 | 0.913 |
| <i>ITGA1</i> | CT | 0.982 (0.688-1.407) | 0.640 | 0.640 | 1.072 (0.800-1.437) | 0.641 | 0.641 | 0.911 (0.679-1.223) | 0.536 | 0.536 |
| Haplotype | AT | 1.316 (0.686-1.407) | 0.086 | 0.129 | 1.341 (1.034-1.741) | 0.027 | 0.045 | 1.121 (0.784-1.603) | 0.532 | 0.536 |
| block 2 | CC | 0.602 (0.212-0.709) | 0.021 | 0.063 | 0.653 (0.483-0.884) | 0.006 | 0.018 | 0.661 (0.233-1.872) | 0.433 | 0.536 |
| <i>ITGA1</i> | CCT | 1.023 (0.707-1.480) | 0.677 | 0.794 | 0.934 (0.705-1.236) | 0.631 | 0.916 | 0.819 (0.555-1.210) | 0.316 | 0.913 |
| Haplotype | TGC | 0.973 (0.641-1.475) | 0.314 | 0.794 | 0.986 (0.761-1.278) | 0.916 | 0.916 | 1.026 (0.644-1.636) | 0.913 | 0.913 |
| block 3 | TGT | 1.418 (0.446-4.507) | 0.794 | 0.794 | 1.084 (0.782-1.505) | 0.627 | 0.916 | 0.957 (0.544-1.683) | 0.879 | 0.913 |
| <i>ITGA1</i> | TA | 0.938 (0.641-1.370) | 0.907 | 0.907 | 0.928 (0.658-1.310) | 0.671 | 0.671 | 0.997 (0.765-1.299) | 0.981 | 0.981 |
| Haplotype | CA | 0.983 (0.536-1.803) | 0.803 | 0.907 | 1.079 (0.832-1.400) | 0.567 | 0.671 | 0.952 (0.523-1.733) | 0.873 | 0.981 |
| block 4 | TG | 1.619 (0.698-3.756) | 0.320 | 0.907 | 0.925 (0.708-1.209) | 0.569 | 0.671 | 1.685 (0.730-3.888) | 0.217 | 0.981 |

¹P values for logistic analysis of three alternative models (codominant, dominant and recessive). The P value for haplotype associations were calculated using single nucleotide polymorphisms Analyzer™ 2.0 software; ²False discovery rate Q value.

DISCUSSION

The present study focused on the association of genetic polymorphisms and haplotypes of the *ITGA1* gene with gastric cancer risk. It has been suggested that the integrin $\alpha 1$ subunit could be involved in gastric cancer carcinogenesis. Integrins on gastric epithelial cells have been reported to serve as a portal for the entry of *H. pylori* *cagA*^[11]. Additionally, the integrin $\alpha 1$ subunit is involved in the adhesion and dissemination of gastric cancer cells to the peritoneum^[18], and an *ITGA2* polymorphism has been reported to be associated with an increase in the risk of gastric cancer^[20]. However, to our knowledge, no previous study has examined the association between *ITGA1* polymorphisms and the risk of gastric cancer.

The SNPs *rs1862610*, *rs2432143* and *rs2447867* were significantly associated with an increase in the risk of gastric cancer. After controlling the FDR, only SNP

rs2432143 in the codominant model was statistically significant. In a gene-based association test, the *ITGA1* gene was found to be significantly associated with gastric cancer.

The C-C type of *ITGA1* haplotype block 2, which includes *rs1862610* and *rs2432143* in intron 1 of the *ITGA1* gene, was found to be a significant protective factor and the A-T type to be a risk factor for gastric cancer. This statistical significance was maintained after controlling the FDR. However, the precise molecular mechanism related to these SNPs is not clear. Based on SNP function prediction using computational methods, SNPs *rs1862610* and *rs2432143* are not predicted to be involved in any structural or functional changes in the integrin $\alpha 1$ subunit. However, we cannot rule out the possibility that these SNPs are either associated with the stability of *ITGA1* mRNA, or in linkage disequilibrium with an as yet unknown functional polymorphism affecting the ex-

pression or function of the integrin $\alpha 1$ subunit.

We used public databases of SNPs related to gastric cancer and assessed the potential functions of selected SNPs with SNP function prediction software. Among the 15 selected SNPs, only two were located in exons, and one was non-synonymous. The potential function was not predicted for any of these SNPs, except for *rs2447867*, which was predicted to be an exonic splicing enhancer (ESE). ESEs are clinically significant because synonymous point mutations in ESEs that were previously thought to be silent mutations can lead to exon skipping and the production of a non-functional protein. As loss of integrin $\alpha 1\beta 1$ has been observed in some other malignancies^[27], non-functional integrin $\alpha 1\beta 1$ could be associated with gastric cancer.

The increased expression of integrin molecules by epithelial cells during inflammation of the underlying lamina propria is probably an adaptive response to prevent extensive epithelial cell sloughing caused by inflammatory mediators. Loss of epithelial integrity due to a decrease in the function of integrin results in more severe injury of the epithelium^[21]. At these sites of tissue injury, bone marrow-derived cells are recruited, and these cells can be a potential source of malignancy^[28]. Because chronic infection with *H. pylori* also induces repopulation of the stomach with bone marrow-derived cells, there is a possibility that a non-functional integrin $\alpha 1$ subunit and *H. pylori* infection would have a synergistic effect in increasing the risk of gastric cancer. The major limitation of the present study is that we did not test for the presence of antibodies against *H. pylori* and the *cagA* antigen in the sera of the case and control subjects.

The OR obtained for SNPs *rs1862610*, *rs2432143*, and *rs2447867* were all below 1.6, while the OR for the *ITGA2* C807T polymorphism in relation to gastric cancer in a Chinese population is 1.57^[20]. These relatively small values can be explained by the promiscuity and redundancy of integrins: one integrin can bind several different ligands, and many different integrins can bind to the same ligand^[29]. Therefore, if an integrin is not functioning, other integrins can compensate for at least some of its function.

In conclusion, the *ITGA1* gene SNPs *rs2432143* and *rs2447867* and the *ITGA1* haplotype block that includes SNP *rs2432143* are significantly associated with gastric cancer risk.

COMMENTS

Background

Integrins mediate signaling events that are essential for stable cell adhesion, cell spreading, migration, survival, proliferation and differentiation. Several integrins, including $\alpha 1\beta 1$, bind to extracellular matrix proteins present in the basal membranes of mature vessels. Tumor progression and the metastasis of various cancers are associated with integrins. The *ITGA1* gene, located on chromosome 5q11.2, encodes the integrin $\alpha 1$ subunit, which is involved in the adhesion of gastric cancer cells to the peritoneum. Adhesion of integrin $\alpha 1$ -positive gastric cancer cells to the extracellular matrix is a critical process in peritoneal dissemination. As integrin $\alpha 1\beta 1$ is up-regulated during inflammation in the gastrointestinal tract mucosa, which is the first step in gastric carcinogenesis,

it is possible that the integrin $\alpha 1$ subunit plays an important role in the development of gastric cancer. It has been suggested that the integrin $\alpha 1$ subunit could be involved in gastric cancer carcinogenesis. Integrins on gastric epithelial cells have been reported to serve as a portal for the entry of *Helicobacter pylori* (*H. pylori*) *cagA*. As integrin $\alpha 1\beta 1$ is up-regulated during inflammation in the gastrointestinal tract mucosa, which is the first step in the gastric carcinogenesis, it is possible that the integrin $\alpha 1$ subunit plays an important role in the development of gastric cancer.

Research frontiers

There are few studies addressing the role of integrins in the development of gastric cancer. An association with an increased risk of gastric cancer has only been reported previously for the *ITGA2* C807T polymorphism in a Chinese population. No earlier study has focused on the association of *ITGA1* gene single nucleotide polymorphisms (SNPs) and haplotypes with gastric cancer risk.

Innovations and breakthroughs

To the best of the authors' knowledge, this present study is the first to suggest a significant association of the genetic polymorphisms and haplotypes of *ITGA1* gene with an increased gastric cancer risk.

Applications

Integrins on gastric epithelial cells have been reported to serve as a portal of entry for *H. pylori* *cagA*, and loss of epithelial integrity due to a decrease in the function of integrins results in more severe injury of the epithelium. Studies are needed addressing the interaction of non-functional integrin $\alpha 1$ subunit and *H. pylori* infection in increasing the risk of gastric cancer.

Peer review

This paper is focused on the *ITGA1* polymorphisms and haplotypes, and gastric cancer risk in a Korean population. The results showed the SNPs *rs1862610*, *rs2432143*, and *rs2447867*, and the *ITGA1* haplotype block which includes SNPs *rs1862610* and *rs2432143* were significantly associated with gastric cancer. It is interesting.

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