Association between CCND1 G870A Polymorphism and Digestive System Tumors: Postprint

Authors: Yang Shumin, Shi Yalin

Date: 2017-12-21T00:00:00+00:00

Abstract

Objective To investigate the correlation between CCND1 gene G870A polymorphism and digestive system tumors. Methods We selected 164 patients with digestive system tumors from our hospital between August 2010 and August 2014 as the case group (divided into gastric cancer and colorectal cancer subgroups, 82 cases each), and 82 healthy individuals undergoing physical examination during the same period as the control group. The polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) method was used to analyze the distribution frequency of CCND1 gene G870A genotypes among the three groups, and its relationship with tumor staging and grading was analyzed. Results The distribution frequencies of GG, GA, and AA at the CCND1 gene G870A locus in both the gastric cancer and colorectal cancer groups showed significant differences compared with the control group ($P<0.05$). The polymorphism of the CCND1 gene G870A locus demonstrated a significant association with increased risk of digestive system tumor onset ($P<0.05$). The risk of developing digestive system tumors in individuals with GA and AA genotypes was significantly higher than that in those with the GG genotype ($P<0.05$). Moreover, the genotype distribution rate was higher in patients with well-differentiated tumors and advanced stage than in those with poorly-differentiated tumors and early stage ($P<0.05$). Conclusion CCND1 gene G870A polymorphism is associated with the risk of digestive system tumor onset, and individuals harboring the A genotype are more susceptible to developing digestive system tumors, which is correlated with tumor differentiation and staging.

Full Text

Preamble

J South Med Univ, 2016, 36(11): 1589-1592
doi: 10.3969/j.issn.1673-4254.2016.11.25

YANG Shumin[1], SHI Yalin[2]
[1]Jiaozuo Worker Medical School, Jiaozuo 454000, China;
[2]Zhengzhou Maternity and Child Healthcare Hospital, Zhengzhou 450012, China

Abstract

Objective To investigate the correlation between CCND1 gene G870A polymorphism and digestive system tumors. **Methods** A total of 164 patients with digestive system tumors treated at our hospital from August 2010 to August 2014 were enrolled as the case group (including 82 gastric cancer patients and 82 colorectal cancer patients), and 82 healthy individuals undergoing physical examination during the same period were selected as the control group. Polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) was used to analyze the distribution frequency of CCND1 gene G870A genotypes in the three groups and its relationship with tumor staging and grading. **Results** The distribution frequencies of GG, GA, and AA genotypes at the CCND1 gene G870A locus in both the gastric cancer and colorectal cancer groups differed significantly from those in the control group ($P<0.05$). The G870A polymorphism showed a significant correlation with increased risk of digestive system tumors ($P<0.05$). The GA and AA genotypes were associated with significantly higher risk of digestive system tumors compared with the GG genotype ($P<0.05$). Moreover, the genotype distribution rates were significantly higher in patients with high differentiation and advanced stages than in those with low differentiation and early stages ($P<0.05$). **Conclusion** The CCND1 gene G870A polymorphism is associated with the risk of digestive system tumors, and the A-containing genotype is more susceptible to digestive system tumors and correlates with tumor differentiation and staging.

Key words: CCND1 gene; G870A locus; polymorphism; digestive system tumors; tumor staging

Introduction

Digestive system tumors encompass both upper and lower gastrointestinal malignancies, including esophageal cancer, gastric cancer, colorectal cancer (rectal and colon cancer), and primary liver cancer [1], with etiology involving multiple factors. Current research has focused extensively on the CCND1 gene, which is located on chromosome 11q13, contains 5 exons and 4 introns spanning approximately 15 kb, and encodes cyclin D1 [2]. A G870A polymorphism exists at the last base of exon 4 in the CCND1 gene, and variations at this locus have been associated with multiple tumor types, including liver, breast, lung, and oral cancers. While numerous studies have investigated the relationship between CCND1 gene G870A polymorphism and various cancers, research

specifically examining its association with digestive system tumors remains limited. Some studies suggest a link between CCND1 G870A polymorphism and digestive system tumorigenesis, but results differ among various gastrointestinal cancers. This study addresses these gaps by investigating the correlation between CCND1 gene G870A polymorphism and the development of esophageal cancer, gastric cancer, rectal cancer, and colon cancer to determine the relationship between this polymorphism and each specific digestive tumor type.

Materials and Methods

Study Population

We enrolled 164 patients with digestive system tumors treated at our hospital from August 2010 to August 2014 as the case group, comprising 82 gastric cancer patients and 82 colorectal cancer patients. Eighty-two healthy individuals undergoing physical examination during the same period served as the control group. All patients were pathologically confirmed as having gastric or colorectal cancer and provided informed consent. Patients with severe brain, liver, kidney dysfunction, or hematological disorders were excluded. The three groups showed no significant differences in gender, age, or other demographic characteristics ($P>0.05$,), ensuring comparability.

DNA Extraction and Genotyping

Reagents and Equipment: TaqDNA polymerase reagent, dNTPs, proteinase K, electrophoresis agarose, DNA marker, PCR amplifier, UV spectrophotometer, electrophoresis apparatus, and gel imaging analysis system.

DNA Extraction: Two milliliters of EDTA-Na2 anticoagulated blood were collected from both control and case group subjects. Genomic DNA was extracted from peripheral blood using the conventional phenol-chloroform method, and DNA concentration and purity were measured and stored at low temperature.

Genotyping of CCND1 Gene G870A Polymorphism: The primers for CCND1 gene G870A polymorphism genotyping were: forward primer 5' -AGTTCATTTCCAATCCGCCC-3'and reverse primer 5'-TTCCGTGGCACTAGGTGTC-3' . The PCR reaction system [5] contained 10× Buffer 2.0 L, MgCl 2.0 L, dNTPs, 0.4 L each of forward and reverse primers, 0.4 L TaqDNA polymerase, 0.25 L DNA template, and double-distilled water to a final volume of 20 L. PCR conditions [6] included initial denaturation at 95°C for 5 min, followed by 35 cycles of denaturation at 95°C for 45 s, annealing at 52°C for 45 s, and extension at 72°C for 1.5 min, with a final extension at 72°C for 15 min. PCR products were detected by 1.5% agarose gel electrophoresis. The PCR products were digested with restriction endonuclease at 37°C for 16 h, and the digested products were analyzed by 1.5% agarose gel electrophoresis to obtain the DBCCR1 polymorphism genotypes.

Statistical Analysis

Data analysis was performed using SPSS 19.0 statistical software. Data conforming to normal distribution were analyzed using the gene counting method to calculate genotype and allele distribution frequencies in case and control groups. Allele frequency was calculated as (2 × number of homozygotes + number of heterozygotes) / (2 × total number of subjects). Count data were expressed as cases and percentages and analyzed using the ^{2} test. $P<0.05$ was considered statistically significant.

Results

Genotype Distribution

The distribution frequencies of GG, GA, and AA genotypes at the CCND1 gene G870A locus in both gastric cancer and colorectal cancer groups differed significantly from those in the control group ($P<0.05$,). As shown in and , the GA and AA genotypes were associated with significantly higher risk of digestive system tumors compared with the GG genotype ($P<0.05$). Furthermore, genotype distribution rates were higher in patients with high differentiation and advanced stages than in those with low differentiation and early stages ($P<0.05$).

Association with Tumor Risk

As shown in , the CCND1 gene G870A polymorphism demonstrated a significant correlation with increased risk of digestive system tumors ($P<0.05$).

Discussion

Tumors are considered cell cycle diseases, and dysregulation of the cell growth cycle plays a critical role in tumorigenesis and development [7]. Cyclin D1 is essential for normal cell cycle regulation, and the gene encoding it, CCND1, has a G870A polymorphism associated with various tumors [8]. Studies comparing the correlation between CCND1 G870A polymorphism and cervical cancer, oral cancer, and other diseases have found close relationships between disease onset and genotype distribution. Research [9] has reported that CCND1 G870A polymorphism is associated with digestive system tumors, with differences between upper and lower gastrointestinal tumors.

Lower digestive tract tumors primarily include colon and rectal cancers, which often occur simultaneously. Numerous studies have investigated the relationship between CCND1 G870A polymorphism and colon cancer risk. Reports [10-12] indicate that CCND1 G870A polymorphism increases rectal cancer risk, with the A allele specifically increasing colorectal cancer risk in male patients. Individuals carrying the A allele show increased incidence of colon cancer. Other studies [13] have reported that individuals with a family history of colorectal cancer and AA or AG genotypes have increased colorectal cancer risk, while GG

genotype promotes metastasis. However, some studies show contradictory results. Our findings are consistent with these studies, though we did not analyze sex differences, focusing instead on the relationship between CCND1 G870A polymorphism and colon cancer risk, which similarly demonstrated increased incidence in patients carrying the A allele.

Upper digestive tract tumors mainly include esophageal and gastric cancers. Studies on CCND1 G870A polymorphism and upper gastrointestinal tumors show close relationships between tumor development and this polymorphism. Research [14] on gastric cancer precancerous lesions demonstrated that the AA genotype significantly increases risk of intestinal metaplasia, with A allele carriers showing greater tumor susceptibility. Our study similarly revealed that the A allele or AA genotype of CCND1 G870A confers significantly higher gastric cancer risk than the GG genotype.

This study analyzed the relationship between CCND1 G870A polymorphism and common digestive tumors (gastric and colon cancers) to clarify its association with digestive system tumor risk. We found significant differences in AA, AG, and GG genotype distribution frequencies between gastric/colon cancer patients and healthy individuals. Analysis of the relationship between genotype distribution and tumor differentiation and staging revealed that more severe differentiation and higher stages were associated with increased AA+AG distribution frequencies. Correlation analysis demonstrated that A-containing genotypes confer higher disease susceptibility.

Conclusion

In summary, CCND1 gene G870A polymorphism is associated with digestive system tumor risk, with A-containing genotypes showing increased susceptibility to digestive system tumors and correlation with tumor differentiation and staging.

References

[1] Xu X, Yang X, Ru G, et al. miR-146a gene polymorphism rs2910164 and the risk of digestive tumors: a meta-analysis of 21 case-control studies[J]. Oncol Rep, 2014, 31(1): 472-9.
[2] Wang S, Wang Z. Prognostic value of long noncoding RNA HOTAIR in digestive system malignancies[J]. J Gastroenterol Hepatol, 2015, 30(7): 1123-33.
[3] Chen B, Cao L, Yang P, et al. Cyclin D1 (CCND1) G870A gene polymorphism is an ethnicity-dependent risk factor for digestive tract cancers: a meta-analysis comprising 20, 271 subjects[J]. Cancer Epidemiol, 2012, 36(2): 106-15.
[4] Hu Z, Zhou Z, Xiong G, et al. Cyclin D1 G870A polymorphism and the risk of hepatocellular carcinoma in a Chinese population[J]. Tumour Biol, 2014, 35(6): 5607-12.
[5] Chen X, Zhao T, Li L, et al. CCND1 G870A polymorphism with altered cyclin D1 transcripts expression is associated with the risk of glioma in a Chinese

population[J]. DNA Cell Biol, 2012, 31(6):
[6] Cander S, Erturk E, Karkucak M, et al. Effect of cycline D1 (CCND1) gene polymorphism on tumor formation and behavior in patients with prolactinoma[J]. Gene, 2012, 509(1): 158-63.
[7] Yang J, Zhang G, Chen J. CCND1 G870A polymorphism is associated with increased risk of colorectal cancer, especially for sporadic colorectal cancer and in Caucasians: a meta-analysis[J]. Clin Res Hepatol Gastroenterol, 2012, 36(2): 169-77.
[8] Xue Y, Rong L, Tong N, et al. CCND1 G870A polymorphism is associated with toxicity of methotrexate in childhood acute lymphoblastic leukemia[J]. Int J Clin Exp Pathol, 2015, 8(9):
[9] Zeng Z, Tu J, Cheng J, et al. Influence of CCND1 G870A polymorphism on the risk of HBV-related HCC and cyclin D1 splicing variant expression in Chinese population[J]. Tumour Biol, 2015, 36(9): 6891-900.
[10] Wang S, Huang Y, Su R, et al. Cyclin D1 G870A polymorphism is associated with an increased risk of multiple myeloma[J]. Genet Mol Res, 2015, 14(2): 5856-61.
[11] Ma G, Wang Q, Lv C, et al. The prognostic significance of HOTAIR for predicting clinical outcome in patients with digestive system tumors[J]. J Cancer Res Clin Oncol, 2015, 141(12): 2139-45.
[12] Atac A, Riecke B, Assaf AT, et al. Lack of evidence for predictive and prognostic value of cyclin D1 gene polymorphism CCND1 G870A for oral squamous cell carcinoma[J]. Anticancer Res, 2014, 34(1): 235-8.
[13] Delektorskaia VV, Kushliskiĭ NE. Neuroendocrine tumors of digestive system: morphologic spectrum and cell proliferation (Ki67 index)[J]. Vestn Ross Akad Med Nauk, 2013, (5): 28-37.
[14] Roesch-Dietlen F, Jiménez-García VA, Remes-Troche JM, et al. Epidemiologic behavior of malignant digestive tract tumors over a five year period in Veracruz, Mexico[J]. Rev Gastroenterol Mex, 2012, 77(1): 3-8.

(Edited by: Sun Changpeng)

Note: Figure translations are in progress. See original paper for figures.