RESEARCH ARTICLE

The miR-146a rs2910164 G > C Polymorphism and Susceptibility to Digestive Cancer in Chinese

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Abstract

Background: Several studies have reported the role of the miR-146a rs2910164 G > C polymorphism as a susceptibility factor for several digestive cancers. However, the results have been controversial. Therefore, we conducted the present meta-analysis to obtain the most reliable estimate of the association. Methods: PubMed, Embase and Web of Science databases were searched. Crude odds ratios (ORs) with 95% confidence intervals (CIs) were extracted and pooled to assess the strength of the association between miR-146a rs2910164 G > C polymorphism and digestive cancer risk. A total of four eligible studies including 3,447 cases and 5,041 controls based on the search criteria were included. Results: We observed that miR-146a rs2910164 G > C polymorphism was not significantly correlated with digestive cancer risks when all studies were pooled into the meta-analysis. While we found that miR-146a rs2910164 polymorphism was not associated with gastric cancer, it was significantly linked with hepatocellular cancer risk (the homozygote codominant model: OR = 1.40, 95% CI = 1.04-1.87). In the stratified analysis by ethnicity, significant associations were observed in Chinese population for the allele contrast model (OR = 1.25; 95% CI = 1.12-1.38), for the homozygote codominant model (OR = 1.62; 95% CI = 1.28-2.04), and for the recessive model (OR = 1.38; 95% CI = 1.16-1.64). However, studies with Asian groups presented no significant association for all genetic models. Conclusions: This meta-analysis suggests that the miR-146a rs2910164 G > C polymorphism is a low-penetrant risk factor for digestive cancers in Chinese.

Keywords: MicroRNAs - genetic polymorphisms - mutation - risk - meta-analysis - Chinese

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Introduction

It is well known that we divided RNA into protein-coding and non-coding RNA. Over the past decade, the field of RNA research has rapidly expanded. Many studies found that mRNA accounted for only 2% of all transcripts, and the remaining 98% are non-coding RNAs in the human genome (Lander et al., 2001; Esteller et al., 2011), suggesting that the noncoding portion of the genome is of crucial importance in the development of normal tissue development and disease (Wright et al., 2011). One of the members of these RNAs, microRNAs, an endogenous, small non-coding and 18 – 25 nucleotides RNAs, have a focus on this stage of development. They are encoded in the genome and are generally transcribed by RNA polymerase II, and exert their effects by associating with a group of proteins termed the ‘RNA-induced silencing complex’ (RISC). RISC is directed to target mRNAs via imperfect sequence complementarity between the mature miRNA and 3′-untranslated region (3′-UTR) of target mRNAs. MiRNAs have been shown to play crucial roles in diverse biological processes, such as cell apoptosis, differentiation, development, signal transduction (Bartel et al., 2004; Denli et al., 2004). Watson–Crick complementarity between the target and the seed region (2–8 nucleotides) of the mature miRNA is both necessary and sufficient for targeting and regulating of mRNAs by miRNAs. But the seed region of miRNAs is so short, so its polymorphism may affect the combination of the core area of binding the 3′ UTR of target genes, thus affecting its regulation of target genes (Hu et al., 2008).

A Homo sapiens miR-146a gene located on chromosome, it has been reported that it play a vital role in several human cancers. Recently, single nucleotide polymorphism (SNP, rs2910164) has been identified in the miR-146a gene. More recently, several studies have assessed the relationship between the polymorphism of miR-146a G > C and the risks to digestive cancers, however, the results have been controversial (Xu et al., 2008; Guo et al., 2010; Okubo et al., 2010; Srivastava et al., 2010; Zeng et al., 2010; Akkız et al., 2011; Hishida et al., 2011; Min et al., 2011; Zhang et al., 2011; Zhou et al., 2011). To derive a more precise effect on the association between miR-146a polymorphism and digestive cancers risks, therefore, we conducted this meta-analysis.

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The miR-146a rs2910164 G > C Polymorphism and Susceptibility to Digestive Cancer in Chinese
Materials and Methods

Identification and eligibility of relevant studies

Search terms “microRNA-146a”, “digestive cancer”, “genotype”, “polymorphism” and “variant” were employed to explore publications in PubMed, ISI Web of Knowledge and Embase databases for relevant reports (last search update February 2012). We did not define any minimum number of patients to be included for meta-analysis. When multiple studies of the same patient population were identified, we included the published report with the largest sample size.

Inclusion and exclusion criteria

The following inclusion criteria were used to select literatures for this analysis: (a) evaluation of miR-146a rs2910164 G > C polymorphism and several digestive cancers, (b) only the case-control studies were considered, (c) sufficient published data for estimating an odds ratio (OR) with 95% confidence interval (CI). Major exclusion criteria were: (1) no control population, (2) no available genotype frequency, and (3) duplicated studies.

Data extraction

For each study, the following data were collected: first author’s surname, year of publication, country of origin, ethnicity, criteria of enrolled patients, genotyping method, total numbers of cases and controls as well as numbers of cases and controls with GG, GC and CC genotypes. The strength of the association between miR-146a rs2910164 G > C polymorphism and digestive cancers risks were estimated using OR, with the corresponding 95% CI. Disagreement was resolved by discussion until a consensus was reached between the two authors. We did not define any minimum number of patients for inclusion in our meta-analysis.

Statistical methods

The risk of UC associated with miR-146a rs2910164 G > C was estimated for each study by OR with 95% CI. For five studies, we analysed the relationship for the allele contrast model (G vs C). At the same time, due to lack the specific genotypes of Glas’s literature, so we estimated the association under other four different types of ORs, namely the homozygote codominant model (GG vs CC), the heterozygote codominant model (GC vs CC), the dominant model (GG+GC vs CC) and the recessive model (GG vs GC+CC). Hardy–Weinberg equilibrium (HWE) was tested by the Chi-square test. The Q-statistic was used to investigate the degree of heterogeneity between the trials, and a P-value 0.10 for the Q-test indicated a lack of heterogeneity among studies. Genotype distribution of controls in all studies was consistent with HWE, except for Zhang XW’s study on HCC. We used the fixed-effects model and the random-effects model based on the Mantel–Haenszel method (Jose et al., 2008) and the DerSimonian and Laird method (Kjellson et al., 2008), respectively, to combine values from each of the studies. A sensitivity analysis was also performed by omitting each study in turn to identify potential outliers. All statistical analyses were performed with Review Manage version 12.0 using two-sided P-values.

Results

Study characteristics

We obtained 10 studies about the association between miR-146a rs2910164 polymorphism. Following the above inclusion and exclusion criteria, 9 publications were included in the final meta-analysis (Xu et al., 2008; Guo et al., 2010; Okubo et al., 2010; Zeng et al., 2010; Akkiz et al., 2011; Hishida et al., 2011; Min et al., 2011; Zhang et al., 2011; Zhou et al., 2011). Characteristics of studies focusing on miR-146a rs2910164 G > C are summarized in Table 1. Genotypes and separate P values for miR-146a rs2910164 polymorphism are list in Table 2. As shown in Figure 1, the literatures do not exist in significant heterogeneity.

Table 1. Characteristics of Studies Included in the Meta-analysis

| First author/ Published year | Ethnicity | Method | Cancer Cases Controls | HWE of controls |
|-----------------------------|----------|--------|-----------------------|-----------------|
| Xu, 2008                    | Chinese  | PCR-RFLP | HCC                   | 479 504 0.12    |
| Akkiz, 2011                 | Turkish  | PCR-RFLP | HCC                   | 222 222 0.38    |
| Zhang, 2011                 | Chinese  | PCR-PRA | HCC                   | 925 1593 0.02   |
| Zhou, 2011                  | Chinese  | PCR-RFLP | HCC                   | 186 483 0.06    |
| Hishida, 2010               | Japanese | PCR-CTTP | GC                    | 584 1637 0.74   |
| Okubo, 2010                 | Japanese | PCR-RFLP | GC                    | 552 697 0.28    |
| Zeng, 2010                  | Chinese  | PCR-RFLP | GC                    | 304 304 0.12    |
| Guo, 2010                   | Chinese  | PCR-Snapshot | ESCC                  | 444 468 0.12  |
| Min, 2011                   | Korean   | PCR-RFLP | CRC                   | 446 502 0.06    |
| Srivastava, 2010            | Indian   | PCR-RFLP | GBC                   | 230 224 0.08    |

HCC, hepatocellular cancer; GC, gastric cancer; ESCC, esophageal squamous cell carcinoma; CRC, colorectal cancer; GBC, gallbladder cancer

Table 2. Genotypes and P-values of for miR-146a rs2910164 Polymorphisms Included in the Meta-analysis

| First author | Cases | Controls |
|--------------|-------|----------|
| Xu, 2008     | GG    | G        |
|              | GC    | C        |
|              | CC    | G        |
|              |      | C        |
| Akkiz, 2011  | GG    | G        |
|              | GC    | C        |
|              | CC    | G        |
|              |      | C        |
| Zhang, 2011  | GG    | G        |
|              | GC    | C        |
|              | CC    | G        |
|              |      | C        |
| Zhou, 2011   | GG    | G        |
|              | GC    | C        |
|              | CC    | G        |
|              |      | C        |
| Hishida, 2010| GG    | G        |
|              | GC    | C        |
|              | CC    | G        |
|              |      | C        |
| Okubo, 2010  | GG    | G        |
|              | GC    | C        |
|              | CC    | G        |
|              |      | C        |
| Zeng, 2010   | GG    | G        |
|              | GC    | C        |
|              | CC    | G        |
|              |      | C        |
| Guo, 2010    | GG    | G        |
|              | GC    | C        |
|              | CC    | G        |
|              |      | C        |
| Min, 2011    | GG    | G        |
|              | GC    | C        |
|              | CC    | G        |
|              |      | C        |
| Srivastava, 2010| GG    | G        |
|              | GC    | C        |
|              | CC    | G        |
|              |      | C        |

Meta-analysis fixed-effects estimates (exponential form)

Study omitted

Figure 1. Sensitivity Analysis for All Studies
Table 3. Results of Meta-analysis for miR-146a rs2910164 Polymorphisms and Digestive System Cancer Risks

| Study groups | Variables | Allele contrast model | Homozygote codominant |
|--------------|-----------|-----------------------|-----------------------|
|              | OR (95% CI) | \( P_h \) | OR (95% CI) | \( P_h \) |
| Total        | 1.05 (0.92-1.20) | 0 | 1.15 (0.86-1.53) | 0.001 |
| Two major digestive cancer types |          |                |                       |             |
| HCC          | 1.08 (0.89-1.32) | 0.128 | 1.40 (1.04-1.87) | 0.308 |
| GC           | 1.00 (0.78-1.27) | 0.003 | 0.98 (0.63-1.53) | 0.009 |
| Ethnicity    |          |                |                       |             |
| Asian        | 1.06 (0.92-1.22) | 0 | 1.15 (0.85-1.57) | 0 |
| Chinese      | 1.25 (1.12-1.38) | 0.222 | 1.62 (1.28-2.04) | 0.237 |

\( P_h \), \textit{P}-value of overall effect test; *Statistically significant results

Figure 2. Forest Plot for the Association Between miR-146a rs2910164 G > C and Digestive Cancers Risks in Chinese Population Studies. A. for the allele contrast model; B. for the homozygote codominant model; C. for the recessive model. The study is shown by the point estimate of the OR (the size of the square is proportional to the weight of each study) and 95% CI for OR; pooled OR and 95% CI have been appropriately derived from the fixed effects model.

Figure 3. Funnel Plot Analysis for Odds Ratios of G Allele Compared with C Allele in Overall Meta-analysis results

Table 3 lists the main results of pooled ORs for miR-146a rs2910164 G > C polymorphism and several digestive cancers risks. The results showed that no statistically significant difference were found in the allele and other genotype models of miR-146a rs2910164 G > C polymorphism among digestive cancers including gastric cancer and controls when all studies were pooled into the meta-analysis. Tumor location was taken into consideration for subgroup analysis, we observed that miR-146a rs2910164 polymorphism was not associated with gastric cancer, but it was significantly correlated with hepatocellular cancer risk (the homozygote codominant model: \( \text{OR} = 1.40, \text{95\% CI} = 1.04 – 1.87, P = 0.308 \) for heterogeneity, Figure 1). In the stratified analysis by ethnicity, significant associations were observed in Chinese population for the allele contrast model (\( \text{OR} = 1.25; \text{95\% CI} = 1.12 – 1.38; P = 0.222 \) for heterogeneity, Figure 2 A), for the homozygote codominant model (\( \text{OR} = 1.62; \text{95\% CI} = 1.28 – 2.04; P = 0.237 \) for heterogeneity, Figure 2 B), and for the recessive model (\( \text{OR} = 1.38; \text{95\% CI} = 1.16 – 1.64; P = 0.811 \) for heterogeneity, Figure...
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Identification of susceptibility genes for several diseases, genetic variants that appear to contribute to human disease disorders on a global scale. It has identified scores of matched controls.

and using standardized unbiased genotyping methods, considering gene-gene and gene-environment interactions, of digestive cancers in Chinese population. Of course, G > C polymorphism was associated with increased risk there was an evidence to indicate that miR-146a rs2910164 correlated with hepatocellular cancer risk. Simultaneously not associated with gastric cancer, but it was significantly the meta-analysis. Interestingly, we observed that was of miR-146a rs2910164 G > C polymorphism in the researchers have taken into account age and other factors, and abnormal expression can effect on carcinogenesis and tumor progression. Some SNPs in pre-microRNAs, flanking regions or target sites have been demonstrated to affect certain physiological processes or related with diseases (Gong et al., 2012). Sometimes, single point mutations in the 7mer seed sites of miRNAs may reduce effectiveness or abolish miR mediated repression may induce effectiveness or abolish miR mediated repression (Richardson et al., 2011).

Some researchers have examined the association of miR-146a rs2910164 G > C polymorphism with several cancers risks, and a significant relationship was observed in several but not all studies (Xu et al., 2008; Guo et al., 2010; Okubo et al., 2010; Srivastava et al., 2010; Zeng et al., 2010; Akkız et al., 2011; Hishida et al., 2011; Min et al., 2011; Zhang et al., 2011; Zhou et al., 2011). Such as, some studies have divided cases and controls into HBV infection according to HBV DNA amount, and some researchers have taken into account age and other factors, so each obtained the different results.

The aim of our study was to demonstrate the role of miR-146a rs2910164 G > C polymorphism in the relationship with digestive system cancers risks using the meta-analysis. Interestingly, we observed that was not associated with gastric cancer, but it was significantly correlated with hepatocellular cancer risk. Simultaneously there was an evidence to indicate that miR-146a rs2910164 G > C polymorphism was associated with increased risk of digestive cancers in Chinese population. Of course, we must be noted that our study population was small, so it is still necessary to conduct larger sample studies considering gene-gene and gene-environment interactions, and using standardized unbiased genotyping methods, homogeneous gastric cancer patients, and sufficiently matched controls.

Recently, Genome-wide association studies (GWAS) help scientists understand the inheritance patterns of disorders on a global scale. It has identified scores of genetic variants that appear to contribute to human disease risk. Especially, it can lead to an enormous boost in the identification of susceptibility genes for several diseases, but there are some insurmountable problems, the most critical is that some high-risk sites for low-frequency masked by low-risk sites for high-frequency, so this may result in the analysis is not comprehensive, and lose a lot of meaningful SNP sites.

In summary, our study shows a genetic association between miR-146a rs2910164 G > C variant increases susceptibility to digestive cancers risk in Chinese population, whereas we need to be further evaluated in larger sample collections.

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