Association of miR-21 with esophageal cancer prognosis: a meta-analysis

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ABSTRACT. The present study aimed to explore the relationship between miRNA expression and survival in patients with esophageal cancer (EC) using meta-analysis. We searched PubMed, EMBASE, CNKI, Wanfang, and ISI Web of Science databases without time restrictions, and extracted relevant data, such as the name of first author, publication year, age, gender, number of case, etc. from the studies included. We calculated the pooled hazard ratios (HRs) using the RevMan 5.2 software. A total of five studies involving 504 subjects were included in the meta-analysis, with the purpose of analyzing the association of miRNA-21 expression with EC prognosis. The pooled HR of elevated versus decreased miR-21 expression in EC was 1.87 [95% confidence interval (CI): 1.37–2.55, $P < 0.001$], with elevated miR-21 expression being associated with poorer prognosis for patients with EC. Our results support a prognostic role for miR-21 in EC.

Key words: miR-21; Esophageal cancer; Prognosis; Meta-analysis

INTRODUCTION

Esophageal cancer (EC) is a multifactor disease associated with varied risk factors (Yano et al., 2014). In humans worldwide, EC is considered the sixth leading cause of cancer deaths (Enzinger and Mayer, 2003). A previous study has shown that there were an estimated 482,300 new cases of EC in 2008 worldwide (Shridhar et al., 2013). The survival rate of EC ranges from 10 (Sant et al., 2003) to 20-30% (Kirby and Rice, 1994) in different populations.

MicroRNAs (miRNAs) are a class of small noncoding RNAs that are approximately 23 nucleotides in length (Huang et al., 2014). miRNAs are involved in cell development, differentiation, proliferation, and apoptosis through targeting mRNAs (Bao et al., 2014). A previous study (Calin et al., 2004) suggested that over 50% of miRNA genes are located in fragile sites and cancer associated genomic regions. This result suggested that miRNAs might play a vital role in the pathogenesis of human cancers. Accumulated evidence has indicated that aberrant expression of miRNAs is associated with the prognosis of EC (Hamano et al. 2011; Komatsu et al. 2013). We performed a meta-analysis of the data available from articles published in this field to evaluate the roles of specific microRNAs as prognostic biomarkers in EC.

MATERIAL AND METHODS

Literature search

We searched the PubMed, EMBASE, China Knowledge Resource Integrated (CNKI), Wanfang, and ISI Web of Science databases using the search terms "miRNA", or "microRNA" and "esophageal cancer", or "esophageal neoplasm", or "esophageal carcinoma", or "ESCC" and "prognosis" and "survival" without the publication year or language published. In addition, reference lists from the retrieved articles and from review articles were reviewed manually to identify the missing relevant publications.

Eligibility criteria

Two reviewers independently assessed the eligibility of the retrieved articles. All publications were required to meet the following criteria: 1) reported survival data in patients with EC; 2) provided sufficient data for determining an estimate of hazard ratios (HRs) and 95% confidence intervals (CIs); and 3) enrolled more than 20 patients. Articles were excluded if relevant data were not available, and studies in which participants did not have documented symptoms or diagnostic evidence of EC were also excluded. To test for between-study heterogeneity, we used the Q- and I^2 tests within the RevMan 5.2 software package provided by the Cochrane Collaboration (Oxford, UK; www.cochrane.org).

Data extraction

Two reviewers independently extracted the data from the included studies. We extracted the following data: name of first author, year of publication, country, number of participants, tumor stage, HR with 95%CI, and outcomes.

Statistical analyses

Meta-analysis was performed using the RevMan 5.2 software. We used the Q- and I^2 tests to examine the heterogeneity between each study. We utilized a fixed effects model to summarize pooled HRs and 95%CIs using the generic inverse variance method. Publication bias was weighted by a Begg's funnel plot with the Egger's bias indicator test with $P < 0.05$ being considered statistically significant (Song and Gilbody, 1998).
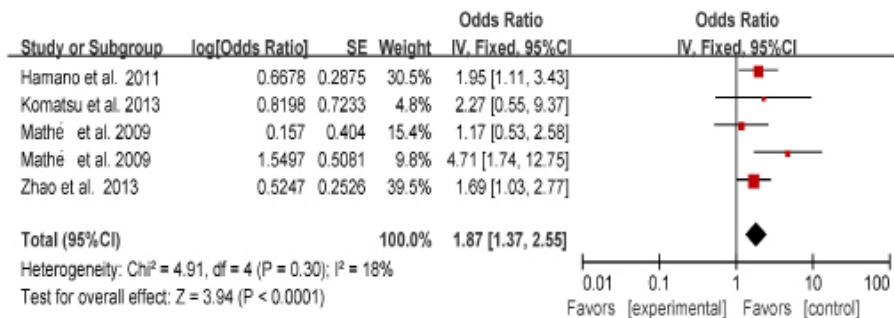
RESULTS

We identified 256 studies of miRNAs associated with the prognosis of EC from a primary literature search in PubMed, EMBASE, CNKI, Wanfang, and ISI Web of Science databases. In these studies, 7 miRNAs were involved including miR-21, miR-16, miR-31, etc. After screening the titles and abstracts, 236 records were excluded because they involved duplicate publication, review articles, or were irrelevant to the current study. Seven articles were further reviewed in detail. Three of these were further excluded because data were unavailable for further analysis. A total of four articles including five independent studies were eventually included. The clinicopathological characteristics of the eligible studies are summarized in Table 1.

For studies evaluating overall survival in EC, no significant heterogeneity was found ($I^2 = 18\%$, $P = 0.30$), therefore, a fixed effects model was applied to calculate the pooled HR (pooled HR = 1.87, 95%CI = 1.37-2.55, $P < 0.001$; Figure 1).

Table 1. Characteristics of studies included.

First author	Publication year	Methods	Population	N	Stage	EC	Cutoff	Outcomes
Hamano et al.	2011	qRT-PCR	Japanese	98	I-IV	U48	Median	OS
Komatsu	2013	qRT-PCR	Japanese	50	I-IV	U6	Normal	OS
Mathé et al.	2009	qRT-PCR	Maryland (USA)	24	0-IV	U66	Median	OS
Mathé et al.	2009	qRT-PCR	Japanese	33	0-IV	U66	Median	OS
Zhao et al.	2013	qRT-PCR	Chinese	178	I-IV	U6	Median	OS

qRT-PCR = quantitative Real-time Polymerase Chain Reaction; EC = esophageal cancer; OS = overall survival.

Figure 1. Forest plot of EC and miR-21. The horizontal lines correspond to the study-specific OR and 95%CI, respectively. The areas of the squares reflect the study-specific weights. The diamond represents the pooled results of HR and 95%CI. EC = esophageal cancer; OR = odds ratio; CI = confidence interval; HR = hazard ratio; SE = standard error.

The potential for publication bias within the studies included was evaluated by Begg's plots and the Egger's test. No significant publication biases were found from the results of the Begg's plots (Figure. 2); the Egger's test reached similar conclusions.
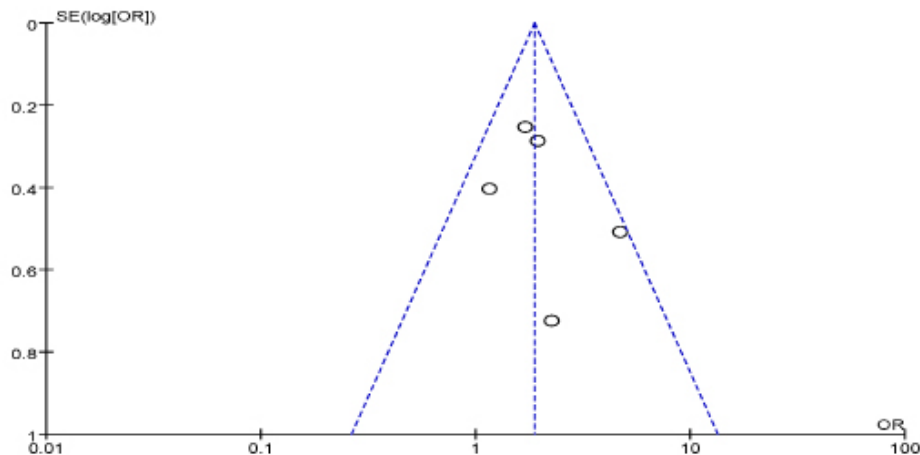
Figure 2. Funnel plot to test for publication bias. Each point represents a separate study for the indicated association. Log OR represents the natural logarithm of the OR. The vertical line represents the mean effects size. OR = odds ratio; SE = standard error.

DISCUSSION

In this meta-analysis, we found that miR-21 was associated with EC using the pooled results from five studies published. miR-21 is an oncogenic miRNA that has been shown to be produced at increased levels in EC, and overexpression of miR-21 plays important roles in increasing cell proliferation, migration, invasion, and survival (Li et al., 2013; Wang et al., 2013). Previous studies (Mathé et al. 2009; Komatsu et al., 2013; Wang et al., 2013; Zhao et al., 2013) have suggested that miR-21 downregulation inhibits cell growth and invasion and induces cells to apoptose through targeting of the *FASL*, *TIMP3*, and *RECK* genes. A study by Hamano et al. (2011) also indicated that miR-21 regulates stem cell function, which is involved in the resistance to chemotherapy in EC. In addition, Ma et al. (2011) have also demonstrated that miR-21 was overexpressed *in vitro* as well as in patients with EC, and promoted cell proliferation by targeting PTEN at posttranscriptional level.

In our study, we found that elevated miR-21 expression predicted poor overall survival in patients with EC (pooled HR = 1.87), as was predicted from the above results. However, the conclusions from our study should be tempered because the pooled risks of miR-21 for survival, although statistically significant, were not strong, with global HRs of 1.87.

Several additional limitations of our study must also be mentioned. First, although miR-21 has been widely investigated, only five studies were eligible for pooled analysis, which might affect the results of the present study. Second, whereas ideally a clear threshold should be set for the cutoff value of miRNA levels for specific outcomes in EC, most investigators used the median or mean value in their studies as the cutoff value and the accurate values were different.

In conclusion, our results support a prognostic role for miR-21 in EC. Further large studies taking into account the effects of gender, ethnicity, and age are warranted to obtain a more robust assessment of the association between miR-21 and the prognosis of EC.

Conflicts of interest

The authors declare no conflict of interest.

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