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ORIGINAL ARTICLE

Link between miR-19b and the mTOR signaling pathway in cancer prognosis*

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Abstract

Objective Previous studies have reported differing conclusions regarding the prognostic value of miR-19b in cancers. Moreover, miR-19b may affect tumor growth by different pathways, mainly targeting PTEN-PI3K-AKT, which activates the downstream mTOR pathway. Therefore, we performed data mining to explore the possible correlation between miR-19b and mTOR in cancer prognosis.

Methods We conducted online search and collected a total of 943 articles. According to different authors cross check and our study including/excluding criteria we at end retained 21 articles with 25 studies in this meta-analysis. Then TCGA data containing miR-19b level with cancer progression were obtained using OncomiR. Furthermore, Trial Sequential Analysis (TSA) was performed to determine whether the results of our meta-analysis could be used in clinical applications. After that, articles regarding the mechanism of miR-19b in various cancers were analyzed and KEGG pathway database was used to find the main regulatory function of miR-19b in human cancers.

Results Overall hazard ratio (HR) results showed that higher levels of miR-19b expression were correlated with shorter overall survival time [HR = 1.54, 95% confidence interval (CI) = 1.20-1.98] by promoting distant metastasis, but had no correlation with disease-free survival (DFS)/progression-free survival (PFS; HR = 0.61, 95% CI = 0.31–1.19). Data from The Cancer Genome Atlas also revealed the role of miR-19b in tumorigenesis. According to trial sequential analysis results, more evidence is required to confirm that miR-19b is not correlated with DFS/PFS. Exploration of the mechanism revealed a possible link between miR-19b and the mTOR pathway.

Conclusion miR-19b may have a pro-carcinogenic role through the mTOR pathway and thus, it is likely to be a therapeutic target for cancers.

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Key words: microRNA; *miR-19b*; prognosis; mechanism; mTOR; cancers

MicroRNAs (miRNAs) are a series of small endogenous single-stranded non-protein-coding RNA molecules with a length of 19–21 nucleotides [1]. They can regulate the expression of their target genes by binding to the 3'-untranslated region and affecting their translation or degradation [2–4]. The dysregulation of these genes plays significant roles in some pathways related to cancer processes, such as the cell cycle, adhesion, and motility [5]. Meanwhile, it is estimated that 60% of human genes are under the regulation of miRNAs [6], indicating that miRNAs might have certain roles in cancer progression [7–9]. Many recent studies have reported that miRNAs can be classified as either oncomiRs or tumor-suppressive miRNAs [3, 10–11]. Their abnormal levels have been associated with different aspects of cancer, including prognosis and clinicopathological features [12].

miR-19b is located on chromosome 13q31.3 and is recognized as the principal element of the *miR-17-92* cluster, which contains *miR-17*, *miR-18a*, *miR-19a*, *miR-19b*, *miR-20a*, and *miR-92* [13–15]. Recently, increasing evidence has demonstrated that *miR-19b* may be a prognostic biomarker in various human cancers, due to its close relationship with cancer prognosis [16–19]. Moreover, many cancer types, either with high or low mortality rates, have been found to be affected by *miR-19b*. These include astrocytic gliomas [20], nasopharyngeal

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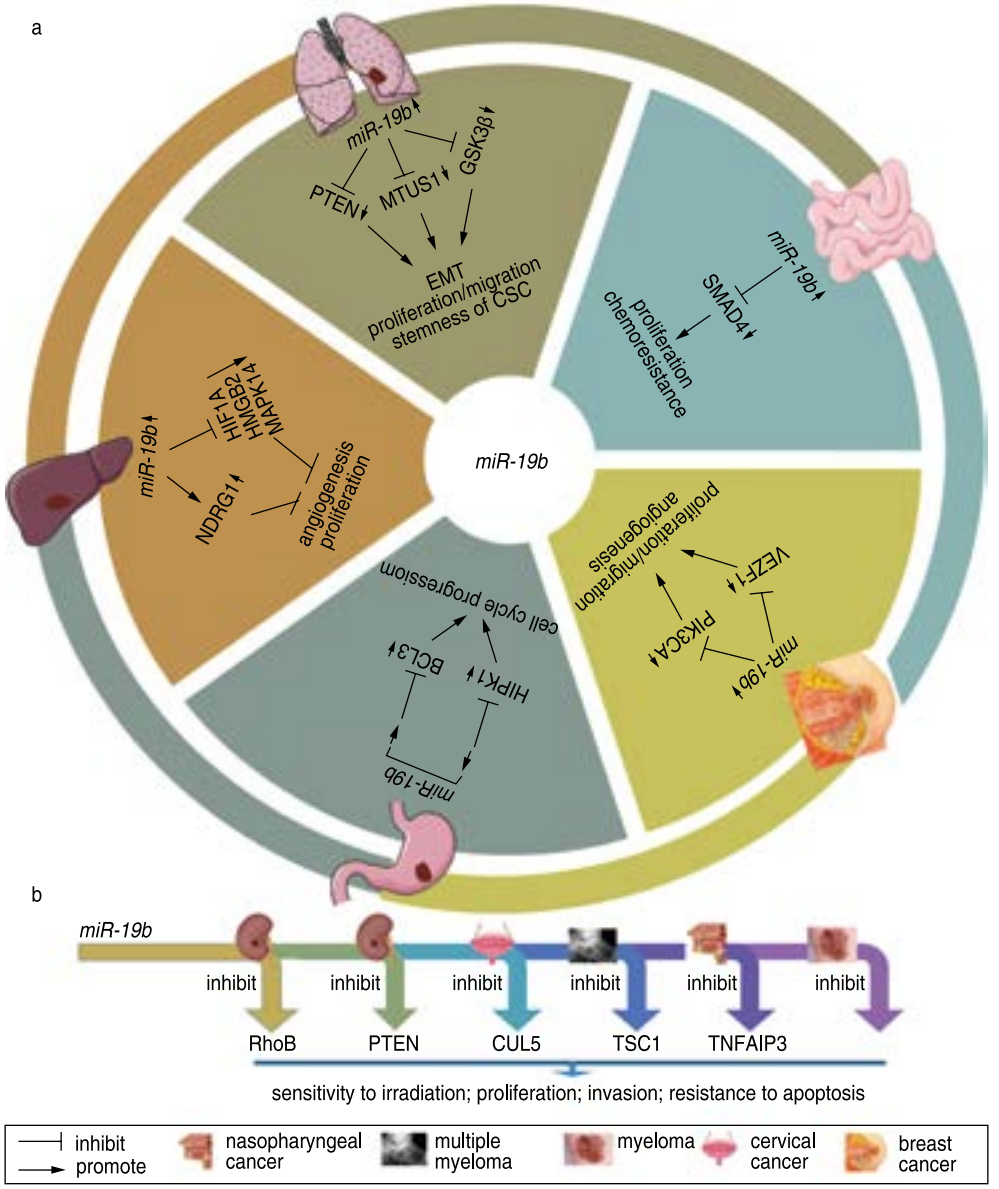

Fig. 1 Correlation between *miR-19b* and prognosis of cancer patients: (a) *miR-19b* roles in cancers with a high mortality rate; (b) *miR-19b* roles in cancers with a low mortality rate.

carcinoma [21], breast cancer [22–26], gastric cancer [27–28], lung cancer [29–31], liver cancer [32], colon cancer [17], renal cancer [33–35], cervical carcinoma [36], ovarian cancer [37], multiple myeloma [38] and melanoma [39]. These studies have confirmed the widespread roles of *miR-19b* in both high- and low-mortality-rate cancer types (Fig. 1). Fluctuating levels of *miR-19b* expression may affect tumor growth through different signaling pathways, but the prognostic role of *miR-19b* in different cancer types remains controversial. In addition, many studies have reported that *miR-19b* targets the PTEN-PI3K-AKT signaling pathway [22, 24–25, 33–34, 36–37, 40–42]. As a key kinase downstream of PI3K-AKT, mTOR can regulate tumor cell proliferation, growth, survival, and angiogenesis [43–44]. Therefore, we speculated that *miR-19b* may play a major role in cancers through the mTOR signaling pathway.

The majority of previously published meta-analyses have evaluated the diagnostic or prognostic value of miRNAs in cancers, but have not evaluated the association of miRNAs with specific pathways. Therefore, we performed this study to first assess the prognostic roles of *miR-19b* in human cancers and further explore the possible link between *miR-19b* and the mTOR signaling pathway based on this meta-analysis. These results may

provide new routes for the prevention and treatment of cancers.

Materials and methods

Our systematic review and meta-analysis was performed according to the recommendations of the PRISMA statement [45].

Literature search strategy

We comprehensively searched literature published up to November 25, 2019 using PubMed, Embase, Web of Science, and Cochrane Library databases. The search terms, [("*miR-19b*" or "microRNA-19b" or "*miR19b*") AND ("cancer" or "carcinoma" or "tumor" or "adenocarcinoma" or "neoplasm" or "neoplasia" or "malignancy" or "malignant") AND ("prognostic" or "prognosis" or "survival" or "outcome" or "recurrence" or "relapse" or "clinical features" or "clinicopathological parameters")] were used to identify the relevant studies.

Inclusion and exclusion criteria

Only those publications that met the following criteria were selected: (1) the relationship between *miR-19b* expression and patient prognosis was analyzed; (2) patients were separated into high/low groups based on *miR-19b* levels; and (3) sufficient data were provided to evaluate the prognostic role of *miR-19b*. The exclusion criteria were: (1) reviews, letters, case reports, animal trials, and expert opinions; and (2) studies without useful information.

Data extraction and quality assessment

Fundamental information from the included articles was carefully extracted by two authors. If the study only provided Kaplan-Meier curves, hazard ratios (HRs) and 95% confidence intervals (CIs) were manually calculated using Engauge Digitizer version 4.1 (https://zenodo.org/record/3941227). We preferably selected multivariate data when the article provided both uni- and multivariate results. The Newcastle-Ottawa Assessment Scale (NOS) was applied to evaluate the quality of the included publications, with a score equal to or greater than 6 indicating high quality.

Extraction and analysis of the Cancer Genome Atlas datasets

We used OncomiR to assess The Cancer Genome Atlas (TCGA) datasets relating *miR-19b* expression with cancer development. *miR-19b* expression data were available for 30 cancer types, including 9497 cases. Log2 mean expression values were used to compare *miR-19b* levels in normal and tumor tissues. Significance in tumor development was determined using a paired Student's *t*-test to compare *miR-19b* expression levels between normal and tumor tissues. Analysis of variance was performed to compare *miR-19b* expression levels between different cohorts for each clinical parameter.

Statistical analysis

Analyses were performed using Stata SE12.0 (STATA Corp, USA). Odds ratios (ORs) and 95% CIs were applied to analyze the relationship between *miR-19b* expression and tumor characteristics. The pooled HR and 95% CI were used to evaluate the prognostic value of *miR-19b*. HRs greater than 1 indicated that *miR-19b* was a factor leading to worse prognosis. Meanwhile, the Q test and I^2 statistics were assessed to evaluate the heterogeneity between included publications. $P < 0.05$ or $I^2 \geq 50\%$ indicated significant heterogeneity, in which case, we selected the random-effects model. Otherwise, we proceeded to the fixed-effects model. Additionally, subgroup and sensitivity analyses were performed to identify the source of heterogeneity, and publication bias was assessed by using a funnel plot and Begg's test.

Trial sequential analysis

Trial sequential analysis (TSA) can be used to avoid the risk of errors related to a small sample size. It can minimize the false positive/negative results caused by random errors. As shown in Fig. 2, curve A only crosses the traditional threshold (Z = 1.96), indicating that a false positive result may be obtained. However, more trials are needed to confirm this. Curve B crosses both the traditional and the TSA threshold, which indicates that a true positive result was obtained and no more trials are needed. Curve C crosses neither of the thresholds [Z = 1.96, TSA threshold and a priori information size (APIS)] and therefore, more trials are required to confirm the negative result. Curve D only exceeds the APIS, which indicates that there is no statistical difference and no more trials are needed to confirm the result. In this meta-analysis, TSA was performed to evaluate the reliability of

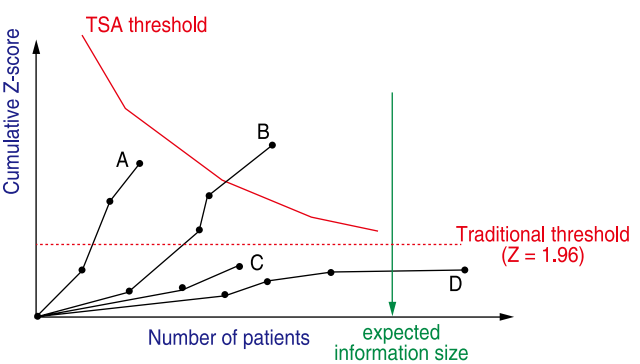

Fig. 2 Diagram of trial sequential analysis

our results, using the criteria of relative risk reduction = 15%, alpha = 5%, and statistical test power = 80%.

Target signaling pathway of *miR-19b*

We first generated an exhaustive collection of articles related to the mechanism of *miR-19b* in cancers. A series of information including the expression level of *miR-19b*, its target genes, its signaling pathways, and its role in cancer progression were extracted. The Kyoto Encyclopedia of Genes and Genomes (KEGG) database was then used to analyze the target mRNAs of *miR-19b* and their different signaling pathways, as reported in the literature. All of these processes were independently performed by two authors. Meanwhile, web-based tools, such as miRDB, miRTarBase, and TargetScan (http://mirdb.org/) were used to determine the effect of *miR-19b* on its target genes. Gene Ontology (GO) and KEGG pathway analyses of these genes were performed using R3.5.3 software, to verify the conclusions of the articles.

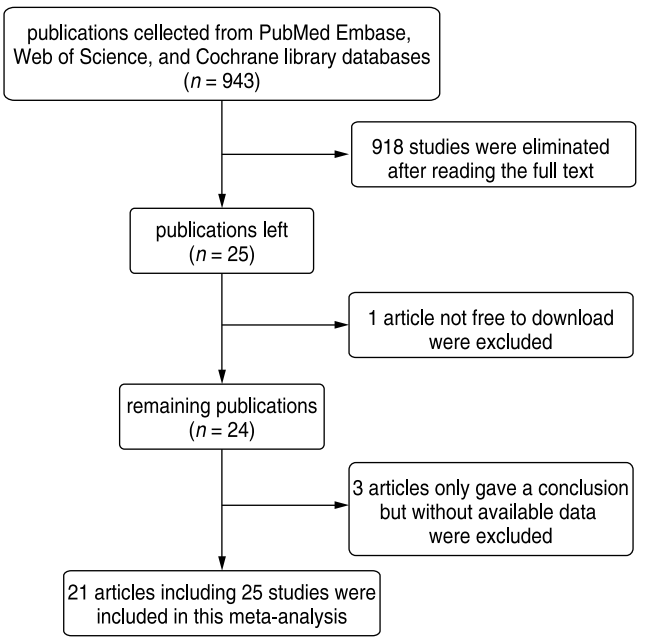

Fig. 3 Article search workflow and information extraction process

Table 1 HRs and 95% CIs of all selected studies

Outcome subgroup	First author, Year	Country	Cancer type	HR (95% CI)	*P* Value
OS (tissue)	Hung, 2015	China	HCC	0.318 (0.120–0.846)	0.022
	Wu, 2014	China	NSCLC	3.466 (1.389–8.650)	0.008
	Huang, 2016	China	NPC	2.967 (1.008–8.772)	< 0.05
	Li, 2018	China	BC	2.560 (1.130–5.796)	0.024
	Jiang, 2017	China	CC	2.23 (1.42–3.58)	0.008
	Wang, 2016	China	GC	0.62 (0.26–0.94)	0.002
	Marcela, 2016	Brazil	BL	0.54 (0.05–5.74)	0.207
	Xu, 2013	China	ESCC	1.77 (0.75–4.21)	0.764
	Zhao, 2017	China	BC	1.95 (1.13–3.37)	0.0092
	Huang, 2017	China	CRC	1.17 (0.21–6.53)	< 0.001
	Shao, 2018	China	GC	1.017 (0.981–1.054)	0.356
	Yu, 2012	China	CC	1.52 (1.09–2.11)	0.367
DFS (tissue)	Hung, 2015	China	HCC	0.455 (0.245–0.845)	0.013
	Jiang, 2017	China	CC	2.73 (1.76–3.89)	0.016
	Wang, 2016	China	GC	0.48 (0.21–0.87)	0.012
	Silvia, 2017	Spain	CRC	0.25 (0.08–0.78)	0.017
	Silvia, 2017	Spain	CRC	0.37 (0.14–0.98)	0.041
OS (Serum/Plasma)	Peng, 2018	China	GC	1.224 (0.856–1.751)	0.268
	Wu, 2014	China	NSCLC	1.800 (1.008–3.216)	0.047
DFS/PFS (Serum/Plasma)	Alfons, 2015	Spain	MM	0.10 (0.01–0.94)	< 0.0001
	Peng, 2018	China	GC	1.28 (0.947–1.729)	0.108
OS (BM)	Zhang, 2018	China	Non-M3 AML	1.68 (0.72–2.82)	0.118
	Zhang, 2018	China	CN AML	2.50 (0.98–5.25)	0.064
PR	Zhang, 2018	China	Whole AML	2.11 (1.62–3.67)	0.047
DFS (Urine)	Stuopelytė, 2016	Lithuania	PCa	0.45 (0.02–0.98)	0.014

Note: HCC: hepatocellular carcinoma; NSCLC: non-small cell lung cancer; NPC: nasopharyngeal carcinoma; BC: breast cancer; CC: colon cancer; GC: gastric cancer; BL: burkitt lymphoma; ESCC: esophageal squamous cell carcinoma; CRC: colorectal cancer; MM: multiple myeloma; AML: acute myeloid leukemia; CN AML: cytogenetically normal AML; PCa: prostate cancer.

Table 2 Pooled ORs for the relationship between *miR-19b* expression levels and clinicopathological features

Clinicopathological features	Studies	Heterogeneity			Model
		ORs (95% CIs)	I^2 (%)	*P*	
Tumor size (≤ 3cm *vs* > 3cm)	7	0.88 (0.46–1.69)	54.9	0.038	Random-effects
Tumor stage (I/II *vs* III/IV)	10	0.74 (0.30–1.81)	86.7	0.000	Random-effects
Vascular invasion (yes *vs* no)	4	0.94 (0.59–1.49)	44.9	0.142	Fixed-effects
LNM (yes *vs* no)	7	1.09 (0.39–3.01)	68.8	0.004	Random-effects
Tumor differentiation (W + M *vs* P)	9	1.00 (0.51–1.95)	66.2	0.003	Random-effects
DM (yes *vs* no)	3	3.43 (1.32–8.90)	56.1	0.102	Random-effects

Results

Study characteristics

Based on the aforementioned inclusion criteria, 943 articles were retrieved from PubMed, Embase, Web of Science, and Cochrane Library databases. After reading the entire text of these articles, 918 articles were eliminated due to the lack of useful information. Among the remaining 25 articles, one was not free to download and three others only gave a conclusion, without available data. Finally, 21 articles encompassing 25 studies were included in this meta-analysis (Fig. 3). A total of 2273 patients with 13 cancer types were distributed among the 21 articles. The fundamental information from these articles is summarized in Supplementary Table 1. All publications had NOS scores between 6 and 9, with an average score of 7 (Table 2). The HRs and 95% CIs of the articles are shown in Table 1.

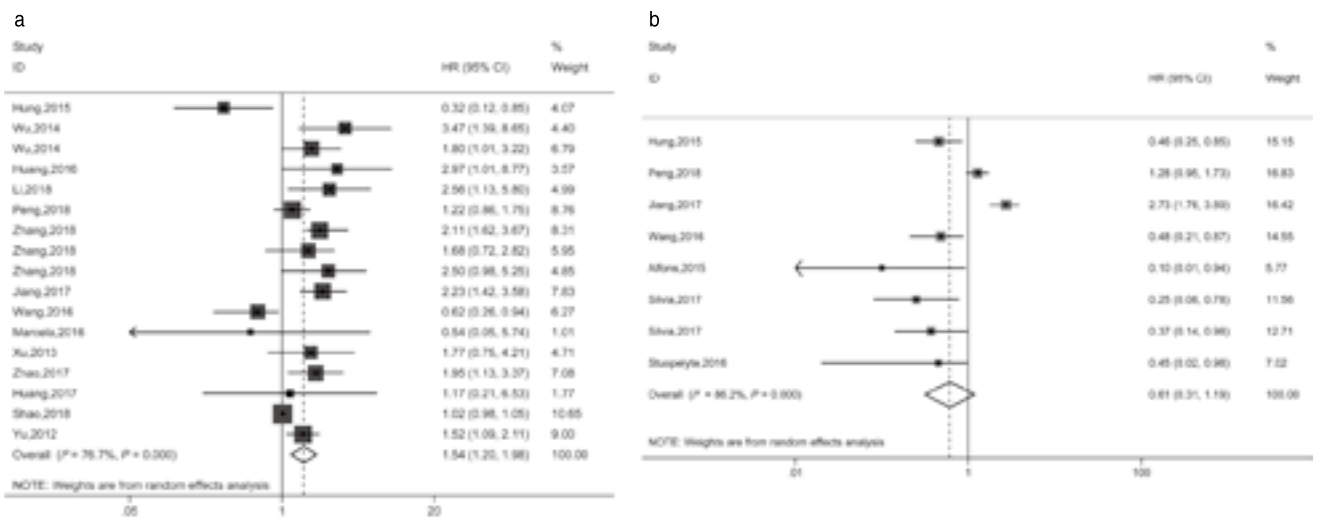

Fig. 4 Forest plots of the relationship between *miR-19b* expression levels and cancer patient prognosis: (a) OS; (b) DFS/PFS

Table 3 Relationship between *miR-19b* expression and cancer progression

Cancer type	Upregulated in	*P* Value	Clinical status	*P* Value	TCGA dataset
Bladder urothelial carcinoma	Tumor	< 0.0001	Pathologic M Status/Clinical T Status	0.0443/0.0265	TCGA-BLCA
Colon adenocarcinoma	Tumor	< 0.0001	Pathologic Stage	0.039	TCGA-COAD
Esophageal carcinoma	Tumor	0.00525	-	-	TCGA-ESCA
Kidney chromophobe	Normal	< 0.0001	Pathologic T Status	0.00184	TCGA-KICH
Liver hepatocellular carcinoma	Normal	0.00763	Pathologic N Status	0.0161	TCGA-LIHC
Lung adenocarcinoma	Tumor	0.00256	Pathologic T Status	0.0144	TCGA-LUAD
Lung squamous cell carcinoma	Tumor	0.000521	-	-	TCGA-LUSC
Prostate adenocarcinoma	Tumor	< 0.0001	-	-	TCGA-PRAD
Rectal adenocarcinoma	Tumor	0.0036	-	-	TCGA-READ
Stomach adenocarcinoma	Tumor	< 0.0001	-	-	TCGA-STAD
Thyroid carcinoma	Normal	< 0.0001	Pathologic Stage	0.000309	TCGA-THCA
Uterine corpus endometrial carcinoma	Tumor	< 0.0001	-	-	TCGA-UCEC

Correlation between *miR-19b* level and OS

A total of 17 studies were used to assess the correlation between *miR-19b* and overall survival (OS). Due to significant heterogeneity (I^2 = 76.7%, P < 0.05), the random-effects model was applied. Higher levels of *miR-19b* expression were found to be associated with shorter OS time (HR = 1.54, 95% CI = 1.20–1.98; Fig. 4a). Subgroup analysis (Fig. 5; Supplementary Table 3) further showed that *miR-19b* overexpression in tissues and bone marrow (BM) was correlated with poor OS in Asian populations (HR: 1.56, 95% CI: 1.21–2.01) and in groups with a sample size greater than 100 (HR = 1.86, 95% CI = 1.47–2.34). When cancers were classified as solid or non-solid tumors, we observed that *miR-19b* was more significantly associated with shorter OS time in non-solid (HR = 2.06, 95% CI = 1.49–2.84) than solid tumors (HR = 1.44, 95% CI = 1.10–1.88).

Correlation between *miR-19b* level and DFS/PFS

A total of 8 studies discussed the correlation between *miR-19b* levels and disease-free/progression-free survival (DFS/PFS). In line with the above analyses on the correlation between *miR-19b* and OS, we used a random-effects model (I^2 = 82.6%, P < 0.05) to explore its relationship with DFS/PFS. There was no correlation found between *miR-19b* levels and DFS/PFS (HR = 0.61, 95% CI = 0.31–1.19, Fig. 4b). Since *miR-19b* level of had no effect on cancer patient DFS/PFS, we did not perform further subgroup analyses.

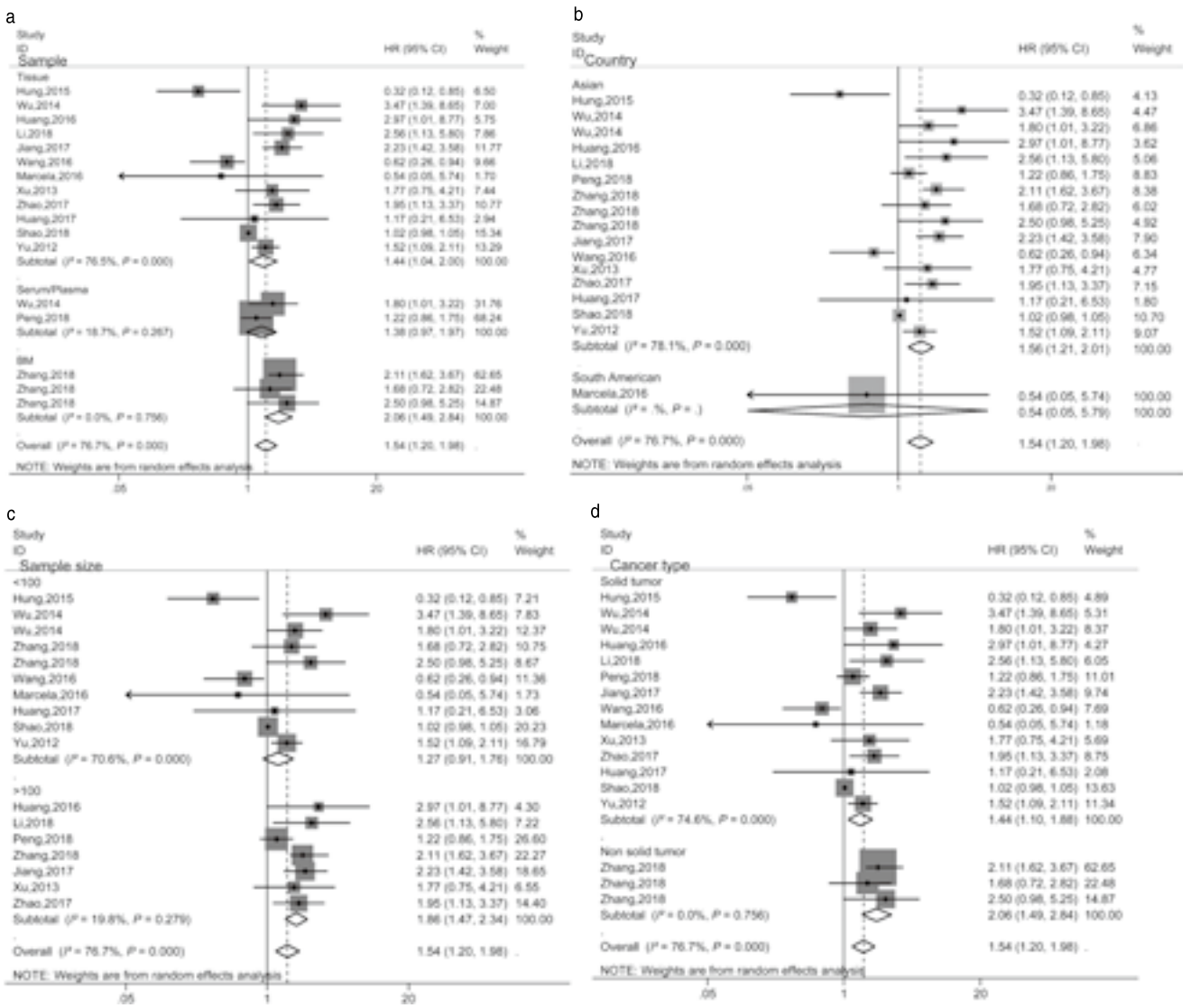

Fig. 5 Subgroup analysis of OS stratified by (a) detection sample; (b) study country; (c) sample size; and (d) cancer type

Correlation between *miR-19b* level and clinicopathological features

There were 13 studies that focused on the relationship between *miR-19b* and clinicopathological features, including tumor size, tumor stage, vascular invasion, lymph node metastasis, and distant metastasis (DM). As shown in Table 2, *miR-19b* level was only related to cancer DM (OR = 3.43, 95% CI = 1.32–8.90). Further subgroup analysis was not performed due to insignificant heterogeneity.

Correlation between *miR-19b* level and tumor progression in TCGA dataset

Under the predetermined significance threshold of $P \leq 0.05$, *miR-19b* expression was significantly associated with tumorigenesis in 12 cancer types from the TCGA dataset. *miR-19b* was significantly up-regulated in nine of these cancer types, but down-regulated in three others. In addition, a correlation between *miR-19b* level and clinicopathological status was also observed in some cancers. The relationship between *miR-19b* expression and cancer progression is presented in Table 3.

Sensitivity analysis and publication bias

Sensitivity analysis was carried out to identify which articles impacted heterogeneity. These results indicated that pooled HRs would not be greatly affected by excluding any study, which indicated that the above analyses were reliable and credible (Fig. 6a and 6b). We next applied funnel plots and Begg's test to estimate the publication bias of the included studies. The funnel plot, displayed in Fig. 6, showed *P* values of 0.902 for OS and 0.063 for DFS/PFS, indicating that there was no publication bias in this Meta-analysis.

Reliability and clinical applicability of results

We performed TSA to evaluate the reliability and clinical applicability of our results. From the results (Fig. 7), we can see that the cumulative Z-curve of OS was similar to curve B in Fig. 2, indicating that a true positive result was obtained. It stipulated that high expression of *miR-19b* was associated with poor OS. No more trials are needed to support this conclusion. In addition, the cumulative Z-curve of DFS/PFS looked like curve C in Fig. 2, which meant that more trials are required to prove

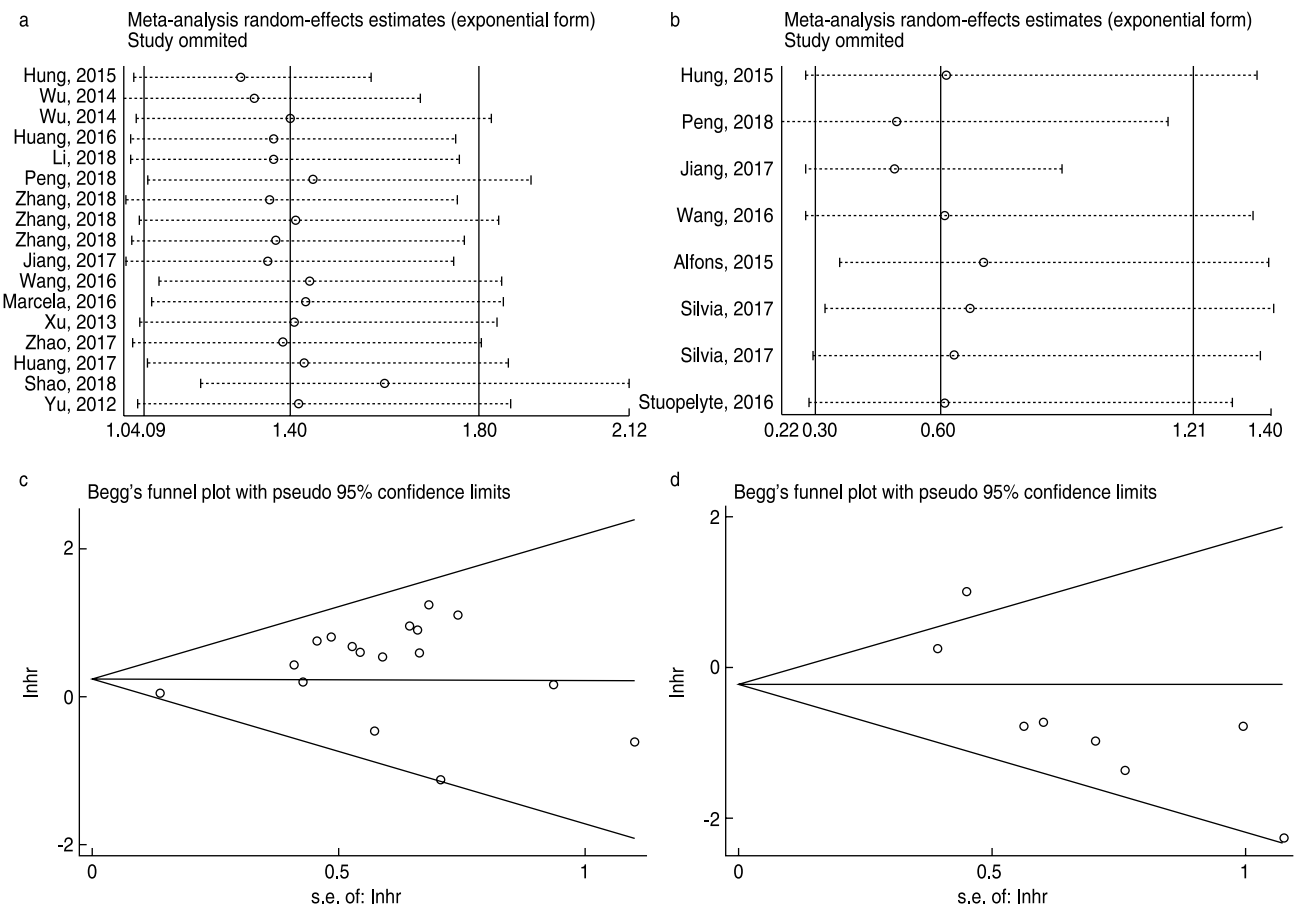

Fig. 6 Sensitivity and publication bias of studies: (a) sensitivity analyses for OS and (b) DFS/PFS and (c) funnel plot for OS (d) and DFS/PFS

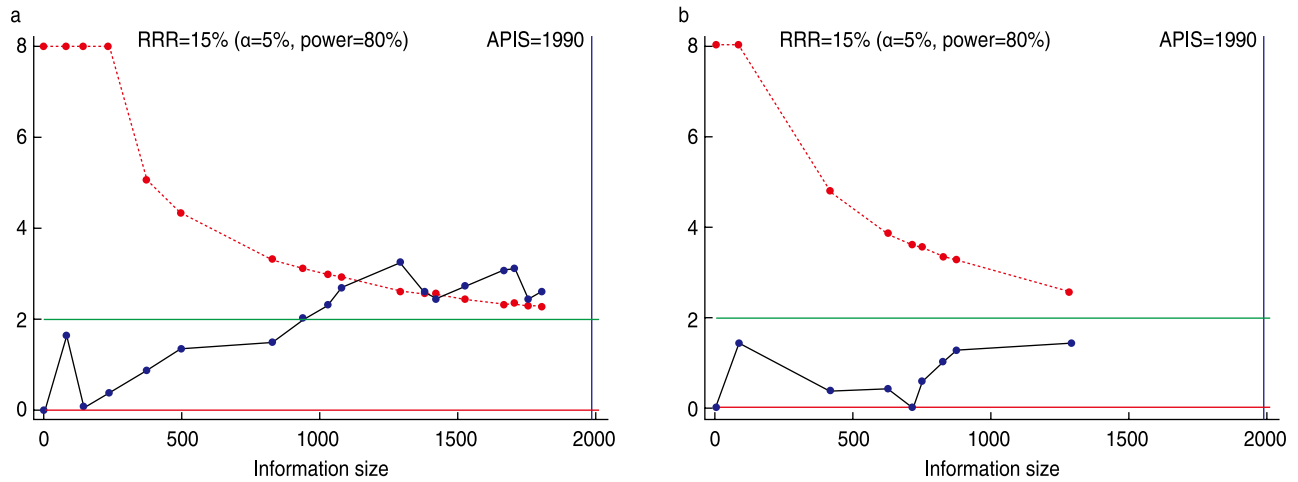

Fig. 7 TSA for cancer prognosis based on APIS: (a) OS; (b) DFS/PFS

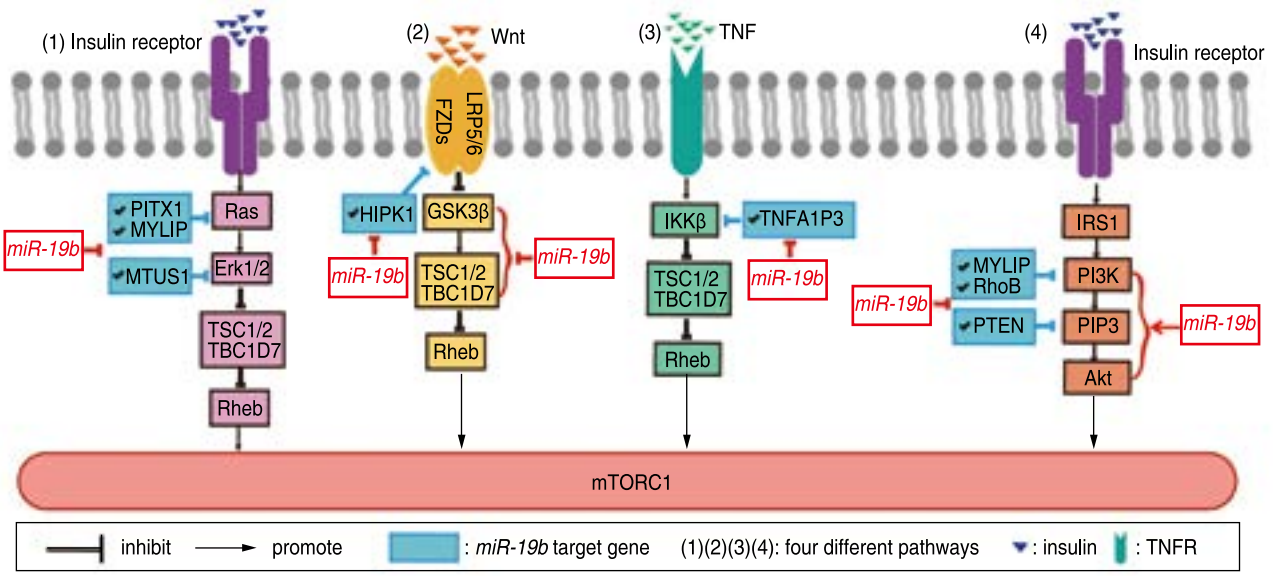

Fig. 8 Molecular mechanism showing the link between *miR-19b* and the mTOR signaling pathway in cancers

the conclusion that *miR-19b* was not correlated with DFS/PFS.

Connection between *miR-19b* and mTOR signaling pathway

To clarify the biological mechanism of the involvement of *miR-19b* in cancers, we reviewed published articles that focused on the target genes of *miR-19b* and its biological mechanism. The relevant information is listed in Supplementary Table 4. These data indicated that *miR-19b* was involved in various signaling pathways, but its involvement in the PTEN-PI3K-AKT signaling pathway was most notable. Some other studies also reported its involvement in the RAS and Wnt/β-catenin signaling pathways. Then, based on data from KEGG, we observed that mTOR was the intersection point of these different signaling pathways. These pathways could affect the mTOR signaling pathway through related molecules. A diagrammatic summary of the mechanism related to the correlation of *miR-19b* with the mTOR signaling pathway is presented in Fig. 8. Three additional databases miRDB, miRTarBase, and TargetScan, were then used to verify our findings. By intersecting the predicted target genes from these tools, 274 genes were selected and the functional annotations of these genes was performed by GO and KEGG analysis. As displayed in Fig. 9, these 274 genes mainly participated in the "mTOR signaling pathway", which verified our findings.

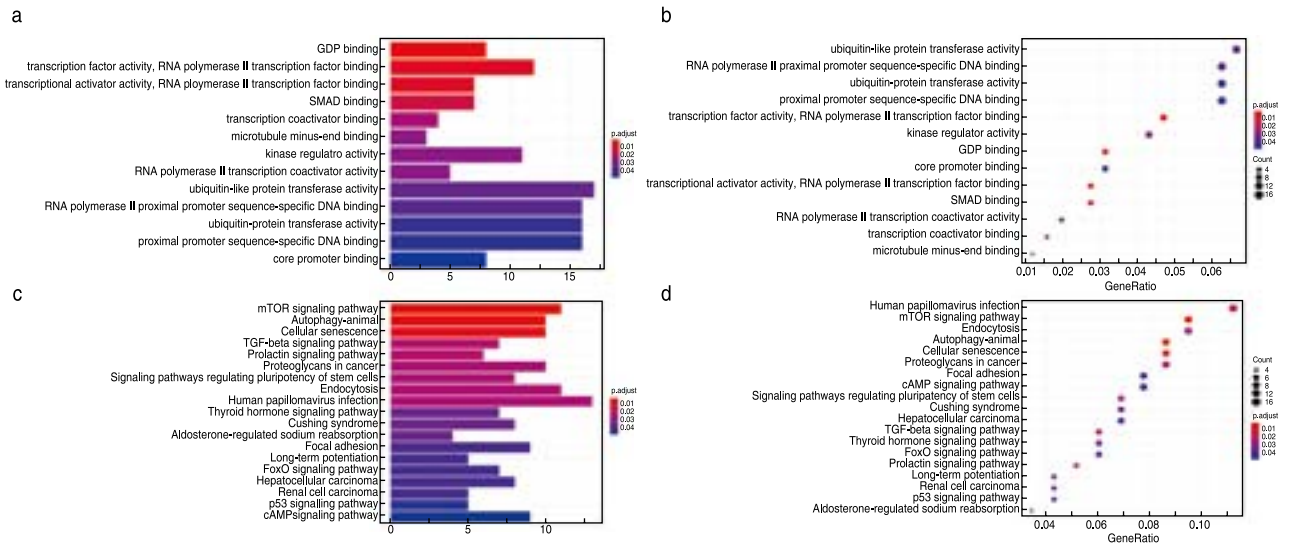

Fig. 9 Bioinformatics supporting the correlation between *miR-19b* and mTOR in cancers: (a) and (b) enrichment of the top 13 Gene Ontology terms, (c) and (d) Kyoto Encyclopedia of Genes and Genomes pathways.

Discussion

miR-19b, is a member of the *miR-17-92* cluster, which contains *miR-17*, *miR-18a*, *miR-19a*, *miR-19b*, *miR-20a*, and *miR-92*, and is located on chromosome 13q31.3. In 2004, Ota *et al* [46] discovered for the first time that the *miR-17-92* cluster is involved in the pathogenesis of B-cell lymphoma. A few years later, Mu *et al* [47] found that *miR-19b* was the principal element of the *miR-17–92* cluster. As a result, it has been widely speculated that *miR-19b* may serve as a biomarker in various types of cancers. Recently, an increasing number of studies have shown that *miR-19b* may serve as a predictor of unfavorable prognosis in cancer patients, because it may lead to the downregulation of tumor suppressor genes or the upregulation of tumor promoter genes. The overexpression of *miR-19b* is associated with poorer prognosis in patients with non-small cell lung cancer [48], nasopharyngeal carcinoma [21], breast cancer [22], ovarian cancer [37], acute myeloid leukemia [49], and colon cancer [17]. Julia *et al* [50] demonstrated that *miR-19b* regulates apoptosis-related activities in tumors through related genes. Wang *et al* [51] reported that *miR-19b* affects tumor growth and grade in gastric cancer. By contrast, some publications have reported that *miR-19b* plays inhibitory roles in tumor development, hence improving patient prognosis. This inhibitory role has been found mainly in hepatocellular carcinoma [32], gastric cancer [27], and multiple myeloma [52]. In other studies, no association between *miR-19b* and cancer prognosis has been found in Burkitt's lymphoma [53] and esophageal squamous cell carcinoma [54]. We can conclude that the prognostic roles of *miR-19b* in different cancers remain controversial. Therefore, we performed this meta-analysis to evaluate the prognostic and clinicopathological role of *miR-19b* in cancers.

The clinically relevant indicators of patient survival include OS, DFS, PFS, and recurrence-free survival (RFS). Due to the lack of relevant literature on RFS, in this meta-analysis, we assessed the relationship between *miR-19b* and OS and DFS/PFS. The results of pooled HRs suggested that high expression levels of *miR-19b* may result in shorter OS time (pooled HR = 1.54, 95% CI=1.20–1.98), whereas there was no influence on DFS/PFS (pooled HR = 0.61, 95% CI = 0.31–1.19). This difference observed in the prediction of OS and DFS/PFS by *miR-19b* may be due to differences in the measurement of these indicators in each study. These differences may result from bias in measuring PFS, difficulties during the end-point collection of DFS data, and the possible influence of other diseases. Nevertheless, OS represents the time from the observation period to the death of the patient.

Due to the large degree of heterogeneity among the included studies, four subgroup analyses were performed to further explore the role of *miR-19b* by sample type, country, sample size, and cancer type. The *miR-19b* level in tissues and BM had a clear impact on OS. This result indicated that *miR-19b* levels in tissues (for solid tumors) and BM (for non-solid tumors) were more meaningful than plasma or serum levels for predicting OS. However, increased *miR-19b* levels in BM were associated with a greater decrease in OS time (HR = 2.06, 95% CI = 1.49–2.84) when compared with increased *miR-19b* levels in tissues (HR = 1.44, 95% CI = 1.04–2.0). This conclusion requires further verified, since there were only three studies in the BM subgroup. When the sample size was

larger than 100, *miR-19b* could predict OS time (HR = 1.86, 95% CI = 1.47–2.34) and showed less heterogeneity, indicating that sample size could influence the accuracy of the conclusions of a study. When grouping patients by cancer type, we observed that *miR-19b* level was a potential biomarker of prognosis in both solid and non-solid tumors.

Based on the above results, we concluded that *miR-19b* has a wide application value in predicting OS time in patients with either solid or non-solid tumors, and high *miR-19b* expression levels lead to shorter OS time in cancer patients. After investigating the publications identified in online database searches, we analyzed TCGA data to further refine the conclusions drawn from the published articles. These data showed that the dysregulation of *miR-19b* was correlated with cancer clinical stage. This further indicated a major role of *miR-19b* in promoting cancers. Combined with the effect of *miR-19b* on DM (OR = 3.43, 95% CI = 1.32–8.90), we speculated that *miR-19b* may affect cancer prognosis by promoting DM. An increasing number of recent studies have suggested that circulating tumor cell (CTC) entry into the bloodstream to reach distant organs is a key step in the initiation of metastasis [55]. Vascular formation, immunosuppression, and epithelial-mesenchymal transition (EMT) of CTCs may be involved in DM. Interestingly, Li *et al* showed that high expression levels of *miR-19b* may promote EMT and thus, enhance the migration and invasion ability of lung cancer cells [29]. Wang *et al* even suggested that *miR-19b* may trigger EMT via exosomes secreted by clear cell renal cell carcinoma stem cells [35]. Mao *et al* [56] came to the same conclusion that *miR-19b* is largely involved in EMT via the *miR-19b*-PTEN-AKT signaling pathway.

Since high levels of *miR-19b* expression were correlated with poor prognosis in various types of cancers, it may be considered as a therapeutic target. A new therapeutic strategy may be possible by targeting the relevant pathway affected by *miR-19b* [57]. However, *miR-19b* plays different roles in cancers through different pathways. The relationship between these pathways and the mechanism by which they involve *miR-19b* in cancers remain unclear. Thus, it is critical to find a single pathway that connects these different pathways involved in the oncogene role of *miR-19b*.

Most of the reviewed articles reported that *miR-19b* overexpression activates the PTEN-PI3K-AKT pathway via the direct targeting of PTEN, RhoB, MYLIP, and CUL5 in pancreatic cancer [42], multiple myeloma [40], breast cancer [22, 24–25, 41], ovarian cancer [37], Wilms' tumor [34], clear cell renal cell carcinoma [33], and cervical carcinoma [36].

Ohira *et al*. found that PITX1, which acts as a negative regulator of the RAS pathway [58], is downregulated by *miR-19b* in melanoma [59]. Moreover, Gu *et al*. demonstrated that *miR-19b* overexpression plays a key role in downregulating MTUS1 [60]. MTUS1 has been shown to interfere with ERK2, which is a part of the RAS pathway [61]. These studies confirmed that *miR-19b* is correlated with tumor progression by targeting the RAS pathway.

Recently, Zhu *et al*. showed that *miR-19b* activates the Wnt/β-catenin pathway by directly targeting GSK3β in lung cancer [31]. Based on the study of Wu *et al*, it is well established that *miR-19b* modulates the Wnt-β-catenin signaling pathway by regulating HIPK1 [28]. Thus, *miR-19b* may affect cancer progression by activating the Wnt-β-catenin pathway.

Wang *et al*. showed that differential expression of *miR-19b* regulates the TSC1/mTOR signaling pathway in multiple myeloma [38]. Furthermore, *miR-19b* has been shown to suppress TNFAIP3 in nasopharyngeal carcinoma [21]. According to the KEGG pathway database, as a negative feedback of the NF-κB axis, the suppression of TNFAIP3 may activate NF-κB through IKKβ, which in turn may regulate mTOR. Interestingly, the study of Jiang *et al* first illustrated that *miR-19b* plays a key role in colon cancer progression via SMAD4 [17]. Voorneveld *et al*. then found that SMAD4 was associated with poor prognosis in colorectal cancer through the BMP pathway, rather than the TGF-β signaling pathway [62]. Moreover, Karner *et al*. confirmed that the BMP family activates the mTORC1 signaling pathway, which promotes the expression of protein anabolism genes [63]. We can conclude that *miR-19b* promotes colorectal cancer progression via the *miR-19b*-SMAD4-BMP-mTOR axis. In brief, the above analyses indicated that the pro-carcinogenic mechanism of *miR-19b* involved the activation of the mTOR signaling pathway.

In summary, these data indicated that *miR-19b* may act as an onco-miR through activation of the PTEN-PI3K-AKT, RAS, Wnt/β-catenin, and mTOR signaling pathways. Thus, it remains important to explore the possible connection between these pathways in the pro-carcinogenic role of *miR-19b*. As shown in Fig. 8, we observed that *miR-19b* played a major role in cancers through the mTOR signaling pathway, which is the intersection point of these different pathways. GO and KEGG analysis verified these findings. Recently, many studies have demonstrated that mTOR may be a therapeutic target for cancers [64–65]. Taken together, these findings indicated the potential to improve cancer treatment by regulating *miR-19b*-related mTOR signaling pathways.

In spite of the rigorous protocols adopted in each process of the analysis, the bias and limitations of this study cannot be ignored. Firstly, four studies were excluded after reviewing the available data. The absence of these articles may have impacted our analysis.

Secondly, HRs were manually extracted from the survival curves for some studies, which may have introduced some errors. Thirdly, we originally planned to also explore the anti-cancer effect of *miR-19b*, but we did not identify a sufficient number of articles for this analysis. Finally, more high-quality, large-sample-size publications are required to confirm our conclusions.

Despite the above limitations, this is the first study to link meta-analysis results with a specific mechanism. This meta-analysis is the first to identify the relationship between *miR-19b* and prognosis, showing that high expression levels of *miR-19b* lead to poor OS by promoting distant metastasis. However, DFS/PFS was not influenced by *miR-19b*. In summary, the results of our study indicate that *miR-19b* may have an oncomiR role through the mTOR signaling pathway.

Conflicts of interest

The authors indicated no potential conflicts of interest.

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