Received: 3 October 2021 | Accepted: 5 November 2021

DOI: 10.1111/1759-7714.14242

ORIGINAL ARTICLE

WILEY

MicroRNA-21 was a promising biomarker for lung carcinoma diagnosis: An update meta-analysis

Haiyan Wang | **Jia Xu** | **Ling Ding**

Department of Respiratory, Hangzhou Third People's Hospital, Hangzhou, China

Correspondence
Haiyan Wang, Department of Respiratory, Hangzhou Third People's Hospital, Hangzhou Zhejiang Province, China.
Email: mishangzhilu@yeah.net

Abstract

Objective: To investigate the diagnostic performance of microRNA-21 detected in serum or sputum as a biomarker for lung carcinoma identification through pooling the open published data.

Methods: Clinical diagnostic studies related to microRNA-21 as a biomarker for lung carcinoma identification were electronically searched in the databases of Pubmed, EMBASE, Cochrane Library, Chinese National Knowledge Infrastructure, Wanfang, and Google Scholar. The data of the included studies was extracted and made pooling of diagnostic sensitivity, specificity, diagnostic odds ratio (DOR), area under the summary receiver operating characteristic curve (ROC) (AUC) for microRNA-21 expression in serum or sputum as a biomarker for lung carcinoma identification. The publication bias was evaluated by Deek's funnel plot.

Results: Seventeen diagnostic studies were finally included and made data pooling. For the included 17 studies, 4 investigated the microRNA-21 expression in sputum and 13 studies in serum. The pooled diagnostic sensitivity and specificity were 0.73 (95% CI, 0.67–0.78) and 0.81 (95% CI, 0.75–0.85), respectively, under random effect model. The combined DOR was 9.65 (95% CI, 6.64–14.03) with the AUC of 0.84 (95% CI, 0.80–0.87). Given a pre-test probability of 50%, the post-test positive probability and post-test negative probability were 79% and 25%, respectively, by using microRNA-21 as a biomarker for lung carcinoma diagnosis. Deek's funnel was obviously asymmetry and indicated significant publication bias ($p < 0.05$).

Conclusion: MicroRNA-21 in serum or sputum was a promising biomarker for lung cancer identification with relative high diagnostic sensitivity and specificity.

KEYWORDS
biomarker, diagnosis, lung carcinoma, meta-analysis, microRNA-21

INTRODUCTION

Carcinoma of the lung is considered as the leading cause of death in both males and femalse for cancer-related diseases.[1] It was reported that 2 206 771 new cases of lung cancer and 1 796 144 deaths from lung carcinoma were identified in the year 2020, which accounted for 11.4% and 18.0% of all cancers diagnosis or death of the same year.[2] Lung cancer has become a major problem of public health and has imposed a heavy burden for the government health care system.[3] However, the general prognosis and long-term survival of lung cancer is low, especially for advanced stage cases mainly due to lack of effective early diagnosis or screening methods.[4] At present, the chest computed tomography (CT) examination and X-ray are major tools for lung cancer screening in high risk subjects such as heavy smokers, elderly people, or people with family history of malignant tumor.[5,6] Bronchoscopy was mainly applied for cases with suspected lung disease or carcinoma, but rarely applied as routine lung cancer screening because of its invasive character. However, chest CT examination and bronchoscopy both have their limitations (eg, low specificity, sensitivity,

This is an open access article under the terms of the Creative Commons Attribution-NonCommercial-NoDerivs License, which permits use and distribution in any medium, provided the original work is properly cited, the use is non-commercial and no modifications or adaptations are made.
© 2021 The Authors. *Thoracic Cancer* published by China Lung Oncology Group and John Wiley & Sons Australia, Ltd.

or invasive). Serum and sputum are optimal humoral components for lung cancer diagnosis.[7–9]

MicroRNA-21 expression levels in serum or sputum applied as a diagnostic biomarker of lung cancer was reported in previously studies.[10,11] However, the exact diagnostic performance was not clear because of the small sample size and clinical heterogeneity between each relevant study. In the present work, we identified all the open published studies relevant to microRNA-21 as a biomarker for lung carcinoma diagnosis and performed data pooling to further evaluate its diagnostic performance and clinical application.

MATERIAL AND METHODS

Publication searching in the databases

Clinical diagnostic studies related to microRNA-21 as a biomarker for lung carcinoma identification were electronically searched in the databases of Pubmed, EMBASE, Cochrane Library, Chinese National Knowledge Infrastructure, Wanfang, and Google Scholar. The key words of "lung cancer," "carcinoma of the lung," "non-small cell lung cancer," "NSCLC," "microRNA-21," "miR-21," and "has-mir-21" were applied for publication electronic databases searching.

Publication inclusion and exclusion criteria

The trials inclusion criteria included: (i) prospective diagnostic studies about microRNA-21 as a biomarker for lung carcinoma diagnosis; (ii) pathology or cytology were applied as gold lung cancer diagnosis standard; (iii) microRNA-21 level was detected in serum or sputum; (iv) the data of true-positive (tp), false-positive (fp), false-negative (fn), and true-negative (tn) can be extracted or calculated form the original study; and (v) the study was published in English or Chinese.The publication exclusion criteria included: (i) studies in animal or case report or literature review; (ii) microRNA-21 expression detected in tumor or corresponding normal tissue not serum or sputum; (iii) duplicated publication or data; and (iv) publication without enough data to calculate tp, fp, fn, and tn.

Data and information extraction from included study

The following information and data was extreacted from the orignianl inlcuded studies: (i) first and corresponding authors; (ii) year of the data published; (iii) area the work performed; (iv) control type; (v) lung cancer type; and (vi) data of tp, fp, fn, and tn.

Quality assessment of the included studies

H.W. and J.X. made the methodological qualities assessment of each included study independently. The QUADAS was applied to evaluate the methodological qualities of the included diagnostic studies.[12]

Publication bias evaluation

The Deek's funnel plot was used to evaluate the publication bias. If the funnel plot was symmetrical and $p > 0.05$, the publication bias was not obvious. Otherwise, it was consider of significant publication bias.

Statistical analysis

Stata12.0 (Stata Corporation; http://www.stata.com) was applied for data managing. Before pooling the data, the statistical heterogeneity between the included 17 studies was tested by χ^2 test. If $p < 0.05$, the data was pooled by random effect model, otherwise by fixed effect model. $p < 0.05$ was considered of statistical significance.

RESULTS

Characteristic of the included studies

Initially, 824 publications relevant to microRNA-21 as a biomarker for lung carcinoma diagnosis were identified by searching the electronic databases. After removing the duplicated data and publications, 798 studies were screened in title and abstract. Twenty-eight studies were reviewed in full text and 17 publications were finally included for meta-analysis (Figure 1). For the included 17 studies, four investigated the

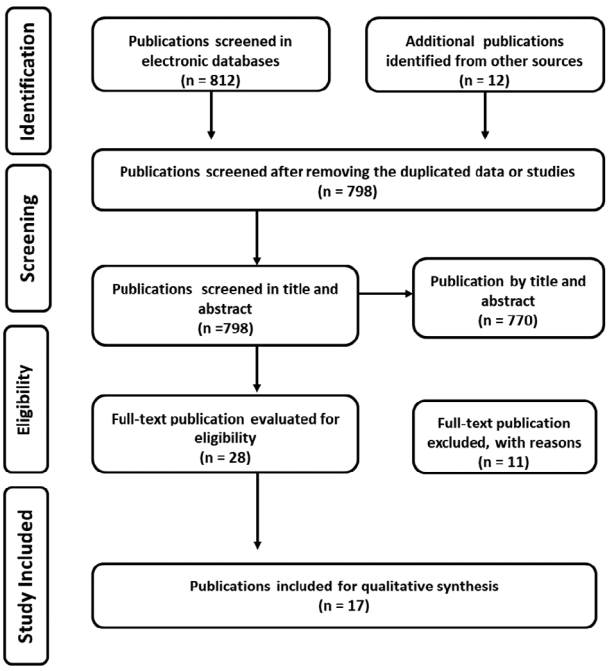

FIGURE 1 Publications searching and screening strategy in the relevant electronic databases

TABLE 1 The main features for the included studies

Author	Year	Country	Tp	Fp	Fn	Tn	Tissue	Race	Cancer type	Control type	QUADAS score
Su et al.[13]	2015	United States	42	14	14	59	Sputum	African, Caucasus	NSCLC	Smoker	11
Xing et al.[14]	2015	United States	47	17	13	45	Sputum	Caucasus	NA	Benign lung disease	9
Yang et al.[15]	2013	China	20	6	4	18	Sputum	Caucasus	NSCLC	Benign lung disease	7
Yu et al.[16]	2010	United States	26	7	10	29	Sputum	African, Caucasus	NSCLC	Healthy control	7
Li et al.[17]	2011	China	16	0	4	10	Serum	East Asian	Lung cancer	Healthy control	12
Wei et al.[18]	2011	China	48	9	15	21	Serum	East Asian	NSCLC	Healthy control	11
Tang et al.[19]	2013	China	30	13	32	47	Serum	East Asian	Lung cancer	Healthy smoker	10
Wei et al.[20]	2011	China	47	6	30	30	Serum	East Asian	NSCLC	Healthy control	8
Chen et al.[21]	2014	China	76	27	24	73	Serum	East Asian	NSCLC	Healthy control	11
Wang et al[22]	2012	China	27	10	4	29	Serum	East Asian	Lung cancer	Normal control	9
Shen et al.[23]	2011	United States	66	46	42	96	Serum	African American, White	NSCLC	Benign lung disease, Healthy control	12
Abd-El-Fattah et al.[24]	2013	United States	56	5	9	32	Serum	Egyptian		Healthy control	12
Le et al.[25]	2012	China	38	4	44	46	Serum	East Asian		Normal control	11
Wang et al.[26]	2021	China	47	2	14	24	Serum	East Asian	Lung cancer	Healthy control	8
Li et al.[27]	2019	China	27	0	5	44	Serum	East Asian	NSCLC	Benign lung disease, Healthy control	7
Zhao et al.[10]	2015	China	59	17	21	43	Serum	East Asian	NSCLC	Healthy control	11
Yang et al.[11]	2015	China	105	87	47	213	Serum	East Asian	NSCLC	Healthy control	10

Abbreviations: Fn, false negative; Fp, false positive; NA, not available; NSCLC, non-small cell lung cancer; Tn: true negative; Tp: true positive.

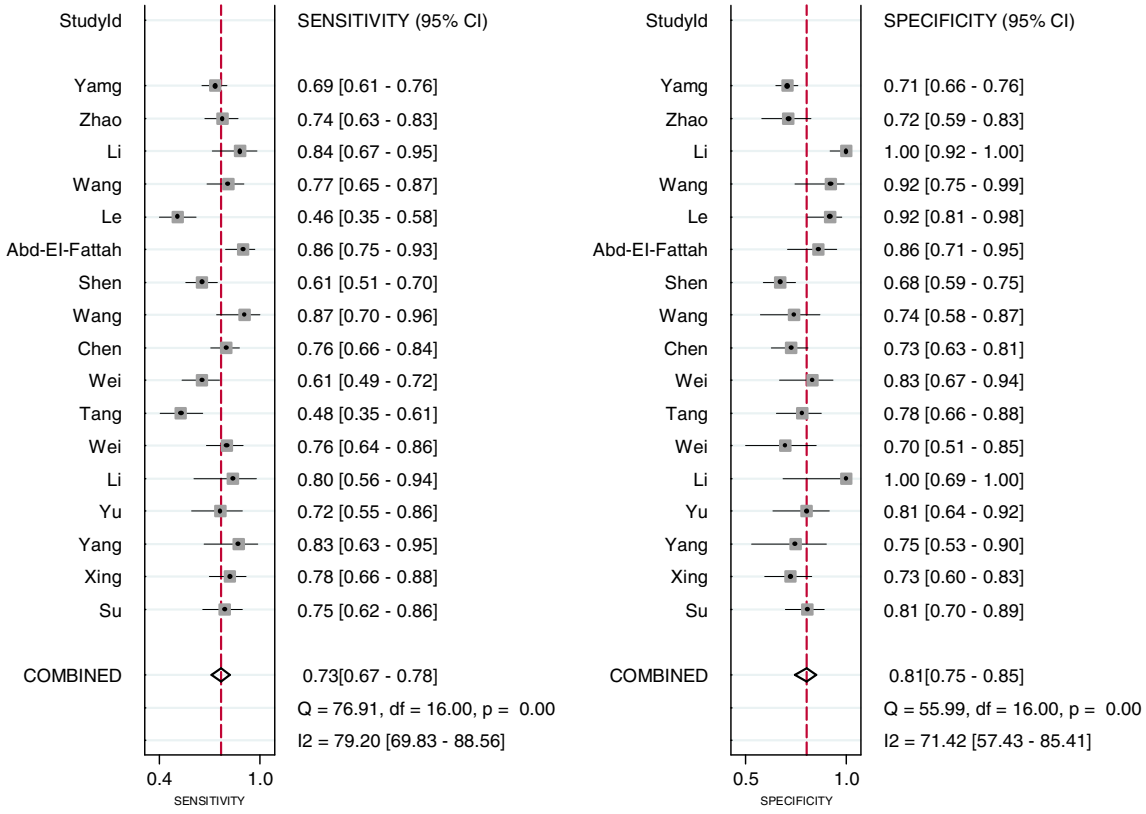

FIGURE 2 Forrest plot of the diagnostic performance of sensitivity and specificity for microRNA-21 as a biomarker in lung carcinoma identification

microRNA-21 expression in sputum and 13 studies investigated the microRNA-21 expression in serum. The general clinical features of the included 17 studies are shown in Table 1.

META-ANALYSIS RESULTS

Diagnostic sensitivity and specificity

The diagnostic sensitivity and specificity were pooled in random effect model because of statistical heterogeneity. The aggregated sensitivity, specificity were 0.73 (95% CI, 0.67–0.78) and 0.81 (95% CI, 0.75–0.85), respectively (Figure 2).

Diagnostic odds ratio and area under the summary receiver operating characteristic

Because of statistical heterogeneity, the diagnostic odds ratio (DOR) was pooled in random effects model (Figure 3). The combined DOR was 9.65 (95% CI, 6.64–14.03). The area under the summary receiver operating characteristic (SROC) was 0.84 (95% CI, 0.80–0.87) (Figure 4).

Fagan's nomogram analysis

Given a pre-test probability of 50%, the post-test positive probability, and post-test negative probability were 79% and 25%, respectively, using microRNA-21 as a biomarker for lung carcinoma diagnosis (Figure 5).

Sub-group analysis

Sub-group analysis was performed according the microRNA-21 detection in sputum or serum. The pooled

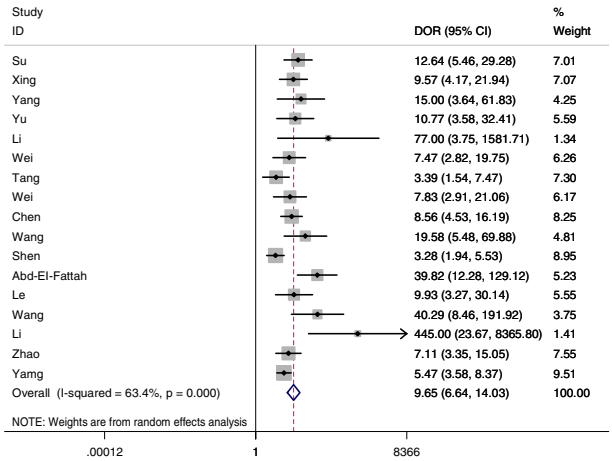

FIGURE 3 Forrest plot of diagnostic odds ratio for microRNA-21 as a biomarker in lung carcinoma identification

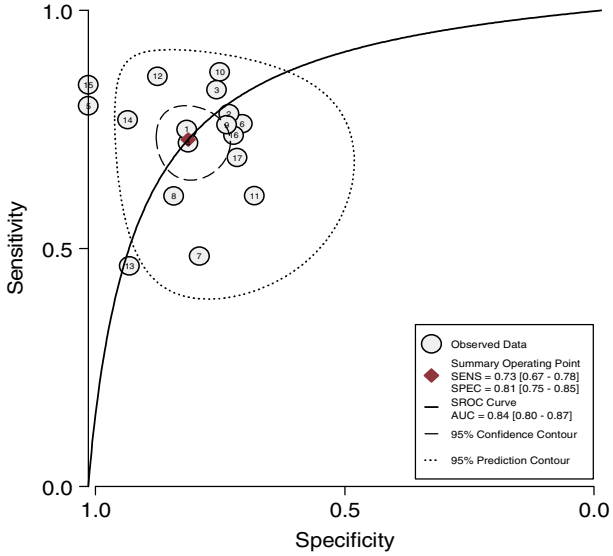

FIGURE 4 SROC of the diagnostic performance of sensitivity and specificity for microRNA-21 as a biomarker in lung carcinoma identification

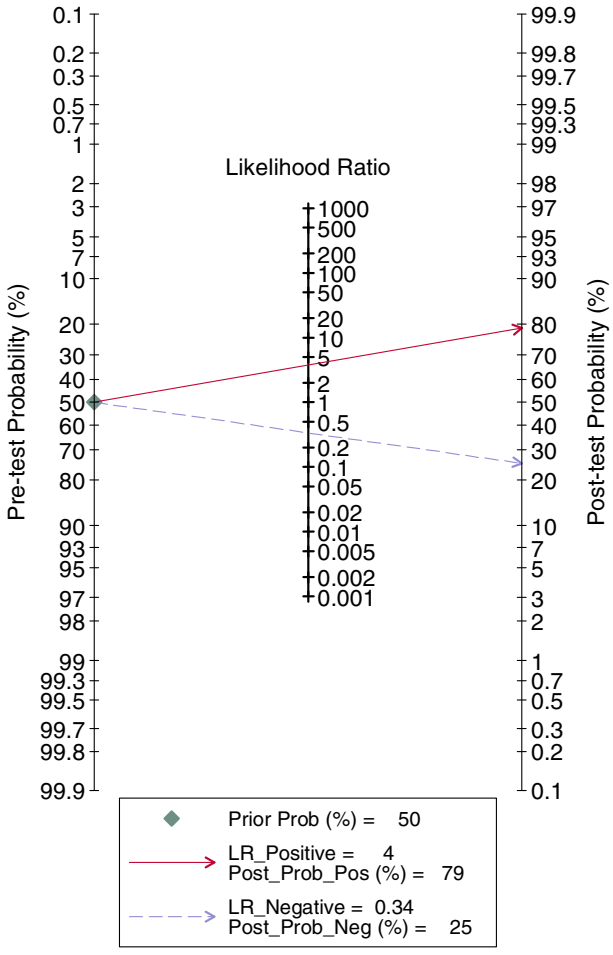

FIGURE 5 Fagan's nomogram of microRNA-21 as a biomarker in lung carcinoma identification

sensitivity, specificity, DOR, AUC, and post-test probability were demonstrated in Table 2.

Publication bias

The publication bias was evaluated by Deek's funnel plot. The plot was obviously asymmetry, which indicated significant publications bias ($p < 0.05$) (Figure 6).

DISCUSSION

At present, the diagnosis of lung cancer relies on chest imaging and cell morphology examination. However, because of poor sensitivity, the aforementioned diagnostic methods cannot identify early stage lung carcinoma.[28,29] Most of the lung cancer cases are diagnosed at middle or advanced stages with poor prognosis and a low long-term survival rate.[30,31] Therefore, it is urgent to find more sensitive and non-invasive biomarkers to improve the lung cancer diagnosis.

TABLE 2 Sub-group analysis according detection samples

Diagnostic performance	Sputum	Serum
Sensitivity	0.77 (0.70–0.82)	0.72 (95% CI, 0.64–0.78)
Specificity	0.77 (0.71–0.83)	0.83 (95% CI, 0.74–0.88)
DOR	11.35 (6.97–18.50)	9.44 (5.04–15.01)
AUC	0.84 (0.80–0.83)	0.84 (0.80–0.87)
Post_Prob_Pos (%)	77	80
Post_Prob_Neg (%)	23	25

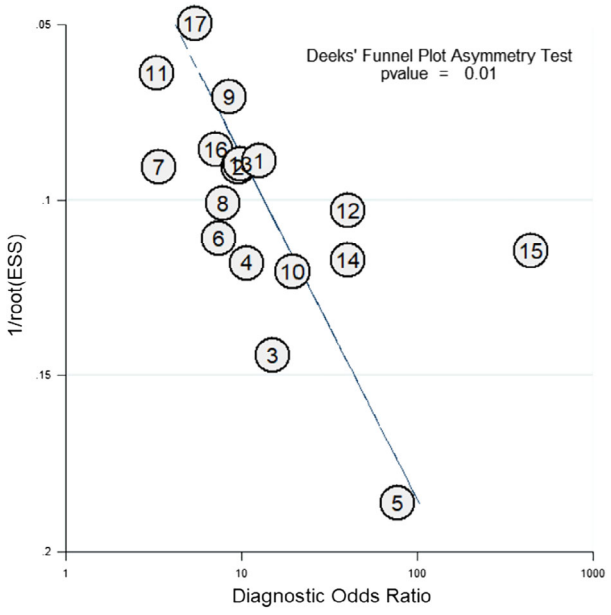

FIGURE 6 Deek's funnel plot for publication bias evaluation

MicroRNA is a kind of non-coding RNA with a length of <25 nucleotides, which is involved in the occurrence and development of a variety of diseases such as cancer,[32,33] cardiovascular disease,[34] infection,[35,36] rheumatic immunity, etc. Studies have confirmed that microRNA can stably exist in body fluids, which provides an opportunity for cancer diagnosis with non-invasive methods. MicroRNA-21 was widely investigated in cancers and most of the studies demonstrated that microRNA-21 was upregulated in cancer tissue and corresponding serum.[37,38] Therefore, the serum level of microRNA-21 may provide useful information for lung cancer identification. However, because of the small sample size, different lung cancer stages, and different histology type, the conclusion was not inconclusive for microRNA-21 as a biomarker in detection lung cancer.

In the present work, we identified all the open published diagnostic studies relevant to microRNA-21 as a biomarker in detection lung cancer and made data combination. Seventeen diagnostic studies were made data pooling and the results demonstrated that the pooled diagnostic sensitivity, specificity were 0.73 (95% CI, 0.67–0.78) and 0.81 (95% CI, 0.75–0.85), respectively, with the diagnostic AUC of 0.84 (95% CI, 0.80–0.87). Given a pre-test probability of 50%, the post-test positive probability and post-test negative probability were 79% and 25%. The pooled results indicated that microRNA-21 in serum or sputum was a promising biomarker for lung cancer identification with relative high diagnostic sensitivity and specificity. The post-test positive probability was elevated for lung cancer and post-test negative probability was decreased for healthy or non-lung cancer subjects.

There are several limitations that may affect the stability of the conclusion: (i) there are obviously statistical heterogeneity in combination the sensitivity, specificity, and DOR; (ii) publication bias existed and may decrease the reliability of results; and (iii) language restriction may lead to studies being omitted.

In conclusion, 17 studies were included and made data combination. Pooled results indicated detection microRNA-21 in sputum or serum can provided useful information for lung cancer identification with high diagnostic performance. However, because of the aforementioned limitations, the conclusion should be further validated by big sample size, well-designed, prospective diagnostic studies.

ORCID

Haiyan Wang https://orcid.org/0000-0002-5693-5046

REFERENCES

1. Siegel RL, Miller KD, Fuchs HE, Jemal A. Cancer statistics, 2021. CA Cancer J Clin. 2021;71:7–33.
2. Sung H, Ferlay J, Siegel RL, Laversanne M, Soerjomataram I, Jemal A, et al. Global cancer statistics 2020: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin. 2021;71:209–49.
3. Zheng W, Zhang H, Shen C, Zhang S, Wang D, Li W, et al. Trend analysis of lung cancer mortality and years of life lost (YLL) rate from

1999 to 2016 in Tianjin, China: does the lung cancer burden in rural areas exceed that of urban areas. Thorac Cancer. 2020;11:867–74.
4. Schegoleva AA, Khozyainova AA, Fedorov AA, Gerashchenko TS, Rodionov EO, Topolnitsky EB, et al. Prognosis of different types of non-small cell lung cancer progression: current state and perspectives. Cell Physiol Biochem. 2021;55:29–48.
5. Tailor TD, Patz EF Jr. Lung cancer screening with chest CT: efficacy confirmed. Radiol Imaging Cancer. 2020;2:e204015.
6. Novellis P, Cominesi SR, Rossetti F, Mondoni M, Gregorc V, Veronesi G. Lung cancer screening: who pays? Who receives? The European perspectives. Transl Lung Cancer Res. 2021;10:2395–406.
7. Smolarz M, Widlak P. Serum Exosomes and their miRNA load-a potential biomarker of lung cancer. Cancers (Basel). 2021;13(6):1–20.
8. Baron AT, Wilken JA, Haggstrom DE, Goodrich ST, Maihle NJ. Clinical implementation of soluble EGFR (sEGFR) as a theragnostic serum biomarker of breast, lung and ovarian cancer. IDrugs. 2009;12:302–8.
9. Tao S, Ju X, Zhou H, Zeng Q. Circulating microRNA-145 as a diagnostic biomarker for non-small-cell lung cancer: a systemic review and meta-analysis. Int J Biol Markers. 2020;35:51–60.
10. Zhao W, Zhao JJ, Zhang L, Xu QF, Zhao YM, Shi XY, et al. Serum miR-21 level: a potential diagnostic and prognostic biomarker for non-small cell lung cancer. Int J Clin Exp Med. 2015;8:14759–63.
11. Yang JS, Li BJ, Lu HW, Chen Y, Lu C, Zhu RX, et al. Serum miR-152, miR-148a, miR-148b, and miR-21 as novel biomarkers in non-small cell lung cancer screening. Tumour Biol. 2015;36:3035–42.
12. Whiting P, Rutjes AW, Reitsma JB, Bossuyt PM, Kleijnen J. The development of QUADAS: a tool for the quality assessment of studies of diagnostic accuracy included in systematic reviews. BMC Med Res Methodol. 2003;3:25.
13. Su J, Anjuman N, Guarnera MA, Zhang H, Stass SA, Jiang F. Analysis of lung flute-collected sputum for lung cancer diagnosis. Biomark Insights. 2015;10:55–61.
14. Xing L, Su J, Guarnera MA, Zhang H, Cai L, Zhou R, et al. Sputum microRNA biomarkers for identifying lung cancer in indeterminate solitary pulmonary nodules. Clin Cancer Res. 2015;21:484–9.
15. Yang XQ, Zhang YH, Sun B, Deng P, Fang J, Shen JH, et al. Diagnostic value of the detection of MicroRNAs in sputum of patients with non-small cell lung cancer. J Clin Pulmon Med. 2013;18:226–9.
16. Yu L, Todd NW, Xing L, Xie Y, Zhang H, Liu Z, et al. Early detection of lung adenocarcinoma in sputum by a panel of microRNA markers. Int J Cancer. 2010;127:2870–8.
17. Li Y, Li W, Ouyang Q, Hu S, Tang J. Detection of lung cancer with blood microRNA-21 expression levels in Chinese population. Oncol Lett. 2011;2:991–4.
18. Juan W, Wen G, Cheng-Jun Z, Yi-Qian L, Zhu M, Ting C, et al. Identification of plasma microRNA-21 as a biomarker for early detection and chemosensitivity of non-small cell lung cancer. Chin J Cancer. 2011;30:407–14.
19. Tang D, Shen Y, Wang M, Yang R, Wang Z, Sui A, et al. Identification of plasma microRNAs as novel noninvasive biomarkers for early detection of lung cancer. Eur J Cancer Prev. 2013;22:540–8.
20. Wei J, Liu LK, Gao W, Zhu CJ, Liu YQ, Cheng T, et al. Reduction of plasma MicroRNA-21 is associated with chemotherapeutic response in patients with non-small cell lung cancer. Chin J Cancer Res. 2011; 23:123–8.
21. Chen GH, Yan H, Sun CY, Zhao J. Expression and significance of microRNA-21 in the cancer tissue and plasma of patients with non-small cell lung cancer. Chinese J Clin Lab Sci. 2014;32:652–5.
22. Wang B, Zhang Q. The expression and clinical significance of circulating microRNA-21 in serum of five solid tumors. J Cancer Res Clin Oncol. 2012;138:1659–66.
23. Shen J, Liu Z, Todd NW, Zhang H, Liao J, Yu L, et al. Diagnosis of lung cancer in individuals with solitary pulmonary nodules by plasma microRNA biomarkers. BMC Cancer. 2011;11:374.
24. Abd-El-Fattah AA, Sadik NA, Shaker OG, Aboulftouh ML. Differential microRNAs expression in serum of patients with lung cancer, pulmonary tuberculosis, and pneumonia. Cell Biochem Biophys. 2013;67: 875–84.
25. Le HB, Zhu WY, Chen DD, He JY, Huang YY, Liu XG, et al. Evaluation of dynamic change of serum miR-21 and miR-24 in pre- and post-operative lung carcinoma patients. Med Oncol. 2012;29: 3190–7.
26. Wang WD, Pei ZH, Xi XX, Cui WL, Zhang T, Chang XT. Excavate target genes of miR-21 in lung cancer patients and its expression diagnostic value in serum. Chinese J Immunol. 2021;37:1118–25.
27. Li SR, Liu Y, Wang ZM, Song MZ, Jiao HJ. Diagnostic value of detecting serum miR-483-5p, miR-21, and miR-25 in patients with non-small-cell lung cancer. Shandong Med J. 2019;59: 19–22.
28. Thandra KC, Barsouk A, Saginala K, Aluru JS, Barsouk A. Epidemiology of lung cancer. Contemp Oncol (Pozn). 2021;25:45–52.
29. Casal-Mouriño A, Ruano-Ravina A, Lorenzo-González M, Rodríguez-Martínez Á, Giraldo-Osorio A, Varela-Lema L, et al. Epidemiology of stage III lung cancer: frequency, diagnostic characteristics, and survival. Transl Lung Cancer Res. 2021;10:506–18.
30. Lin HT, Liu FC, Wu CY, Kuo CF, Lan WC, Yu HP. Epidemiology and survival outcomes of lung cancer: a population-based study. Biomed Res Int. 2019;2019:8148156.
31. Kastner J, Hossain R, White CS. Epidemiology of lung cancer. Semin Roentgenol. 2020;55:23–40.
32. Li H, Huhe M, Lou J. MicroRNA-103a-3p promotes cell proliferation and invasion in non-small-cell lung cancer cells through Akt pathway by targeting PTEN. Biomed Res Int. 2021;2021:7590976.
33. Ding L, Tian W, Zhang H, Li W, Ji C, Wang Y, et al. MicroRNA-486-5p suppresses lung cancer via downregulating mTOR signaling in vitro and in vivo. Front Oncol. 2021;11:655236.
34. Song Z, Gao R, Yan B. Potential roles of microRNA-1 and microRNA-133 in cardiovascular disease. Rev Cardiovasc Med. 2020; 21:57–64.
35. Swaminathan S, Kelleher AD. MicroRNA modulation of key targets associated with T cell exhaustion in HIV-1 infection. Curr Opin HIV AIDS. 2014;9:464–71.
36. Kałużna EM. MicroRNA-155 and microRNA-196b: promising biomarkers in hepatitis C virus infection. Rev Med Virol. 2014;24: 169–85.
37. Li G, Wang Q, Li Z, Shen Y. Serum miR-21 and miR-210 as promising non-invasive biomarkers for the diagnosis and prognosis of colorectal cancer. Rev Esp Enferm Dig. 2020;112:832–7.
38. Liu M, Wang W, Chen H, Lu Y, Yuan D, Deng Y, et al. miR-9, miR-21, miR-27b, and miR-34a expression in HPV16/58/52-infected cervical cancer. Biomed Res Int. 2020;2020:2474235.

How to cite this article: Wang H, Xu J, Ding L. MicroRNA-21 was a promising biomarker for lung carcinoma diagnosis: An update meta-analysis. Thorac Cancer. 2022;13:316–21. https://doi.org/10.1111/1759-7714.14242