INTRODUCTION

Adenocarcinoma in situ (AIS) was formerly classified as a subtype of lung adenocarcinoma that is usually considered pre-invasive. According to the definition of the International Association for the Study of Lung Cancer (IASLC, 2011 edition), the American Thoracic Society (ATS) classification in 2011, and the European Respiratory Society (ERS) classification in 2011, AIS is a localized adenocarcinoma.
lesion of less than or equal to 3 cm with pure lepidic growth pattern.\(^1\) In WHO classification of thoracic tumors (5th edition) 2021, AIS and atypical adenomatous hyperplasia (AAH) were adjusted from preinvasive lesions to precursor glandular lesions.\(^2\)

Previous studies revealed that AIS shows distinctive clinicopathological features, genetic signatures, microenvironmental conditions, and DNA methylation levels and destined to be different from AAH, minimally invasive adenocarcinoma (MIA), and invasive adenocarcinoma (IA).\(^3\)–\(^6\) More importantly, patients with AIS demonstrated superior prognosis after curative resection, with 5-year disease-specific survival (DFS) and overall survival (OS) of nearly 100%.\(^7\)–\(^8\) Because of its noninvasive nature, AIS has been proposed to be an appropriate candidate for limited resection.\(^9\) Therefore, preoperative identification of AIS is of great concern. However, a reliable method is scarce. Positron emission tomography/computed tomography (PET/CT) showed limited sensitivity in identifying AIS.\(^10\) By comparison, perioperative frozen section diagnosis typically has high specificity, it shows limited sensitivity and usually overestimates the tumor invasiveness.\(^11\) At the right time, circulating tumor cell (CTC) detection technology may provide insights into tumor invasiveness and help identify AIS prior to surgery. Over the past decade, different techniques have been developed to isolate, detect, and quantitate CTCs, and the clinical significance of CTC has been well studied.\(^12\) The Cytoploraare® CTC enrichment and detection kit enumerates folate receptor-positive circulating tumor cells (FR+CTCs) using negative enrichment and ligand-targeted polymerase chain reaction (PCR) techniques. The FR+CTC level has proven effective in determining the malignancy of pulmonary nodules.\(^13\)–\(^17\) Furthermore, FR+CTCs were found to be present in patients with preinvasive pulmonary diseases.\(^18\)–\(^19\) Thus, in the present study, we aimed to investigate the performance of FR+CTC as a noninvasive biomarker to preoperatively identify lung precursor lesions.

### 2 | MATERIAL AND METHODS

#### 2.1 | Patient recruitment

The observational study conducted in Shanghai Pulmonary Hospital. A total of 380 patients with indeterminate pulmonary nodules were recruited from July 2018 to June 2019. Inclusion criteria were as follows: (1) patients with indeterminate pulmonary nodules of less than or equal to 30 mm according to CT scan; (2) expected to undergo surgical resection; and (3) signed informed consent and agreed to provide a preoperative peripheral blood sample for FR+CTC analysis. Exclusion criteria were as follows: (1) uncertain pathological diagnosis of surgically resected specimens; (2) insufficient or abnormal morphology preoperative blood sample for FR+CTC analysis; (3) the interval between sampling and surgery was more than 7 days; and (4) prior cancer history. All the procedures were conducted in accordance with the Helsinki Declaration and the study was approved by the institutional ethics committee of the Shanghai Pulmonary Hospital. Written informed consent was obtained from all participants upon enrollment.

#### 2.2 | Data collection

Data were collected from the clinician records through the standard case report form (CRF), including demographic characteristics, clinical symptoms, surgical methods, and CT image information.

#### 2.3 | CT scan

All participants received a chest CT scan (Revolution CT, GE Healthcare,) before inclusion into this study. CT image characteristics including lesion size, type, and location were observed and agreed upon by at least two experienced radiologists. A multidisciplinary team then evaluated the malignancy of the identified nodules. Patients with nodules likely to be malignant subsequently undergo surgical resection.

#### 2.4 | Pathological assessment

Resected tumors specimens were pathologically assessed by at least two experienced pathologists to determine the tumor invasiveness according to the IASLC/ATS/ERS classification of lung adenocarcinoma.\(^2\) In brief, AAH was defined as a localized proliferation of less than or equal to 5 mm; AIS was defined as a localized lesion of less than or equal to 30 mm with a pure lepidic pattern; MIA was defined as a solitary adenocarcinoma of less than or equal to 30 mm with a predominantly lepidic pattern and less than or equal to 5 mm invasion; and IA was defined as an adenocarcinoma lesion with other/mixed subtype and invasion.

#### 2.5 | FR + CTC enumeration

Upon enrollment, 3 ml of peripheral venous blood was collected from each participant. FR+CTC level was analyzed by using the Cytoploraare® kit (Genosaber Biotech,) according to the manufacturer’s protocol. Detailed procedures were as previously described.\(^15\) Briefly, the first step negatively enriches CTCs through lysis of red blood cells and depletion of white blood cells using antibody-coated magnetic beads. The second step labels FR+CTCs with a folate receptor alpha-targeting detection probe containing a specific oligonucleotide sequence and detects its signal using quantitative polymerase chain reaction (qPCR). A serial of diluted oligonucleotide samples corresponding to 2–2×10\(^5\) FR+CTCs served as standards to calibrate the FR+CTC level. The unit used for FR+CTC quantitation was defined as folate receptor unit (FU)/3 ml of peripheral blood.
2.6 | Statistical analysis

Continuous variables were compared using Mann-Whitney t-test. Categorical outcomes were compared using Fisher’s exact test and chi-square test. Receiver operating characteristic (ROC) curve was used to analyze the diagnostic performance of FR+CTC. Youden index was calculated to determine the optimal cutoff values. Multivariate analyses assessing predictors of precursor lesions (adjusted for gender, age, total number of pulmonary nodules, type of nodules, maximum tumor diameter [MTD], and location) was performed using logistic regression. Statistical analysis was performed by using Prism 8 (GraphPad Software,) and R 4.0.0 (R Foundation for Statistical Computing.).

3 | RESULTS

3.1 | Baseline characteristics

A total of 380 patients were enrolled in this study with indeterminate pulmonary nodules. 26 patients were excluded: 2 did not undergo surgery for physical reasons, 15 for blood sample reason (5 due to lack of operation time information, it is impossible to determine whether the blood was collected before surgery, 3 due to postoperative blood, 3 due to the interval between sampling and surgery was more than 7 days and 4 blood clots), 8 had uncertain pathological diagnosis, and 1 had prior cancer history. 354 patients underwent surgery and CTC analysis and were included in the final analysis (Figure S1). Of these 354 patients, 57 had benign lung disease, 133 were confirmed to precursor lesions, 136 were adenocarcinoma, and the remaining 28 had unspecified pathological types. Patient demographics and detailed clinical characteristics are shown in Table 1.

3.2 | FR + CTC level in determining precursor lesions

The median FR+CTC level of the precursor lesions group was higher than that of the benign group (10.60 vs. 7.52 FU/3 ml) (Table S1), the difference was statistically significant (p < 0.001) (Figure 1A). FR+CTC can effectively differentiate precursor from benign lesions (cutoff value: 9.22 FU/3 ml; sensitivity: 69.17%; specificity: 82.46%) (Figure 2A, Table 2). The median FR+CTC levels of the precursor lesions and the malignant lesions group were 10.60 FU/3 ml and 10.44 FU/3 ml, respectively (Table S1). The difference was not statistically significant (p = 0.71) (Figure 1A). FR+CTC cannot differentiate precursor lesions from malignant tumors (cutoff value: 11.61 FU/3 ml; sensitivity: 63.97%; specificity: 44.36%) (Figure 2B, Table 2). The median FR+CTC levels of the single pulmonary lesion and the multiple pulmonary lesions group were 10.15 FU/3 ml and 9.33 FU/3 ml, respectively (Figure 1B). And the median FR+CTC levels of the benign, the precursor lesion, and malignant tumors group in single pulmonary lesion patients were 7.37 FU/3 ml, 10.60 FU/3 ml, and 10.55 FU/3 ml (Table S1, Figure 1C), The difference between benign and precursor was statistically significant (p = 0.004), while the difference between precursor and malignant was not statistically significant (p = 0.57). FR+CTC can effectively differentiate precursor lesions from benign lesions (cutoff value: 9.03 FU/3 ml; sensitivity: 75.20%; specificity: 83.00%) (Figure 2C, Table 2) and FR+CTC cannot differentiate malignant tumors from precursor lesions (cutoff value: 13.13 FU/3 ml; sensitivity: 76.32%; specificity: 31.86%) (Figure 2D, Table 2). The median FR+CTC levels of the benign, the precursor glandular lesion, and malignant tumors group in multiple pulmonary lesion patients were 7.88 FU/3 ml, 10.44 FU/3 ml, and 9.39 FU/3 ml (Figure 1D). For patients with multiple pulmonary lesions, FR+CTC neither differentiate precursor lesions from benign lesions (cutoff value: 12.64 FU/3 ml; sensitivity: 88.89%; specificity: 45.00%) (Figure 2E, Table 2) nor precursor lesions from malignant tumors (cutoff value: 12.38 FU/3 ml; sensitivity: 72.73%; specificity: 45.00%) (Figure 2F, Table 2).

| TABLE 1 | Patients’ demography of 354 eligible patients |
|----------|-----------------------------------------------|
| **Number of patients** | **N (%)** |
| **Gender** | |
| Male | 135 (38.1) |
| Female | 219 (61.9) |
| Age (SD), years | 50.7 (11.5) |
| **Total number of pulmonary nodules identified** | |
| 1 | 277 (78.2) |
| >1 | 76 (21.5) |
| **Pathological subtype** | |
| Benign | 57 (16.1) |
| Precursor lesions | 133 (37.6) |
| Malignant lesions | 136 (38.4) |
| Complex pathology | 28 (7.9) |
| **Type of nodules** | |
| Pure GGN | 276 (78.0) |
| Pure-solid GGN | 20 (5.6) |
| Solid nodule | 36 (10.2) |
| **Location** | |
| Upper | 180 (50.9) |
| Middle | 32 (9.0) |
| Lower | 107 (30.2) |
| **Surgery** | |
| Wedge resection | 123 (34.7) |
| Segmentectomy | 129 (36.4) |
| Lobectomy | 64 (18.1) |
| Complex | 38 (10.7) |

*a* one case no data.

*b* both precursor and malignant lesions are present in multiple lesions cases.

*c* contains at least two of the above three resections.
3.3 Univariate and multivariate analysis for precursor lesions

To identify independent factors that influence the differentiation between malignant lesions and precursor lesions, the univariate analysis indicated that age, total number of pulmonary nodules, MTD, location (lower vs upper) were statistically significant (all \( p < 0.05 \)), and for multivariate analysis, multiple pulmonary lesion was 3.104 times more likely to malignant lesions than the single multiple pulmonary lesions (AOR: 3.104, 95% CI: 1.525, 6.316), the MTD ≥1 cm was more likely to be malignant lesions (AOR: 3.148, 95% CI: 1.722, 5.754), lower lobe were more likely to malignant lesions than upper lobe (AOR: 2.098, 95% CI: 1.132, 3.888) (Table 3).

4 DISCUSSION

Based on accurate preoperative diagnosis, physicians can help patients in deciding whether the patient should undergo surgical treatment and the extent of the surgery. However, there is a lack of effective methods to differentiate precursor lesions from benign pulmonary diseases or other pathological subtypes of lung cancer currently. Due to their good prognosis, AIS are considered low-grade malignant tumors. However, since most patients with lung cancer exhibit middle or late stage disease at the initial visit, early detection, early diagnosis, and correct staging play an extremely important role in improving their survival rate. Therefore, noninvasive, preoperative differentiation of precursor lesions from benign or malignant is essential in guiding the clinical management of these patients.
FIGURE 2  ROC curves. (A) The performance of FR + CTC in differentiating precursor glandular lesions from benign; (B) The performance of FR + CTC in differentiating precursor glandular lesions from malignant; (C) The performance of FR + CTC in differentiating precursor glandular lesions from benign in single pulmonary lesions; (D) The performance of FR + CTC in differentiating precursor glandular lesions from malignant in single pulmonary lesions; (E) The performance of FR + CTC in differentiating precursor glandular lesions from benign in multiple pulmonary lesions; (F) The performance of FR + CTC in differentiating precursor glandular lesions from malignant in multiple pulmonary lesions.
TABLE 2  Results of ROC analysis

|                       | Area under curve | 95% confidence interval | p value | Cutoff | Sensitivity | Specificity |
|-----------------------|------------------|-------------------------|---------|--------|-------------|-------------|
| Benign vs precursor   | 0.8069           | 0.7441–0.8697           | <0.0001 | 9.220  | 0.6917      | 0.8246      |
| Precursor vs malignant| 0.5130           | 0.4435–0.5824           | 0.7145  | 11.610 | 0.6397      | 0.4436      |
| Benign vs precursor (single lesions) | 0.8417 | 0.7800–0.9035 | <0.0001 | 9.025  | 0.7520      | 0.8300      |
| Precursor vs malignant (single lesions) | 0.5065 | 0.4308–0.5822 | 0.8667  | 13.13  | 0.7632      | 0.3186      |
| Benign vs precursor (multiple lesions) | 0.6472 | 0.4385–0.8559 | 0.1101  | 12.64  | 0.8889      | 0.45        |
| Precursor vs malignant (multiple lesions) | 0.5364 | 0.3563–0.7164 | 0.6922  | 12.375 | 0.7273      | 0.45        |

TABLE 3  Univariate and multivariate analysis for precursor glandular lesions

| Factors                          | Univariate analysis |                          | Multivariate analysis |                          |
|----------------------------------|---------------------|--------------------------|-----------------------|--------------------------|
|                                  | OR, 95% CI          | p value                  | OR, 95% CI            | p value                  |
| FR + CTC level (≥11.615)         | 0.69(0.43–1.11)     | 0.12                     | 0.7276(0.416–1.273)   | 0.2653                   |
| Gender (Female)                  | 0.82(0.5–1.33)      | 0.42                     | –                     | –                        |
| Age (≥60)                        | 2.36(1.36–4.08)     | <0.01                    | 1.840(0.972–3.516)    | 0.0610                   |
| Total number of nodules (>1)     | 2.33(1.3–4.18)      | <0.01                    | 3.104(1.525–6.316)    | 0.0018                   |
| MTD (≥1 cm)                      | 3.76(2.24–6.32)     | <0.01                    | 3.148(1.722–5.754)    | <0.01                    |
| Solid nodule                     | Reference           |                          |                       |                          |
| Pure solid GGN                   | 3.139,272(0–Inf)    | 0.98                     | 6.934,000(0–Inf)      | 0.9882                   |
| Pure GGN                         | 0.23(0.03–1.98)     | 0.18                     | 0.742(0.029–2.599)    | 0.2595                   |
| Location                         |                      |                          |                       |                          |
| Upper                            | Reference           |                          |                       |                          |
| Middle                           | 0.77(0.34–1.75)     | 0.53                     | 0.836(0.325–2.148)    | 0.710                    |
| Lower                            | 1.82(1.05–3.17)     | 0.03                     | 2.098(1.132–3.888)    | 0.0186                   |

MTD: Maximum tumor diameter.

In the present study, median FR + CTC of the malignant group was 10.44 FU/3 ml, which was significantly higher than 7.52 FU/3 ml of the benign group. We confirm that whether the multiple nodules have the same infiltration property or not, in the multiple lesions subgroup, median FR + CTC of the malignant group was 9.39 FU/3 ml, while that of the benign group was 7.88 FU/3 ml, the result was similar. However, due to the sample size being too small and the lack of more genotypes and other data, it is impossible to determine whether they are multiple primary or intra pulmonary metastasis. The differences in their biological characteristics lead to complex effects on CTC level, but the overall judgment of benign and malignant by CTC is not affected (in single lesion group). The FR + CTC levels between benign and precursor lesions in patients with single pulmonary lesion is significantly different (p < 0.001); while FR + CTC levels between precursor lesions and malignant lesions is not statistically different (p > 0.05). In this group of patients, benign and precursor lesions can be distinguished, while precursor lesions and malignant lesions was not well differentiated, especially in the multiple lesions group, the possible reason is small sample size, and we need further studies. Recently, Hu et al. reported that early spread of colorectal cancer usually occurs at a very early stage when the basement membrane is not breached and the cancer is undetectable in clinical (usually less than 0.01 cm³), although the mechanism is still unclear, it is likely to be detected by CTC.22

Since the report from the North American Lung Cancer Study Group of a three-fold increase in local recurrence rate and a decrease in overall survival following limited resection, lobectomy has been the standard of care for early-stage nonsmall cell lung cancer (NSCLC) in the past two decades,23 but non-IA patients may be candidates for limited surgical resection.21 Nevertheless, recent studies have shown that sublobar resection could be acceptable for patients with noninvasive adenocarcinomas.24 For patients with stage IA NSCLC, segmentectomy has showed a comparable survival and a better preservation of lung function.24,25 Indeed, the NCCN guideline suggests that sublobar resection is appropriate in patients with peripheral nodule ≤ 2 cm, and pure adenocarcinoma in situ (AIS) histology. Therefore, development of a noninvasive and sensitive strategy for predicting the invasiveness of lung nodules, particularly in NSCLC, before selecting eligible patients for limited lung resection will significantly benefit patients with noninvasive lung carcinoma, particularly early-stage, undefined small-sized SPNs, by avoiding major lung resections. In the long run, a reliable differentiating strategy for SPNs will significantly contribute to the reduction of lung cancer-associated mortality by guiding adequate and timely treatment and avoiding overtreatment of noninvasive SPNs. Our result of FR + CTC level cannot in favor of distinguishing precursor lesions and malignant lesions, and because of that we explored the risk factors.
of malignant, the results showed that MTD >10mm were independent risk factors for malignant lesions, it is consistent with previous study, however, location (lower lobe) is inconsistent. Total number of nodules > 1 was showed to be an independent risk factor for malignant lesions, and the age was almost an independent risk factor in our study (p = 0.061). Zhou et al. demonstrated that in small-sized lung adenocarcinoma, FR+CTC level was significantly lower in non-invasive adenocarcinomas than that in invasive adenocarcinomas. And multivariate analysis revealed that smaller tumor size and lower FR+CTC level are significant independent differentiators of non-invasive cancer from invasive cancer.18 With the excessive care in lung cancer screening, an efficacious biomarker for assisting LDCT to determine the malignancy and tumor invasiveness of lung nodules is urgently warranted.26 Increasing effort is being made to leverage FR as over expression as a diagnostic marker and/or to enable more precise cancer surgery. Although lobectomy is the standard surgical procedure for lung cancer, a limited resection is suggested for some carefully selected patients, in particular, patients with small-sized lesions. A previous report suggested that noninvasive adenocarcinomas are potential candidates for limited resection and have excellent prognosis.27 In practice, it is sometimes difficult to eliminate tumor invasion through intraoperative morphological diagnosis based on frozen sections.28 Lee et al. reported that lesion border and lesion margin on CT imaging were that were routinely used in clinical settings showed only moderate interobserver agreement,29 thereby calling for a more objective and robust parameter other than CT imaging to differentiate noninvasive adenocarcinomas from invasive ones. Preoperative CTC detection combined with the maximum tumor diameter can effectively evaluate the subnodular infiltrates and the subnodular tumoticle like IPA.18

Several limitations of the current study should be mentioned, firstly, the present study was a retrospective study although the CTC was collected prospectively, it resulted in the loss of crucial information and information bias, especially imaging morphological information, furthermore, we were unable to screen predictors based on morphology and adjust for some potential confounders. Secondly, the small number of included patients, and the difference in the number between the benign and precursor or malignant groups, increase uncertainty.

5 | CONCLUSIONS
FR + CTC can be identified in precursor glandular lesions and utilized to differentiate from benign pulmonary diseases. Total number of pulmonary nodules, MTD, location (lower vs upper) were independent risk factors for malignancy. FR + CTC is expected to be an alternative biomarker for preoperative surgical decision making. However, further studies with large samples are needed for confirmation.

ACKNOWLEDGMENTS
None

FUNDING INFORMATION
The study was funded by the Programs of Shanghai Pulmonary Hospital (no. fxxr1904).

CONFLICT OF INTEREST
The author has no conflicts of interest to declare.

DATA AVAILABILITY STATEMENT
The data that support the findings of this study are available from the corresponding author upon reasonable request.

ORCID
Xiaogang Zhao  https://orcid.org/0000-0001-6071-0424

REFERENCES
1. Travis WD, Brambilla E, Noguchi M, et al. International association for the study of lung cancer/american thoracic society/european respiratory society international multidisciplinary classification of lung adenocarcinoma. J Thorac Oncol. 2011;6:244-285.
2. WHO classification of tumors Editorial Board. Thoracic Tumours: WHO classification of tumours. 5th ed. 2021. WHO Press.
3. Ishida H, Shimizu Y, Sakaguchi H, et al. Distinctive clinicopathological features of adenocarcinoma in situ and minimally invasive adenocarcinoma of the lung: a retrospective study. Lung Cancer. 2019;129:16-21.
4. Li D, Yang W, Zhang Y, et al. Genomic analyses based on pulmonary adenocarcinoma in situ reveal early lung cancer signature. BMC Med Genomics. 2018;11:106.
5. Naito M, Aokage K, Saruwatari K, et al. Microenvironmental changes in the progression from adenocarcinoma in situ to minimally invasive adenocarcinoma and invasive lepidic predominant adenocarcinoma of the lung. Lung Cancer. 2016;100:53-62.
6. Selamat SA, Galler JS, Joshi AD, et al. DNA methylation changes in atypical adenomatous hyperplasia, adenocarcinoma in situ, and lung adenocarcinoma. PLoS One. 2011;6:e21443.
7. Behera M, Owonikoko TK, Gal AA, et al. Lung adenocarcinoma staging using the 2011 IASLC/ATS/ERS classification: a pooled analysis of adenocarcinoma in situ and minimally invasive adenocarcinoma. Clin Lung Cancer. 2016;17:e57-e64.
8. Boland JM, Froemmig AT, Wampfler JA, Maldonado F, Peikert T, Hyland C, de Andrade M, Aubry MC, Yang P, Yi ES Adenocarcinoma in situ, minimally invasive adenocarcinoma, and invasive pulmonary adenocarcinoma--analysis of interobserver agreement, survival, radiographic characteristics, and gross pathology in 296 nodules. Hum Pathol 2016;51:41-50, Adenocarcinoma in situ, minimally invasive adenocarcinoma, and invasive pulmonary adenocarcinoma--analysis of interobserver agreement, survival, radiographic characteristics, and gross pathology in 296 nodules.
9. Wang R, Hu H, Pan Y, et al. RET fusions define a unique molecular and clinicopathologic subtype of non-small-cell lung cancer. J Clin Oncol. 2012;30:4352-4359.
10. Hattori A, Suzuki K, Matsunaga T, et al. Tumour standardized uptake value on positron emission tomography is a novel predictor of adenocarcinoma in situ for c-Stage IA lung cancer patients with a part-solid nodule on thin-section computed tomography scan. Interact Cardiovasc Thorac Surg. 2014;18:329-334.
11. He P, Yao G, Guan Y, Lin Y, He J. Diagnosis of lung adenocarcinoma in situ and minimally invasive adenocarcinoma from intraoperative frozen sections: an analysis of 136 cases. J Clin Pathol. 2016;69:1076-1080.
12. Sharma S, Zhuang R, Long M, et al. Circulating tumor cell isolation, culture, and downstream molecular analysis. Biotechnol Adv. 2018;36:1063-1078.
13. Xue Y, Cong W, Xie S, Shu J, Feng G, Gao H. Folate-receptor-positive circulating tumor cells as an efficacious biomarker for the diagnosis of small pulmonary nodules. J Cancer Res Ther. 2018;14:1620-1626.
14. Wang L, Wu C, Qiao L, et al. Clinical significance of folate receptor-positive circulating tumor cells detected by ligand-targeted polymerase chain reaction in lung cancer. J Cancer. 2017;8:104-110.
15. Chen X, Zhou F, Li X, et al. Folate-receptor-positive circulating tumor cell detected by LT-PCR-based method as a diagnostic biomarker for non-small-cell lung cancer. J Thorac Oncol. 2015;10:1163-1171.
16. Yu Y, Chen Z, Dong J, et al. Folate receptor-positive circulating tumor cells as a novel diagnostic biomarker in non-small cell lung cancer. Transl Oncol. 2013;6:697-702.
17. Lou J, Ben S, Yang G, et al. Quantification of rare circulating tumor cells in non-small cell lung cancer by ligand-targeted PCR. PLoS One. 2013;8:e80458.
18. Zhou Q, Geng Q, Wang L, et al. Value of folate receptor-positive circulating tumour cells in the clinical management of indeterminate lung nodules: A non-invasive biomarker for predicting malignancy and tumour invasiveness. EBioMedicine. 2019;41:236-243.
19. Ding C, Zhou X, Xu C, et al. Circulating tumor cell levels and carcinoembryonic antigen: an improved diagnostic method for lung adenocarcinoma. Thorac Cancer. 2018;9:1413-1420.
20. Song Z, Zhu H, Guo Z, Wu W, Sun W, Zhang Y. Prognostic value of the IASLC/ATS/ERS classification in stage I lung adenocarcinoma patients—Based on a hospital study in China. Eur J Surg Oncol. 2013;39:1262-1268.
21. Fan L, Fang M, Li Z, et al. Radiomics signature: a biomarker for the preoperative discrimination of lung invasive adenocarcinoma manifesting as a ground-glass nodule. Eur Radiol. 2019;29:889-897.
22. Hu Z, Ding J, Ma Z, et al. Quantitative evidence for early metastatic seeding in colorectal cancer. Nat Genet. 2019;51:1113-1122.
23. Ginsberg RJ, Rubinstein LV. Randomized trial of lobectomy versus limited resection for T1 N0 non-small cell lung cancer. Lung Cancer Study Group. Ann Thorac Surg. 1995;60:615-622. discussion 622-613.
24. Altorki NK, Yip R, Hanaoka T, et al. Sublobar resection is equivalent to lobectomy for clinical stage 1A lung cancer in solid nodules. J Thorac Cardiovasc Surg. 2014;147:754-762. Discussion 762-754.
25. Harada H, Okada M, Sakamoto T, Matsuoka H, Tsubota N. Functional advantage after radical segmentectomy versus lobectomy for lung cancer. Ann Thorac Surg. 2005;80:2041-2045.
26. Ten Haaf K, van der Aalst CM, de Koning HJ. Clinically detected non-aggressive lung cancers: implications for overdiagnosis and overtreatment in lung cancer screening. Thorax. 2018;73:407-408.
27. Yoshizawa A, Motoi N, Riely GJ, et al. Impact of proposed IASLC/ATS/ERS classification of lung adenocarcinoma: prognostic subgroups and implications for further revision of staging based on analysis of 514 stage I cases. Mod Pathol. 2011;24:653-664.
28. Walts AE, Marchevsky AM. Root cause analysis of problems in the frozen section diagnosis of in situ, minimally invasive, and invasive adenocarcinoma of the lung. Arch Pathol Lab Med. 2012;136:1515-1521.
29. Lee SM, Park CM, Goo JM, Lee HJ, Wi JY, Kang CH. Invasive pulmonary adenocarcinomas versus preinvasive lesions appearing as ground-glass nodules: differentiation by using CT features. Radiology. 2013;268:265-273.

SUPPORTING INFORMATION
Additional supporting information can be found online in the Supporting Information section at the end of this article.

How to cite this article: Li Z, Cai J, Zhao Y, Cai J, Zhao X. Folate receptor-positive circulating tumor cells in the preoperative diagnosis of indeterminate pulmonary nodules. J Clin Lab Anal. 2022;36:e24654. doi: 10.1002/jcla.24654