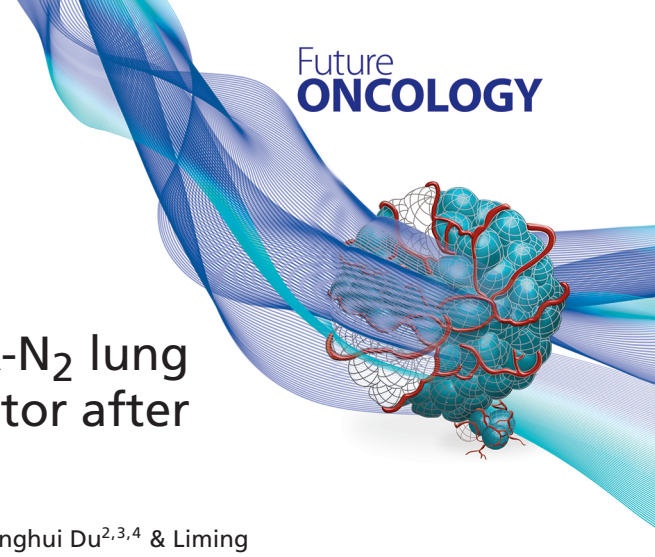
Research Article

For reprint orders, please contact: reprints@futuremedicine.com



Micropapillary pattern of stage IIIA-N_2 lung adenocarcinoma is a prognostic factor after adjuvant chemoradiotherapy

Jian Zeng[1,2,3], Xiaoying Cui[2,3,4,5], Lei Cheng[2,3,4], Ying Chen[1,2,3,4], Xianghui Du[2,3,4] & Liming Sheng*[,2,3,4]

[1]Department of Thoracic Surgery, Cancer Hospital of The University of Chinese Academy of Sciences (Zhejiang Cancer Hospital), Hangzhou, Zhejiang, China

[2]Institute of Cancer & Basic Medicine (IBMC), Chinese Academy of Sciences, Hangzhou, Zhejiang, China

[3]Department of Radiotherapy, Cancer Hospital of The University of Chinese Academy of Sciences (Zhejiang Cancer Hospital), Hangzhou, Zhejiang, China

[4]Key Laboratory Diagnosis & Treatment Technology on Thoracic Oncology, Zhejiang, China

[5]The Second Clinical Medical College, Zhejiang Chinese Medical University

*Author for correspondence: m05shengliming@126.com

Aim: This study aims to investigate the significance of a micropapillary pattern in stage IIIA-N_2 lung adenocarcinoma after adjuvant chemoradiotherapy. **Patients & methods:** A total of 257 patients with stage IIIA-N_2 lung adenocarcinoma were enrolled in this study. Patients were classified into three groups based on the proportion of micropapillary components: micropapillary negative, micropapillary minor component and micropapillary predominant component. **Results:** The micropapillary predominant group had the shortest median disease-free survival and overall survival times compared with the micropapillary minor component and micropapillary negative groups (median overall survival time: 54 months vs 64 months vs not reached; $p = 0.004$). Furthermore, the micropapillary pattern was an independent prognostic factor for disease-free survival and overall survival ($p < 0.05$). **Conclusion:** The micropapillary pattern of IIIA-N_2 lung adenocarcinoma is related to worse prognosis.

First draft submitted: 9 June 2020; Accepted for publication: 14 August 2020; Published online: 1 September 2020

Keywords: adjuvant chemoradiotherapy • micropapillary pattern • non-small-cell lung cancer • prognosis

Non-small-cell lung cancer (NSCLC) is the most common malignant neoplasm in the world. Squamous cell carcinoma and adenocarcinoma are the most common histologic subtypes, accounting for about 80–85% of all lung cancer. For early-stage NSCLC, surgery has been the standard treatment for many decades. For advanced metastatic disease, systemic treatment can prolong the life of patients. Completely resected stage IIIA-N_2 NSCLC is a heterogeneous disease with a 5-year overall survival (OS) rate of 14–51% [1–4]. There is still no clear consensus for the optimal management of these patients; a multimodality treatment is regarded as the most superior strategy [5]. For bulky unresectable stage IIIA-N_2 NSCLC, concurrent chemoradiotherapy is the main treatment [6]. Surgery and postoperative adjuvant chemotherapy are the primary treatments for resected stage IIIA-N_2 NSCLC. Both induction chemotherapy and induction chemoradiotherapy followed by surgery are treatment options [3]. After such intensive treatments, 40% of patients still have a local-regional recurrence. A retrospective analysis of the ANITA study indicates that postoperative adjuvant radiotherapy (PORT) for patients with pathologic IIIA-N_2 NSCLC can prolong survival [7]. Furthermore, many retrospective studies show that PORT is associated with an increase in survival in patients with N_2 nodal disease [8,9]. However, the role of adjuvant radiotherapy in this setting remains controversial before the results of large-scale randomized clinical trials. The ongoing randomized controlled trial LUNG ART (ClinicalTrials.gov: NCT00410683) will give a clear answer [10].

In 2011 invasive lung adenocarcinoma was classified into several subtypes based on the predominant pattern, including lepidic, acinar, papillary, solid and micropapillary; invasive mucinous adenocarcinoma was also classified on the basis of the novel histologic classification proposed by the International Association for the Study of Lung

Cancer (IASLC), the American Thoracic Society (ATS) and the European Respiratory Society (ERS) [11]. Studies reported that these subtypes were strongly linked to the risk of recurrence of lung adenocarcinoma [12,13]. The subtypes are organized into three groups according to the risk of recurrence and metastasis: low-risk group (lepidic, invasive mucinous adenocarcinoma), intermediate-risk group (acinar and papillary) and high-risk group (solid and micropapillary). Most lung adenocarcinomas contain mixed types. Tumors containing some micropapillary components, even if these are not predominant, are more aggressive than tumors without micropapillary components [14]. The micropapillary predominant (MPP) subtype is related to mediastinal lymph node metastasis and short disease-free survival (DFS) [15]. In early-stage NSCLC, patients with the MPP subtype are more prone to local-regional recurrence, with important implications for adjuvant radiotherapy and chemotherapy [16]. Based on previous studies, whether lung adenocarcinoma subtypes are associated with tumor recurrence and prognosis remains unknown and needs further investigation. Therefore we analyzed 257 consecutive patients who were diagnosed with completely resected pathologic stage IIIA-N_2 NSCLC between January 2013 and December 2017 in the Cancer Hospital of the University of Chinese Academy of Sciences (Zhejiang Cancer Hospital, Hangzhou, China). This study aims to investigate the role of the MPP subtype in completely resected IIIA-N_2 lung adenocarcinoma.

Methods

Patients

Between January 2013 and December 2017, patients with pathologic stage IIIA-N_2 lung adenocarcinoma who received post-lobectomy adjuvant chemoradiotherapy were enrolled in the study. Patients should undergo systematic thoracic lymph node dissection to ensure the accuracy of postoperative disease staging. Before surgery, patients underwent chest and abdomen CT scan, brain MRI and bone emission CT scan to exclude distant metastasis. A PET-CT scan was also recommended whenever possible. Pathologic staging for all patients was based on the American Joint Committee on Cancer's (AJCC) Tumor/Node/Metastases (TNM) classification system (8th edition). Patients who underwent palliative resection, segmentectomy, sleeve lobectomy, wedge resection or total pneumonectomy were excluded from this study. Furthermore, patients who received neoadjuvant chemotherapy or were diagnosed with previous or coexisting cancer other than lung adenocarcinoma were excluded. A total of 257 eligible patients were enrolled in this retrospective study. This study was approved by the institutional review board of Cancer Hospital of the University of Chinese Academy of Sciences (Zhejiang Cancer Hospital). Clinicopathologic information such as age, gender, tumor location, histopathology, tumor stage, tumor diameter, resection margins, recurrence and survival was obtained from patients' charts.

Pathologic evaluation

Surgical specimens were routinely dehydrated, embedded and fixed according to standard pathologic practice. Histopathologic analysis of each slide was performed independently by two experienced pathologists. Lung adenocarcinoma was classified into one of six subtypes according to the 2011 IASLC/ATS/ERS lung adenocarcinoma classification system: lepidic, acinar, papillary, micropapillary, solid predominant subtypes and invasive mucinous adenocarcinoma [11]. For mixed specimens with more than two subtypes, the histologic subtype was defined as the predominant subtype, which occupied the highest percentage. Based on the proportion of micropapillary subtype, histopathologic subtypes were divided into three groups: micropapillary predominant component (MPP), micropapillary minor component (MPM) and micropapillary negative (MPN).

Adjuvant chemotherapy & radiotherapy

All patients underwent postoperative adjuvant chemotherapy with pemetrexed plus cisplatin. Intramuscular injection of vitamin B12 and folic acid was administered 1 week before pemetrexed chemotherapy. Pemetrexed was intravenously administered at a dose level of 500 mg/m^2 (day 1), and cisplatin was administered at a dose level of 25 mg/m^2 (days 1–3) every 21 days for four cycles. Pemetrexed plus carboplatin could also be used in patients aged over 70 years or with renal insufficiency. Carboplatin was administered at a fixed dose of area under the plasma concentration–time curve of 5 mg/ml/min. If the patient had serious complications from chemotherapy at the prescribed dose, then the dose could be reduced by 20%. Chemotherapy could be suspended if intolerable side effects occurred or if the patient refused to continue.

Sequential radiotherapy was initiated after the completion of adjuvant chemotherapy. PORT was delivered by intensity-modulated radiotherapy, which was designed by Philips Radiation Oncology Systems (Pinnacle version 8.0, ADAC Laboratories, CA, USA). The clinical target volume included the bronchial stump, mediastinal nodal

stations and corresponding high-risk draining lymph node stations, as defined by Femke *et al.* [17]. The plan target volume was defined as the clinical target volume plus 5–15 mm to account for the daily setup variation and respiratory movement. Radiotherapy was administered in 2.0 Gy once daily for 5 days per week up to a total dose of 50 Gy. The dose limitations of the surrounding organs at risk were as follows: the lung mean dose was below 16.0 Gy; V_{20} (volume receiving >20 Gy) less than 30%, V_{30} (volume receiving >30 Gy) below 20%, and V_5 (volume receiving >5 Gy) less than 60%. The V_{40} of the heart was below 40%. The maximum spinal cord dose was 45.0 Gy. The plan target volume encompassed at least a 95% isodose line. The dose–volume histogram was obtained for the plan target volume, spinal cord, lung and heart.

Statistical analysis

A Pearson chi-square test was used to assess the correlation between lung adenocarcinoma subtypes and clinicopathologic variables such as gender, age, tumor location, tumor diameter and T stage. DFS and OS were analyzed by Cox proportional hazard regression and Kaplan–Meier curves. Furthermore, we used the Cox proportional hazard model with the backward selection method for multivariate analysis. The factor analyzed in univariate analysis ($p < 0.05$) was included in the multivariate analysis. All statistical calculations were performed with SPSS 13.0 for Windows (SPSS Inc, IL, USA). A p-value of less than 0.05 was considered statistically significant.

Results

Patients' characteristics

A total of 257 patients were enrolled in this retrospective study. The characteristics of these patients were summarized in Table 1. This study included 127 (49.4%) men and 130 (50.6%) women. Patients' ages ranged from 25 to 78 years (median: 58 years). Based on the AJCC 8th TNM staging system, 103 (40.1%), 126 (49.0%) and 28 (10.9%) patients were T_1, T_2 and T_3, respectively. Lesions of the left and right lungs were found in 99 and 158 cases, respectively. Tumor diameter ranged from 0.9 to 6.8 cm, with a median diameter of 2.5 cm. A total of 4353 lymph nodes were removed after pathologic examination and the number of positive lymph nodes was 1380. The number of positive mediastinal lymph nodes was distributed from 1 to 26. A total of 124 patients had driver gene alterations: 106 with *EGFR* mutation and 18 with *ALK* gene rearrangement. Of the 257 patients, 22 had a micropapillary-predominant lung adenocarcinoma, 34 papillary-predominant, 32 solid-predominant, 38 lepitic-predominant, 89 acinar-predominant and 42 invasive mucinous lung adenocarcinoma.

Relationship between clinicopathologic variables & histologic patterns

The relationship between clinicopathologic variables and histologic patterns in lung adenocarcinoma patients is presented in Table 1. According to the proportion of micropapillary subtype, 22 patients were classified as MPP, 78 as MPM and 157 as MPN. The histologic patterns and patients' ages were unrelated ($p = 0.770$). Compared with male patients, female patients were more likely to have MPP or MPM disease, but the difference was not statistically significant ($p = 0.098$). Lung adenocarcinoma with MPP or MPM had a large mass, advanced stage and mediastinal lymph node metastasis, but no statistical difference was found ($p > 0.05$). The histologic pattern was related to driver gene alteration and pleural invasion ($p < 0.05$). MPP or MPN was significantly high in patients with positive pleural invasion ($p = 0.011$) and positive *EGFR* mutation or *ALK* gene rearrangement ($p = 0.006$).

Survival analysis

The median follow-up time was 40.0 months (range: 5–94 months). Tumor recurrence was observed in 129 patients (50.2%). Sixty-seven patients (26.1%) died because of disease progression. The 5-year DFS and OS rates were 41.0 and 62.7%, respectively. The median DFS and OS were 46.0 and 83.0 months, respectively.

The 5-year DFS and OS rates for different histologic patterns were 20.5 and 39.5% for MPP, 24.5 and 55.1% for MPM, and 53.6 and 72.2% for MPN, respectively (Figure 1 & Figure 2). Patients with MPP showed significantly shorter DFS time than the other two groups (median DFS 27.0 months for MPP vs 39.0 months for MPM vs 62.0 months for MPN; $p = 0.032$).

Furthermore, histologic patterns were divided into two groups (with and without micropapillary) based on the presence or absence of micropapillary components. Univariate analysis indicated that micropapillary components (hazard ratio [HR]: 1.56; $p = 0.012$), tumor diameter (HR: 1.57; $p = 0.013$), T stage (HR: 2.02; $p = 0.037$) and the total number of positive lymph nodes (HR: 1.42; $p = 0.049$) were significantly associated with DFS (Table 2). These prognostic variables were incorporated into the multivariate analysis. The results indicated that micropapillary

Table 1. The relationship between clinicopathologic variables and histologic pattern in lung adenocarcinoma patients.

Variables	n	MPP, n (%)	MPM, n (%)	MPN, n (%)	p-value
Gender					
– Female	130	13 (10.0)	46 (35.4)	71 (54.6)	0.098
– Male	127	9 (7.1)	32 (25.2)	86 (67.7)	
Age					
– ≥60	155	12 (7.7)	49 (31.6)	94 (60.6)	0.770
– <60	102	10 (9.8)	29 (28.4)	63 (61.8)	
Tumor location					
– Left	99	10 (10.1)	33 (33.3)	56 (56.6)	0.482
– Right	158	12 (7.6)	45 (28.5)	101 (63.9)	
Tumor diameter					
– ≤3 cm	176	13 (7.4)	52 (29.5)	111 (63.1)	0.503
– >3 cm	81	9 (11.1)	26 (32.1)	46 (56.8)	
T stage					
– T_1	103	7 (6.8)	24 (23.3)	72 (69.9)	0.060
– $T_{2–3}$	154	15 (9.7)	54 (35.1)	85 (55.2)	
Total number of positive LN					
– ≤4	147	10 (6.8)	44 (29.9)	93 (63.3)	0.466
– >5	110	12 (10.9)	34 (30.9)	64 (58.2)	
Total number of positive MLN					
– ≤2	174	13 (7.5)	48 (27.6)	113 (64.9)	0.182
– >3	83	9 (10.8)	30 (36.1)	44 (53.0)	
Station status of MLN					
– Single	161	13 (8.1)	42 (26.1)	106 (65.8)	0.117
– Multiple	96	9 (9.4)	36 (37.5)	51 (53.1)	
Driver gene alteration					
– Negative	133	11 (8.3)	29 (21.8)	93 (69.9)	0.006
– Positive	124	11 (8.9)	49 (39.5)	64 (51.6)	
Pleural invasion					
– Negative	143	9 (6.3)	35 (24.5)	99 (69.2)	0.011
– Positive	114	13 (11.4)	43 (37.7)	58 (50.9)	
Lymphovascular invasion					
– Negative	191	14 (7.3)	55 (28.8)	122 (63.9)	0.240
– Positive	66	8 (12.1)	23 (34.8)	35 (53.0)	
Nerve invasion					
– Negative	234	22 (9.4)	72 (30.8)	140 (59.8)	0.224
– Positive	23	0 (0)	6 (26.1)	17 (73.9)	

LN: Lymph node; MLN: Mediastinal lymph node; MPM: Micropapillary minor; MPN: Micropapillary negative; MPP: Micropapillary predominant.

components, tumor diameter and T stage were the three independent prognostic factors for DFS. The prognostic variables identified in the univariate analysis of OS, including micropapillary (HR: 2.13; p = 0.002), the total number of positive lymph nodes (HR: 1.68; p = 0.033) and pleural invasion (HR: 1.69; p = 0.034), remained significant in the multivariate analysis. Patients with micropapillary components had an elevated risk of disease progression and death compared with those without micropapillary. The HR was 1.58 (95% CI: 1.08–2.30) for disease progression and 1.85 (95% CI: 1.09–3.12) for death.

Discussion

This study aimed to investigate the relationship between the subtype classification of lung adenocarcinoma and prognosis in patients with stage pIIIA-N_2 disease. This study included a relatively large sample of 257 patients with lung adenocarcinoma. The histologic pattern was related to driver gene alteration, pleural invasion and prognosis. Multivariate analysis indicated that the micropapillary component was associated with poor patient outcomes.

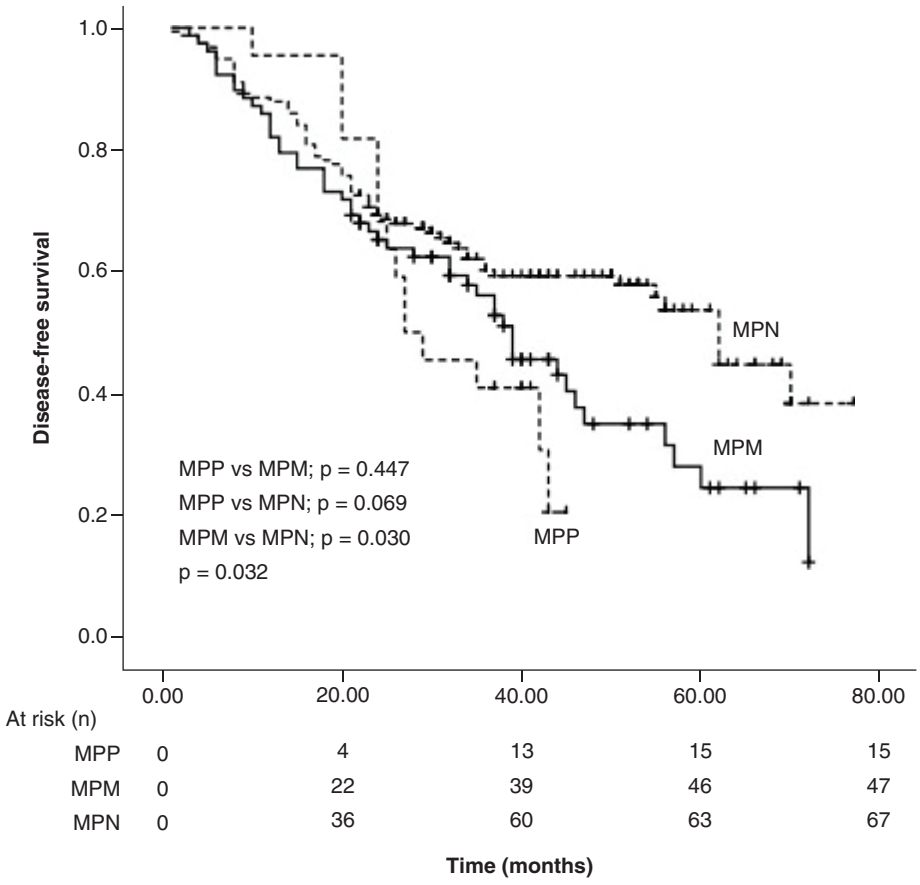


Figure 1. Kaplan–Meier survival curves of disease-free survival for different histologic patterns.
MPM: Micropapillary minor; MPN: Micropapillary negative: MPP: Micropapillary predominant.

Stage pIIIA-N2 lung adenocarcinoma is a heterogeneous group of diseases. For resectable patients, surgery is the standard treatment. Despite its potential for cure, 60% of patients are still prone to relapse and metastasis after treatment, resulting in death [18]. Therefore the treatment of patients with stage pIII-N_2 lung adenocarcinoma should be multidisciplinary, including surgery, chemotherapy and radiotherapy. However, most treatment options and layout are still in the exploration stage. The 5-year survival rate after surgery for stage III lung cancer is only 46%, which is very unsatisfactory [19]. Although there are currently no randomized clinical studies directly targeting stage III lung cancer, the results of the ANITA trial of mixed stage I–III patients show that platinum-based adjuvant chemotherapy can improve survival [20]. Vinorelbine and cisplatin are the most commonly used drugs in postoperative adjuvant chemotherapy, but their toxicity is relatively high, and half of the patients discontinue the drug due to excessive toxicity [21]. Pemetrexed is widely used in non-squamous-cell carcinoma because of its low toxicity. For patients with *EGFR* sensitive mutations, 2-year tyrosine kinase inhibitor (TKI) treatment after surgery can help prolong survival [22]. ADJUVANT/CTONG1104 established a clear role for adjuvant EGFR-TKI therapy in treating stage II–IIIA (N1–N2) *EGFR*-mutant NSCLC by comparing gefitinib versus vinorelbine plus cisplatin. Patients with completely resected, stage II–IIIA (N1-N2), *EGFR*-mutant (exon 19 deletion or exon 21 Leu858Arg) NSCLC were enrolled in this study. Gefitinib 250 mg once daily for 24 months) improved DFS to 28.7 months, versus 18.0 months (p = 0.0054) with adjuvant chemotherapy [23]. The role of PORT for patients with stage pIIIA-N_2 NSCLC remains controversial. In 1998, an individualized meta-analysis based on nine randomized controlled clinical trials indicated that the use of PORT was detrimental to unselected NSCLC populations [24]. The study results showed that PORT harms patient survival, directly increasing the relative risk of death by 21%. Subgroup analysis suggested that PORT reduced the survival of $N_{0–1}$ patients. However, for patients with N_2 disease, the use of PORT did not jeopardize survival and did not benefit survival. After the meta-analysis was published, the usage rate of PORT gradually decreased until the ANITA trial results were announced [20].

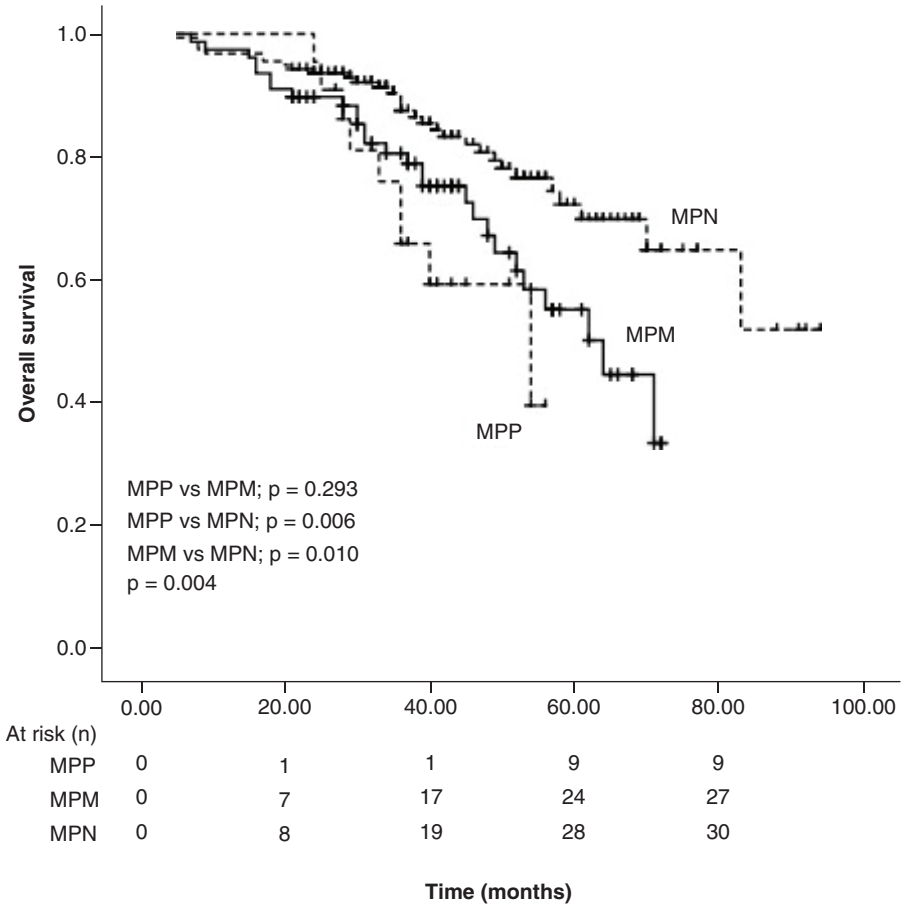


Figure 2. Kaplan–Meier survival curves of overall survival for different histologic patterns.
MPM: Micropapillary minor; MPN: Micropapillary negative; MPP: Micropapillary predominant.

In 2008, the ANITA trial explored the difference between postoperative chemotherapy and placebo in operative NSCLC and analyzed the effect of PORT on patient survival [7]. The results indicated that pN_2 patients treated with postoperative chemotherapy combined with sequential PORT could obtain longer median survival (47.4 months vs 23.8 months) and higher 5-year survival rate (47.4 vs 34.0%) than those treated with postoperative chemotherapy alone. Many large retrospective studies based on the Surveillance, Epidemiology and End Results database or the National Cancer Database showed that PORT could greatly reduce the risk of death in patients with stage IIIA-N_2 resectable NSCLC [9,25]. A small sample prospective randomized controlled study published in 2014 compared the effect of postoperative adjuvant chemoradiotherapy (POCRT) and postoperative adjuvant chemotherapy (POCT) in patients with pIIIA N_2 [26]. Patients in the POCRT group received PORT (50.4 Gy/28 fractions) concurrently with two cycles of chemotherapy followed by another two cycles of consolidative chemotherapy. A total of 135 patients were enrolled in this study. Compared with the POCT group, the median DFS of the POCRT group was significantly prolonged (28 months vs 18 months). However, given the small sample size, no difference in OS was found between the two groups. A Phase III randomized controlled study called 'LUNG ART' which explores the role of PORT in stage IIIA-N_2 NSCLC is underway [27]. In this study, all patients with stage pIIIA-N_2 NSCLC underwent postoperative adjuvant chemoradiation. The total dose of PORT was 50 Gy. The 5-year DFS and OS rates of the entire group of patients were 41.0 and 62.7%, respectively. This result was better than that presented in historical data [9,28,29] because half of the patients had a positive driver gene mutation. After the treatment failed, the patients underwent corresponding targeted therapy, which prolonged their survival. Moreover, this result was due to the use of PORT, which improved the local and regional control rates of these patients.

Previous studies have explored the prognostic factors of stage pIIIA N_2 NSCLC, including primary tumor size [30], lymph node ratio [31], number of lymph node stations [32] and mediastinal lymph node size [33]. In this

Table 2. Univariable and multivariable analyses of disease-free survival and overall survival using Cox's regression.

Variables	Univariate analysis			Multivariate analysis		
	HR	95% CI	p-value	HR	95% CI	p-value
Disease-free survival						
– Age (≥60 vs <60)	1.17	0.83–1.67	0.372			
– Gender (male vs female)	0.96	0.68–1.35	0.792			
– Micropapillary (present vs absent)	1.56	1.10–2.01	0.012	1.58	1.08–2.30	0.019
– Tumor location (right vs left)	0.72	0.51–1.02	0.063			
– Tumor diameter (>3 cm vs ≤3 cm)	1.57	1.10–2.25	0.013	2.14	1.30–3.54	0.003
– T stage ($T_{2–3}$ vs T_1)	2.02	1.04–3.89	0.037	1.99	1.02–3.86	0.043
– Total number of positive LN (>5 vs ≤4)	1.42	1.00–2.00	0.049			
– Total number of positive MLN (>3 vs ≤2)	1.05	0.73–1.51	0.808			
– Station number of MLN (multiple vs single)	1.24	0.87–1.77	0.225			
– Pleural invasion (positive vs negative)	1.17	0.73–1.67	0.354			
– Lymphovascular invasion (positive vs negative)	1.13	0.76–1.67	0.536			
– Nerve invasion (positive vs negative)	0.95	0.51–1.77	0.882			
Overall survival						
– Age (≥60 vs <60)	0.88	0.54–1.42	0.592			
– Gender (male vs female)	1.30	0.80–2.10	0.290			
– Micropapillary (present vs absent)	2.13	1.31–3.46	0.002	1.85	1.09–3.12	0.022
– Tumor location (right vs left)	1.18	0.71–1.97	0.514			
– Tumor diameter (>3 cm vs ≤3 cm)	1.21	0.74–2.00	0.447			
– T stage ($T_{2–3}$ vs T_1)	1.68	1.04–2.72	0.033	1.79	1.08–2.98	0.025
– Total number of positive LN (>5 vs ≤4)	1.24	0.76–2.00	0.393			
– Total number of positive MLN (>3 vs ≤2)	1.42	0.84–2.38	0.186			
– Station number of MLN (multiple vs single)	1.33	0.82–2.15	0.255			
– Pleural invasion (positive vs negative)	1.69	1.04–2.74	0.034	2.55	1.13–5.79	0.025
– Lymphovascular invasion (positive vs negative)	1.65	1.00–2.74	0.053			
– Nerve invasion (positive vs negative)	1.07	0.46–2.48	0.873			

HR: Hazard ratio; LN: Lymph node; MLN: Mediastinal lymph node.

study, tumor size was an independent prognostic factor. Our findings were consistent with that of a previous study based on a relatively small sample size [30]. Chen *et al.* [30] retrospectively analyzed 77 consecutive patients with stage pIIIA-N_2 NSCLC and found that tumor size <3 cm was associated with good prognosis. This study showed that the component of micropapillary was related to driver gene alteration and pleural invasion. Furthermore, MPP was associated with poor patient outcomes. Cai *et al.* [34] analyzed the data from 261 lung adenocarcinoma patients to investigate the relationship between intratumoral heterogeneity and treatment response. *EGFR*-sensitive mutation occurred frequently in papillary, lepidic and micropapillary components. This finding indicated that EGFR-TKI treatment should be limited to patients with micropapillary or papillary lung adenocarcinoma. Similarly, Tsuta *et al.* [35] found that *EGFR*, *KRAS* and *ALK* gene mutations were statistically prevalent in MPP lung adenocarcinomas ($p = 0.00001$). The component of micropapillary has changed some treatment indications. In a large retrospective study [36], stage IB NSCLC patients with micropapillary components could benefit from postoperative adjuvant chemotherapy. The usage of chemotherapy in patients with a MPP pattern reduced lung cancer-specific death by 54%. Another interesting finding in our study is the component of micropapillary borderline statistically significantly associated with T stage but not N stage. Furthermore, the T stage has a significant impact on OS while the N parameters do not; this finding is slightly different from other research results [37,38]. Lung adenocarcinoma with a micropapillary pattern is prone to tumor spread through air spaces, which is an important factor affecting the T stage [39]. It seems histologic pattern first serves as a local regulator, then as a systematic mediator. Although based on the results of this study, T stage is a better predictor than N stage, the number of positive lymph nodes was also related with OS. In a large-scale retrospective study, the use of PORT for stage IIIA pathologic N_2 NSCLC demonstrated better survival with more than three positive lymph nodes (n >3), but not for patients with three or fewer positive lymph nodes [40]. Single N_2 station involvement was a better predictor of benefit from PORT [41].

The component of micropapillary in *EGFR*-mutant NSCLC was related to early brain metastasis, indicating that conducting rigorous follow-up evaluations of the CNS for these patients is necessary to detect brain metastases early and seek better control. Leeman *et al.* [16] treated stage I–IIA lung adenocarcinoma with stereotactic body radiation therapy. One hundred and nineteen patients were enrolled. Of the patients who were diagnosed with solid and/or micropapillary pattern lung adenocarcinoma, 30% were defined as having a high-risk subtype. The presence of a micropapillary component determines an increased risk of tumor recurrence. Thus elevating the total radiation dose or adjuvant systemic or targeted therapy is necessary for early-stage lung adenocarcinoma patients who received stereotactic body radiation therapy.

Our study is limited by its retrospective nature and selective biases. In addition, information on post-treatment recurrence is insufficient, which may vary the reported survival of patients. Second, the postoperative chemotherapy regimen did not choose a unified drug. Some older patients received carboplatin instead of cisplatin. A prospective study is required to determine the prognostic value of histologic subtypes. Moreover, this study lacks a control group that did not undergo PORT. Therefore drawing a conclusion about which histologic subtype can benefit from PORT is impossible.

Conclusions

We evaluated the association between the histologic patterns of lung adenocarcinoma and the prognosis, and found that the micropapillary component was associated with poor outcomes. These findings might help identify patients with stage pIIIA-N_2 NSCLC who are eligible for PORT.

Summary points

- The component of micropapillary in lung adenocarcinoma was associated with poor patient outcomes.
- A total of 257 patients with IIIA-N_2 lung adenocarcinoma received adjuvant chemoradiotherapy.
- The micropapillary predominant component group had the shortest median disease-free survival and overall survival time compared with the micropapillary minor component and micropapillary negative groups.

Author contributions

J Zeng and L Sheng: data collection and analysis and the drafting of the manuscript. X Cui: analysis and interpretation of the data. L Cheng: conception and study design. X Du: drafting of the manuscript. All authors read and approved the final manuscript.

Acknowledgments

We thank all the patients who participated in this study and their families.

Funding & competing interests disclosure

This study was supported by grants from the Nature Science Foundation of Zhejiang Province (no. LY14H160012). The authors have no other relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter or materials discussed in the manuscript apart from those disclosed.

No writing assistance was utilized in the production of this manuscript.

Ethical conduct of research

The authors state that they have obtained appropriate institutional review board approval or have followed the principles outlined in the Declaration of Helsinki for all human or animal experimental investigations. In addition, for investigations involving human subjects, informed consent has been obtained from the participants involved.

Availability of data & material

All data generated or analysed during this study are included in this published article.

References

1 van Schil P, van Meerbeeck J, Kramer G *et al.* Morbidity and mortality in the surgery arm of EORTC 08941 trial. *Eur. Respir. J.* 26(2), 192–197 (2005).

2 van Meerbeeck JP, Kramer GW, van Schil PE *et al.* Randomized controlled trial of resection versus radiotherapy after induction chemotherapy in stage IIIA-N2 non-small-cell lung cancer. *J. Natl Cancer Inst.* 99(6), 442–450 (2007).

3 Eberhardt WE, Pottgen C, Gauler TC *et al.* Phase III study of surgery versus definitive concurrent chemoradiotherapy boost in patients with resectable stage IIIA(N_2) and selected IIIB non-small-cell lung cancer after induction chemotherapy and concurrent chemoradiotherapy (ESPATUE). *J. Clin. Oncol.* 33(35), 4194–4201 (2015).

4 Rajaram R, Correa AM, Xu T *et al.* Locoregional control, overall survival, and disease-free survival in stage IIIA (N_2) non-small-cell lung cancer: analysis of resected and unresected patients. *Clin. Lung Cancer*21(4), e294–e301 (2020).

5 Bograd AJ, Vallieres E. Surgery as a component of multimodality care for known stage IIIA-N2 non-small cell lung cancer. *Semin. Respir. Crit. Care Med.* 41(3), 346–353 (2020).

6 Bradley JD, Hu C, Komaki RR *et al.* Long-term results of NRG oncology RTOG 0617: standard- versus high-dose chemoradiotherapy with or without cetuximab for unresectable stage III non-small-cell lung cancer. *J. Clin. Oncol.* 38(7), 706–714 (2020).

7 Douillard JY, Rosell R, De Lena M *et al.* Impact of postoperative radiation therapy on survival in patients with complete resection and stage I, II, or IIIA non-small-cell lung cancer treated with adjuvant chemotherapy: the Adjuvant Navelbine International Trialist Association (ANITA) randomized trial. *Int. J. Radiat. Oncol. Biol. Phys.* 72(3), 695–701 (2008).

8 Lally BE, Zelterman D, Colasanto JM *et al.* Postoperative radiotherapy for stage II or III non-small-cell lung cancer using the surveillance, epidemiology, and end results database. *J. Clin. Oncol.* 24(19), 2998–3006 (2006).

9 Robinson CG, Patel AP, Bradley JD *et al.* Postoperative radiotherapy for pathologic N2 non-small-cell lung cancer treated with adjuvant chemotherapy: a review of the National Cancer Data Base. *J. Clin. Oncol.* 33(8), 870–876 (2015).

10 Le Pechoux C, Dunant A, Faivre-Finn C *et al.* Postoperative radiotherapy for pathologic N2 non-small-cell lung cancer treated with adjuvant chemotherapy: need for randomized evidence. *J. Clin. Oncol.* 33(26), 2930–2931 (2015).

11 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.* 6(2), 244–285 (2011).

12 Yoshiya T, Mimae T, Tsutani Y *et al.* Prognostic role of subtype classification in small-sized pathologic N0 invasive lung adenocarcinoma. *Ann. Thorac. Surg.* 102(5), 1668–1673 (2016).

13 Zhao X, Zhang Y, Qian K *et al.* Prognostic significance of the International Association for the Study of Lung Cancer/American Thoracic Society/European Respiratory Society classification of stage I lung adenocarcinoma: a retrospective study based on analysis of 110 Chinese patients. *Thorac. Cancer* 8(6), 565–571 (2017).

14 Wang W, Hu Z, Zhao J *et al.* Both the presence of a micropapillary component and the micropapillary predominant subtype predict poor prognosis after lung adenocarcinoma resection: a meta-analysis. *J. Cardiothorac. Surg.* 15(1), 154 (2020).

15 Warth A, Muley T, Meister M *et al.* The novel histologic International Association for the Study of Lung Cancer/American Thoracic Society/European Respiratory Society classification system of lung adenocarcinoma is a stage-independent predictor of survival. *J. Clin. Oncol.* 30(13), 1438–1446 (2012).

16 Leeman JE, Rimner A, Montecalvo J *et al.* Histologic subtype in core lung biopsies of early-stage lung adenocarcinoma is a prognostic factor for treatment response and failure patterns after stereotactic body radiation therapy. *Int. J. Radiat. Oncol. Biol. Phys.* 97(1), 138–145 (2017).

17 Spoelstra FO, Senan S, Le Pechoux C *et al.* Variations in target volume definition for postoperative radiotherapy in stage III non-small-cell lung cancer: analysis of an international contouring study. *Int. J. Radiat. Oncol. Biol. Phys.* 76(4), 1106–1113 (2010).

18 Goldstraw P, Crowley J, Chansky K *et al.* The IASLC Lung Cancer Staging Project: proposals for the revision of the TNM stage groupings in the forthcoming (seventh) edition of the TNM classification of malignant tumours. *J. Thorac. Oncol.* 2(8), 706–714 (2007).

19 Okami J, Shintani Y, Okumura M *et al.* Demographics, safety and quality, and prognostic information in both the seventh and eighth editions of the TNM classification in 18,973 surgical cases of the Japanese Joint Committee of Lung Cancer Registry Database in 2010. *J. Thorac. Oncol.* 14(2), 212–222 (2019).

20 Douillard JY, Rosell R, De Lena M *et al.* Adjuvant vinorelbine plus cisplatin versus observation in patients with completely resected stage IB-IIIA non-small-cell lung cancer (Adjuvant Navelbine International Trialist Association [ANITA]): a randomised controlled trial. *Lancet Oncol.* 7(9), 719–727 (2006).

21 Alam N, Shepherd FA, Winton T *et al.* Compliance with post-operative adjuvant chemotherapy in non-small cell lung cancer. An analysis of National Cancer Institute of Canada and intergroup trial JBR.10 and a review of the literature. *Lung Cancer* 47(3), 385–394 (2005).

22 Tachihara M, Dokuni R, Okuno K *et al.* Phase II study of adjuvant chemotherapy with pemetrexed and cisplatin with a short hydration method for completely resected nonsquamous non-small cell lung cancer. *Thorac. Cancer* 11(9), 2536–2541 (2020).

23 Zhong WZ, Wang Q, Mao WM *et al.* Gefitinib versus vinorelbine plus cisplatin as adjuvant treatment for stage II–IIIA (N1–N2) *EGFR*-mutant NSCLC (ADJUVANT/CTONG1104): a randomised, open-label, Phase 3 study. *Lancet Oncol.* 19(1), 139–148 (2018).

24 PORT Meta-analysis Trialists Group. Postoperative radiotherapy in non-small-cell lung cancer: systematic review and meta-analysis of individual patient data from nine randomised controlled trials. *Lancet* 352(9124), 257–263 (1998).

25 Wei S, Xie M, Tian J *et al.* Propensity score-matching analysis of postoperative radiotherapy for stage IIIA–N_2 non-small cell lung cancer using the Surveillance, Epidemiology, and End Results database. *Radiat. Oncol.* 12(1), 96 (2017).

26 Shen WY, Ji J, Zuo YS *et al.* Comparison of efficacy for postoperative chemotherapy and concurrent radiochemotherapy in patients with IIIA-pN_2 non-small cell lung cancer: an early closed randomized controlled trial. *Radiother. Oncol.* 110(1), 120–125 (2014).

27 Le Pechoux C. Role of postoperative radiotherapy in resected non-small cell lung cancer: a reassessment based on new data. *Oncologist* 16(5), 672–681 (2011).

28 Asamura H, Chansky K, Crowley J *et al.* The International Association for the Study of Lung Cancer Lung Cancer Staging Project: proposals for the revision of the n descriptors in the forthcoming 8th edition of the TNM classification for lung cancer. *J. Thorac. Oncol.* 10(12), 1675–1684 (2015).

29 Groome PA, Bolejack V, Crowley JJ *et al.* The IASLC Lung Cancer Staging Project: validation of the proposals for revision of the T, N, and M descriptors and consequent stage groupings in the forthcoming (seventh) edition of the TNM classification of malignant tumours. *J. Thorac. Oncol.* 2(8), 694–705 (2007).

30 Chen CY, Wu BR, Chen CH *et al.* Prognostic value of tumor size in resected stage IIIA-N_2 non-small-cell lung cancer. *J. Clin. Med.* 95), 1307 (2020).

31 Han H, Zhao Y, Gao Z *et al.* A prognostic score system with lymph node ratio in stage IIIA-N_2 NSCLC patients after surgery and adjuvant chemotherapy. *J. Cancer Res. Clin. Oncol.* 145(8), 2115–2122 (2019).

32 Yoon HY, Lee JC, Kim SW *et al.* Prognosis of multi-level N2-positive non-small cell lung cancer according to lymph node staging using endobronchial ultrasound-transbronchial biopsy. *Thoracic Cancer* 9(6), 684–692 (2018).

33 Rice SR, Molitoris JK, Vyfhuis MAL *et al.* Lymph node size predicts for asymptomatic brain metastases in patients with non-small-cell lung cancer at diagnosis. *Clin. Lung Cancer* 20(1), e107–e114 (2019).

34 Cai Y, Wu H, Shi X *et al.* Heterogeneous components of lung adenocarcinomas confer distinct *EGFR* mutation and *PD-L1* expression. *BMC Cancer* 20(1), 148 (2020).

35 Tsuta K, Kawago M, Inoue E *et al.* The utility of the proposed IASLC/ATS/ERS lung adenocarcinoma subtypes for disease prognosis and correlation of driver gene alterations. *Lung Cancer* 81(3), 371–376 (2013).

36 Qian F, Yang W, Wang R *et al.* Prognostic significance and adjuvant chemotherapy survival benefits of a solid or micropapillary pattern in patients with resected stage IB lung adenocarcinoma. *J. Thorac. Cardiovasc. Surg.* 155(3), 1227–1235 e1222 (2018).

37 Chang C, Sun X, Zhao W *et al.* Minor components of micropapillary and solid subtypes in lung invasive adenocarcinoma (</= 3 cm): PET/CT findings and correlations with lymph node metastasis. *Radiol. Med.* 125(3), 257–264 (2020).

38 Pani E, Kennedy G, Zheng X *et al.* Factors associated with nodal metastasis in 2-centimeter or less non-small cell lung cancer. *J. Thorac. Cardiovasc. Surg.* 159(3), 1088–1096.e1 (2020).

39 Kadota K, Nitadori JI, Sima CS *et al.* Tumor spread through air spaces is an important pattern of invasion and impacts the frequency and location of recurrences after limited resection for small stage I lung adenocarcinomas. *J. Thorac. Oncol.* 10(5), 806–814 (2015).

40 Wang S, Ma Z, Yang X *et al.* Choice of postoperative radiation for stage IIIA pathologic N2 non-small cell lung cancer: impact of metastatic lymph node number. *Radiat. Oncol.* 12(1), 207 (2017).

41 Yuan C, Tao X, Zheng D *et al.* The lymph node status and histologic subtypes influenced the effect of postoperative radiotherapy on patients with N2 positive IIIA non-small cell lung cancer. *J. Surg. Oncol.* 119(3), 379–387 (2019).