BMC Pulmonary Medicine

RESEARCH

Open Access

Transbronchial lung cryobiopsy for interstitial lung disease: early experience, learning curve, and the impact of sedation on complication rates at a single centre in Japan

Shota Kaburaki[1*], Toru Tanaka[1], Koichiro Kamio[1], Yosuke Tanaka[1], Kazuo Kasahara[1] and Masahiro Seike[1]

Abstract

Background Transbronchial lung cryobiopsy (TBLC) has emerged as a promising diagnostic tool for interstitial lung disease (ILD). This study aimed to assess the initial experience, procedural learning curve, and influence of sedative medications on complication rates, particularly bleeding and pneumothorax, in the implementation of a TBLC program for ILD diagnosis.

Methods In this retrospective cohort study, we analysed 119 patients who underwent TBLC at Nippon Medical School Hospital from April 2021 to March 2024. Procedural times, complication rates, and histopathological outcomes were evaluated. The learning curve was assessed using cumulative sum (CUSUM) analysis, focusing on procedure time and biopsy yield. The association between sedative medication dosages and bleeding risk was also examined.

Results The overall diagnostic yield was high, with alveolar tissue obtained in 97.5% of cases and a definitive pathological diagnosis achieved in 81.5% of patients. CUSUM analysis revealed a proficiency threshold at approximately 56 cases, with improved efficiency and biopsy yield in the consolidation phase. Fentanyl dosage was significantly associated with reduced bleeding complications (odds ratio 0.51, 95% confidence interval 0.27–0.97, $p = 0.041$).

Conclusions TBLC is a safe and effective diagnostic tool for ILDs, with a manageable learning curve for procedural efficiency. Sedation, particularly fentanyl dosage, may plays a crucial role in minimizing complications, but further research is needed to clarify this relationship. These findings support the adoption of TBLC as a standard diagnostic approach for ILD and highlight the importance of adequate training and optimized sedation protocols to ensure safety and efficacy in clinical practice.

Keywords Transbronchial lung cryobiopsy, Interstitial lung disease, Cumulative sum learning curve analysis, Sedation, Fentanyl

*Correspondence:
Shota Kaburaki
s-kaburaki@nms.ac.jp
[1]Department of Pulmonary Medicine and Oncology, Graduate School of Medicine, Nippon Medical School, 1-1-5, Sendagi, Bunkyo-ku, Tokyo 113-8603, Japan

Introduction

Diagnosis of interstitial lung disease (ILD) often requires a multidisciplinary approach integrating clinical, radiological and pathological data. When pathological information is needed to make a definitive diagnosis, surgical lung biopsy (SLB) has traditionally been the gold standard for obtaining adequate lung-tissue samples. However, SLB is associated with significant morbidity and mortality, especially in patients with advanced disease [1, 2]. Transbronchial lung cryobiopsy (TBLC) is a promising new alternative to SLB for obtaining lung tissue in the diagnostic evaluation of ILD. Studies have shown that larger specimens with fewer artefacts are produced by TBLC than by conventional transbronchial forceps biopsy, and the diagnostic yield of TBLC for ILD appears to be comparable to that of SLB in the context of multidisciplinary discussion [3, 4]. However, TBLC is not without risks. Complications, such as haemorrhage, pneumothorax and acute exacerbation of ILD have been reported [4]. In a recent study, when TBLC was first introduced into clinical practice at their institution, the incidence of complications, including life-threatening bleeding, was particularly high [5]. This raises concerns about the real-world safety of TBLC as it transitions from specialised research centres to widespread use.

As TBLC is a novel procedure, understanding the learning curve is crucial for its safe and effective implementation. In addition to operator experience and patient selection, appropriate airway management and anaesthesia may also affect the safety of TBLC. Establishing training and competency standards to guide the safe introduction of TBLC would be beneficial. A commonly used statistical method for quantitatively assessing the learning curve by analysing changes in key performance indicators over consecutive cases is the cumulative sum (CUSUM) method [6]. Previous CUSUM analysis has been applied to TBLC, which showed that diagnostic yield and sample size improved after 50 to 70 cases, suggesting a plateau in the learning curve [7]. However, the diagnostic yield in ILD is strongly influenced by patient-related factors; therefore, an evaluation of the learning curve with respect to the efficiency of the procedural technique is necessary.

The study aim was to evaluate our centre's early experience with implementing a programme for diagnosing ILD in patients admitted to undergo TBLC. We focused on identifying risk factors for complications, used CUSUM analysis of the procedure time per biopsy to define the learning curve, and analysed the association between sedative medications and bleeding risk. We hypothesised that procedural efficiency would be low during the initial learning phase but would increase with experience and that various factors, such as patient characteristics, procedural techniques and sedation, might influence the risk of adverse events. By analysing the outcomes and trends in our initial cohort, we hope to provide insights for optimising the performance and safety of TBLC and improving the diagnosis and management of ILDs.

Methods

Study design and patient population

This retrospective cohort study included patients who underwent TBLC for the diagnosis of ILD at Nippon Medical School Hospital between April 2021 and March 2024. The Nippon Medical School Hospital Institutional Review Board approved this study (approval number B-2024-875) and waived informed consent due to its retrospective nature.

Eligible patients were adults (≥18 years) with suspected ILD based on clinical and radiographic findings who were referred for TBLC after multidisciplinary discussion. Patients taking antiplatelet or anticoagulant medications were required to discontinue these medications until they were no longer effective before undergoing the procedure. The exclusion criteria included inability to tolerate bronchoscopy, haemorrhagic disease, severe pulmonary hypertension or other conditions determined by the attending physician.

Demographic and clinical data were abstracted from the electronic medical record, including age, sex, body-mass index, smoking history, comorbidities, pulmonary function tests and chest high-resolution computed tomography (HRCT) findings.

TBLC procedure

All TBLC procedures were performed by a team of more than three pulmonologists in the bronchoscopy suite who used an endotracheal tube with an inner diameter of 8.0 mm with the patient under local anaesthesia. TBLC procedures were conducted three times a week by three rotating groups. Due to the nature of our university hospital, these groups changed every six months. Each group consisted of at least three physicians, always including a respiratory specialist with over 15 years of experience and a third-year respiratory medicine resident. The experience of other physicians ranged from 4 to 15 years. Additionally, each procedure always included at least one physician who had completed training in at least 10 TBLC cases at a tertiary referral centre with extensive experience in TBLC before the initiation of our TBLC program.

Patient monitoring included continuous pulse oximetry, electrocardiography and intermittent blood-pressure assessments. After airway examination using a flexible bronchoscope (BF-1TQ290, Olympus, Tokyo, Japan), a 1.9-mm cryoprobe (Erbe, Tubingen, Germany) was introduced through the working channel and advanced to the target region under fluoroscopic guidance, maintaining

a distance of 10–20 mm from the pleura. The duration of cryoadhesion (freeze time) ranged from 5 to 7 s. The bronchoscope and cryoprobe were simultaneously removed en-bloc, and the obtained specimen was immediately thawed in saline. A 4-Fr Fogarty Catheter (Edwards Lifesciences, California, United States) was prophylactically placed to tamponade any potential bleeding. At least one biopsy specimen was obtained from distinct segments of the lower lobe, specifically targeting areas of abnormality identified on pre-procedure HRCT. Post-procedure, a chest X-ray was taken immediately and then repeated the following day to look for pneumothorax. Anaesthesia was intermittently administered during the procedure with intravenous doses of fentanyl and midazolam at the pulmonologist's discretion, with doses ranging from 20 to 40 μg for fentanyl and 2 to 6 mg for midazolam per administration. The maximum total doses allowed were 100 μg of fentanyl and 30 mg of midazolam, respectively. Hypoxia was managed by administering oxygen via an endotracheal tube. Data collected from TBLC sampling encompassed the number and anatomical locations of the biopsies as well as the freeze-time durations.

Complications and safety assessments

Intra- and post-procedural complications were recorded, including bleeding, pneumothorax, respiratory-tract infection, hypotension, respiratory failure and death. Bleeding severity was graded as mild (requiring suction only), moderate (requiring topical iced saline, vasoconstrictors or thrombin) or severe (caused procedure stop or hemodynamic instability or required transfusion, surgical intervention or admission to the intensive care unit) [8]. Significant bleeding was defined as moderate or greater. Pneumothorax was classified as small (<2 cm from chest wall to lung edge) or large (≥2 cm or symptomatic, requiring chest tube). Complications were attributed to TBLC if they occurred within 30 days of the procedure.

Histopathological assessment

The TBLC specimens were fixed in formalin, embedded in paraffin, sliced into 4-μm thick sections and stained with haematoxylin & eosin. A pathologist specialising in ILDs performed the histopathological assessment. Specimens were considered adequate if they contained alveolated lung parenchyma. A histopathological diagnosis was reached by integrating the clinical, radiological and pathological data according to standard guidelines.

CUSUM learning-curve analysis

The TBLC learning curve was assessed by performing CUSUM analysis, a method commonly used to evaluate the learning process for surgical procedures [9]. The CUSUM analysis examined the procedure time per number of biopsies. These CUSUM values were then plotted against the case numbers, with the target time defined as the mean procedure time for the entire cohort. The learning curve was considered complete when an inflection point was observed, indicating sustained improvement in efficiency. The number of procedures needed to achieve proficiency was determined. Additionally, a risk-adjusted cumulative summation (RA-CUSUM) analysis was performed to account for patient-specific factors that might influence procedural outcomes. Patient factors for risk adjustment included age, sex, body mass index, percent predicted forced vital capacity (FVC), and percent predicted diffusing capacity for carbon monoxide (DLCO). A linear regression model was constructed to predict the expected procedure time per biopsy based on these risk factors. The RA-CUSUM was calculated by cumulatively summing the differences between observed and expected procedure times across all cases.

Statistical analysis

Descriptive statistics were used to summarise patient characteristics, TBLC parameters and complications. Continuous variables were reported as the mean ± standard deviation or median and interquartile range. Categorical variables were expressed as counts and percentages.

Univariate and multivariable logistic regression were performed to determine potential risk factors for complications, with results reported as odds ratios with 95% confidence intervals. Variables with $p<0.05$ on univariate analysis were included in the multivariable model. A two-tailed $p<0.05$ was accepted as indicating statistical significance. R version 3.61 with R studio was used to perform all statistical analyses.

Subgroup analysis

To further investigate the findings from our primary analysis, we planned a subgroup analysis. The study cohort was divided into two phases based on the proficiency threshold identified by the CUSUM analysis: an initial phase and a consolidation phase. For factors that showed statistically significant associations ($p<0.05$) with procedural outcomes in the primary analysis, we performed separate logistic regression analyses in each phase.

Post-hoc power analysis

A post-hoc power analysis was conducted to evaluate the statistical power of two key analyses: the CUSUM analysis comparing procedural times between the initial and consolidation phases, andthe logistic regression model assessing factors significantly associated with the risk of complications, particularly significant bleeding.

For the CUSUM analysis, power was calculated using a two-sample t-test, with the effect size (Cohen's d) computed from the difference in mean procedural times between phases, divided by the pooled standard deviation. For the logistic regression analysis, the observed odds ratio of factors found to be significantly associated with complications in the primary analysis was used to calculate power. The significance level (α) was set at 0.05 for both analyses. The significance level (α) was set at 0.05 for both analyses.

Power calculations were performed using the statsmodels package (version 0.12.2) in Python 3.8, utilizing the TTestIndPower and NormalIndPower classes for the CUSUM and logistic regression analyses, respectively.

Table 1 Characteristics of the patients

Variable	*n* = 119 (100%)
Demographics	
Age (years)	68.6 ± 9.0
Sex, male	65 (54.6)
BMI (kg/m^2)	23.4 ± 4.1
Never smoker	54 (45.4)
Pulmonary Function Tests	
FVC %predicted (%)	84.3 ± 22.1
FEV1/FVC (%)	79.6 ± 15.2
FEV1%predicted (%)	91.0 ± 20.4
DLCO %predicted (%)	76.2 ± 20.7
Procedural Details	
Procedural time (min)	38.5 ± 8.6
Fentanyl (μg)	92.4 ± 14.0
Fentanyl (μg/kg/h)	2.58 ± 0.88
Midazolam (mg)	11.8 ± 4.9
Midazolam (mg/kg/h)	0.32 ± 0.14
Number of biopsies	2.3 ± 0.9
Number of biopsies per 30 min	1.9 ± 0.9
Biopsy from upper lobes	38 (31.9)
Complications	
Pneumothorax small	1 (0.8)
large	1 (0.8)
Bleeding mild	82 (68.9)
moderate	26 (21.8)
severe	5 (4.2)
Significant bleeding	31 (26.1)
Respiratory tract infection	3 (2.5)
90-day mortality	0 (0.0)
Histopathological Findings	
Alveolar tissue obtained	116 (97.5)
Pathological diagnosis achieved	97 (81.5)

Data are presented as mean ± SD or n (%). BMI, body mass index; DLCO, diffusing capacity of the lung for carbon monoxide; FEV1, forced expiratory volume in 1 s; FVC, forced vital capacity

Results

Study design and patient population

A total of 119 patients were included in this retrospective cohort study. Their mean age was 68.6±9.0 years, and 65 (54.6%) were male. The mean body-mass index was 23.4±4.1 kg/m^2, and 54 (45.4%) were never smokers. The mean FVC percent predicted, the ratio of forced expiratory volume in 1 s (FEV1) to FVC, FEV1% predicted and DLCO percent predicted were 84.3% ± 22.1%, 79.6% ± 15.2%, 91.0% ± 20.4% and 76.2% ± 20.7%, respectively (Table 1). No patient exhibited severe renal dysfunction, defined as an estimated glomerular filtration rate of <30 mL/min/1.73 m².

TBLC procedure

The mean procedural time was 38.5±8.6 min. Patients received an average of 92.4±14.0 μg of fentanyl (2.58±0.88 μg/kg/h) and 11.8±4.9 mg of midazolam (0.32±0.14 mg/kg/h). The mean number of biopsies was 2.3±0.9, with an average of 1.9±0.9 biopsies performed per 30 min. Biopsies were taken from the upper lobes in 38 (31.9%) patients (Table 1).

Complications and safety assessments

Pneumothorax occurred in 2 (1.6%) patients, with 1 (0.8%) small and 1 (0.8%) large. Bleeding was observed in 113 (94.9%) patients, with 82 (68.9%) mild, 26 (21.8%) moderate and 5 (4.2%) severe. Significant bleeding (moderate or above) occurred in 31(26.1%) patients. Respiratory-tract infection was reported in 3 (2.5%) patients. No deaths were reported within 90 days of the procedure. No patient exhibited hypotension necessitating the use of vasopressors or respiratory failure requiring assisted ventilation.

Histopathological assessment

Adequate alveolar tissue was obtained in 97.5% of cases, and the histopathological diagnoses based on the TBLC specimens were as follows: nonspecific interstitial pneumonia in 28 (23.5%), hypersensitivity pneumonitis in 23 (19.3%), drug-induced ILD in 15 (12.6%), idiopathic pulmonary fibrosis in 10 (8.4%), cryptogenic organising pneumonia in 7 (5.9%), connective tissue disease-related ILD in 7 (5.9%), eosinophilic pneumonia in 3 (2.5%) and multi-centric Castleman's disease, pulmonary alveolar proteinosis, sarcoidosis and silicosis in 1 (0.8%) patient each. However, 19 (16.0%) of cases remained as unclassifiable interstitial pneumonia (UCIP) despite the high rate of alveolar-tissue retrieval.

CUSUM learning-curve analysis

As shown in the scatter plot of procedure time per number of biopsies (Fig. 1A), there was a downward trend in procedure time with increasing case numbers. The fitted

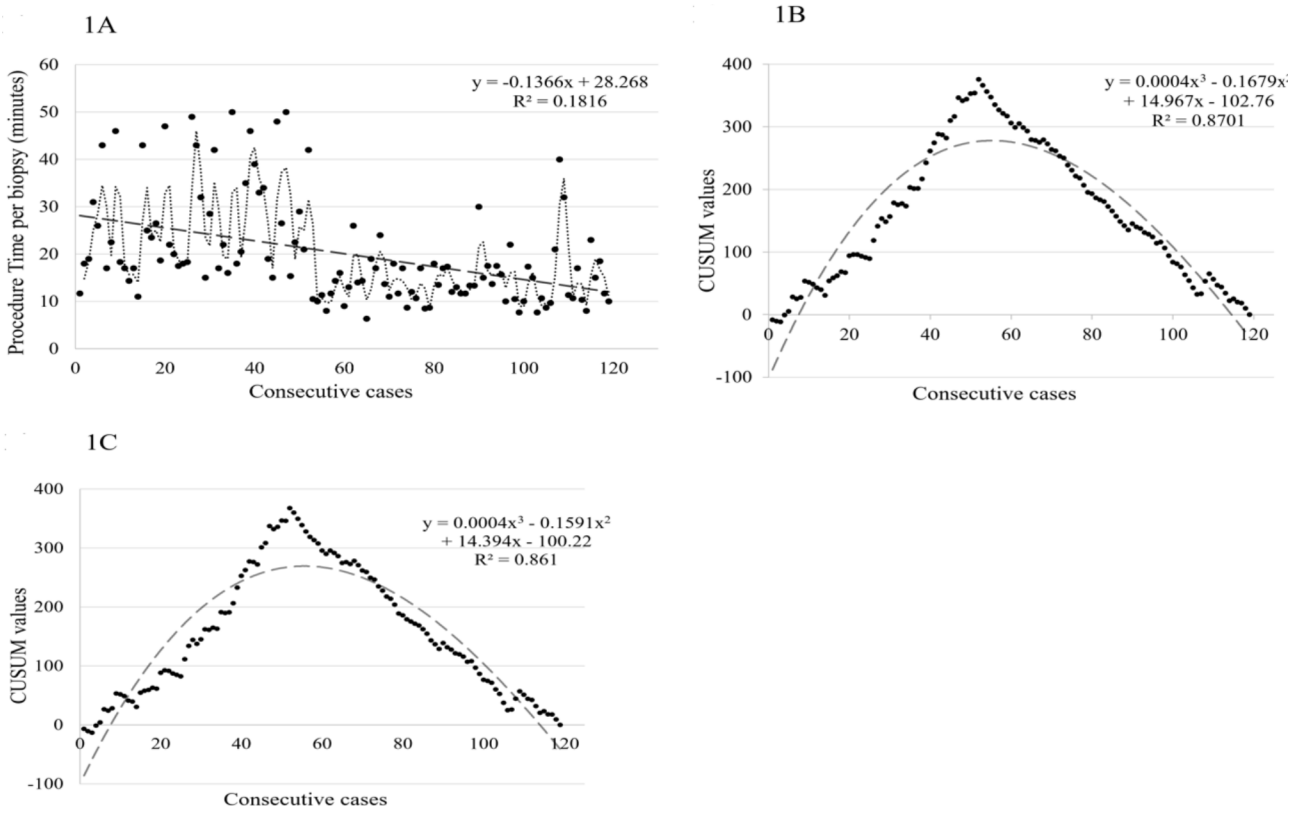

Fig. 1 The trend of the procedural time and CUSUM analyses. **A**: The trend of the procedural time per number of biopsies. **B**: The learning curve (CUSUM chart) of the procedural time per number of biopsies (56 was the vertex of the curve). **C**: The learning curve (RA-CUSUM chart) of the procedural time per number of biopsies (58 was the vertex of the curve)

curve was plotted in the CUSUM analysis of procedure time per number of biopsies (Fig. 1B). The formula for the scatter plot polynomial fitting curve was.

$y=0.0004\times^3-0.1679\times^2+14.967x-102.76$,

where y represented the procedure time per number of biopsies and x represented the number of procedures.

The fit of the curve was good, with a correlation coefficient R^2 = 0.8701. The CUSUM analysis showed that after approximately 56 cases, a change in performance occurred, as evidenced by a reduction in the procedure time per number of biopsies. The RA-CUSUM analysis revealed a similar learning curve pattern with an inflection point at 58 cases (Fig. 1C), closely aligning with the primary CUSUM analysis.

The learning curve was divided into two phases: the initial phase (first 56 cases) and the consolidation phase (remaining 63 cases). Compared with the initial phase, the consolidation phase had a significantly shorter procedure time (35.1±6.9 vs. 42.4±8.9 min, $p<0.001$), higher fentanyl dosage (95.2±11.2 vs. 89.3±16.2 μg, $p=0.020$; 2.78±0.92 vs. 2.36±0.77 μg/kg/h, $p=0.008$), higher midazolam dosage per body weight and time (0.34±0.16 vs. 0.29±0.11 mg/kg/h, $p=0.024$) and more biopsies taken (2.6±0.7 vs. 1.9±0.8, $p<0.001$). No significant differences in patient characteristics, complications, alveolar-tissue retrieval (94.6% vs. 100%, $p=0.101$) or pathological diagnosis (80.4% vs. 81.0%, $p=1.000$) were found between the two phases (Table 2).

Predictors of significant bleeding

In the univariable analysis, male sex [odds ratio (OR) 2.56, 95% CI 1.06−6.17, $p=0.037$] and fentanyl dosage (OR 0.46, 95% CI 0.24−0.87, $p=0.016$) were significantly associated with significant bleeding. In the multivariable analysis, only fentanyl dosage per body weight and time remained a significant predictor of significant bleeding (OR 0.51, 95% CI 0.27−0.97, $p=0.041$) (Table 3).

Subgroup analysis results

In the initial phase, logistic regression analysis revealed a significant association between higher fentanyl dosage and reduced risk of significant bleeding, with an odds ratio of 0.20 (95% CI: 0.05–0.72, $p=0.011$). In the consolidation phase, a similar trend was observed, with an odds ratio of 0.55 (95% CI: 0.25–1.21, $p=0.138$), suggesting that higher fentanyl dosage was associated with lower bleeding risk, although this association did not reach statistical significance.

Table 2 Comparison between initial phase and consolidation phase

Variable	Initial phase (*n* = 56)	Consolidation phase (*n* = 63)	*P*-values
Demographics			
Age (years)	68.3 ± 8.3	68.7 ± 9.6	0.806
Sex, male	27 (48.2)	38 (60.3)	0.202
BMI (kg/m^2)	22.8 ± 4.1	23.9 ± 4.1	0.146
Never smoker	26 (46.4)	28 (44.4)	0.855
Pulmonary Function Tests			
FVC %predicted (%)	80.3 ± 19.2	87.9 ± 24.1	0.073
FEV1/FVC (%)	80.7 ± 13.3	78.5 ± 16.9	0.472
FEV1%predicted (%)	80.3 ± 19.2	87.3 ± 24.5	0.108
DLCO %predicted (%)	73.8 ± 20.2	78.5 ± 21.2	0.266
Procedural Details			
Procedural time (min)	42.4 ± 8.9	35.1 ± 6.9	< 0.001
Fentanyl (μg)	89.3 ± 16.2	95.2 ± 11.2	0.020
Fentanyl (μg/kg/h)	2.36 ± 0.77	2.78 ± 0.92	0.008
Midazolam (mg)	11.2 ± 3.8	12.4 ± 5.7	0.187
Midazolam (mg/kg/h)	0.29 ± 0.11	0.34 ± 0.16	0.024
Number of biopsies	1.9 ± 0.8	2.6 ± 0.7	< 0.001
Biopsy from upper lobes	14 (25.0)	24 (38.1)	0.168
Complications			
Pneumothorax	2 (1.6)	0 (0.0)	0.129
Significant bleeding	12 (21.4)	19 (30.2)	0.303
Respiratory tract infection	2 (3.6)	1 (1.6)	0.601
Histopathological Findings			
Alveolar tissue obtained	53 (94.6)	63 (100.0)	0.101
Pathological diagnosis achieved	45 (80.4)	51 (81.0)	1.000

Data are presented as mean ± SD or n (%). BMI, body mass index; DLCO, diffusing capacity of the lung for carbon monoxide; FEV1, forced expiratory volume in 1 s; FVC, forced vital capacity

Table 3 Predictors of significant bleeding

Variable	Univariable Analysis		Multivariable Analysis	
	OR (95% CI)	*P*-values	OR (95% CI)	*P*-values
Age	0.98 (0.94–1.03)	0.412		
Male	2.56 (1.06–6.17)	0.037	2.12 (0.85–5.28)	0.106
Never smoker	0.48 (0.20–1.13)	0.091		
BMI (kg/m^2)	1.08 (0.97–1.19)	0.151		
FVC %predicted (%)	1.01 (0.99–1.03)	0.440		
FEV1/FVC (%)	1.00 (0.97–1.02)	0.806		
FEV1%predicted (%)	1.00 (0.98–1.02)	0.946		
DL_{CO} %predicted (%)	1.02(0.99–1.05)	0.059		
Number of biopsies	0.91 (0.56–1.47)	0.687		
Biopsy from upper lobes	1.02 (0.43–2.45)	0.964		
Fentanyl (μg/kg/h)	0.46 (0.24–0.87)	0.016	0.51 (0.27–0.97)	0.041
Midazolam (mg/kg/h)	9.65 (0.57–165.0)	0.117		
UIP in histopathology	1.28 (0.41–4.02)	0.667		
Consolidation phase	1.58 (0.69–3.66)	0.281		

BMI, body mass index; CI, confidence interval, DLCO, diffusing capacity of the lung for carbon monoxide; FEV1, forced expiratory volume in 1 s; FVC, forced vital capacity, OR, odds ratio

Post-hoc power analysis results

For the CUSUM analysis comparing procedural times between the initial and consolidation phases, the effect size (Cohen's d) was 0.92. With a significance level of 0.05 (two-tailed), the post-hoc power was calculated to be 99.88%. In the logistic regression analysis of fentanyl dosage predicting significant bleeding, the post-hoc power was 99.94%. Both analyses demonstrated very high statistical power, indicating the study was well-equipped to detect the observed effects.

Discussion

This retrospective study investigated the safety, diagnostic yield and learning curve of TBLC for diagnosing ILDs at a single centre in Japan. The overall diagnostic yield was high, with adequate alveolar tissue obtained in 97.5% of cases. However, a definitive diagnosis could not be reached in 16.0% of patients, who were classified as having UCIP. This finding highlights the limitations of TBLC in providing a definitive diagnosis in some cases, despite the high success rate in obtaining representative lung tissue. The proportion of UCIP cases was higher in our study than in previous studies, suggesting that further refinement of the TBLC technique and standardisation of histopathological interpretation may be necessary to improve diagnostic accuracy [10].

A key finding of our study is the significant improvement in procedural efficiency after the initial 56 cases observed in our CUSUM analysis of the learning curve. The CUSUM analysis showed a reduction in procedure time and an increase in the number of biopsies obtained per case in the consolidation phase, indicating that bronchoscopists became more proficient in performing TBLC over time. The RA-CUSUM analysis, accounting for patient-specific factors, corroborated our primary CUSUM findings. This consistency suggests that the observed improvement in procedural efficiency is robust and not merely a result of patient characteristic variations. This finding is in a similar range to the study by Almeida et al., which reported a learning curve plateau

after approximately 70 procedures [7]. Almeida's study assessed the learning curve using diagnostic yield, sample length, and sample area. In contrast, Davidsen et al. used CUSUM analysis for diagnostic yield and complications, finding satisfactory performance throughout their study without a clear learning curve [10]. These diverse approaches collectively enhance our understanding of proficiency development in TBLC performance.

Another significant finding of our study is the association between fentanyl dosage and the risk of bleeding during TBLC. In the multivariable analysis, lower fentanyl dosage per body weight and time was found to be a significant predictor of bleeding complications. This novel finding suggests that adequate sedation with fentanyl may have a crucial role in reducing the risk of bleeding during TBLC, which is consistent with recent reviews recommending the use of midazolam in combination with short-acting opioids for their beneficial cough suppression effects [11, 12]. The mechanism behind this association may be related to fentanyl's ability to minimise patient movement and coughing during the procedure, thereby reducing the risk of trauma to the lung tissue and blood vessels [13–15]. Additionally, fentanyl has been shown to attenuate the haemodynamic response to tracheal intubation, which may further contribute to its protective effect against bleeding during TBLC [16–18]. Our results highlight the importance of optimising sedation protocols for TBLC and provide a basis for further research into the role of sedative medications, particularly opioids, in reducing complications during the procedure.

The safety profile of TBLC in our study was comparable to that reported in previous studies, with a low incidence of pneumothorax (1.6%) and no procedure-related deaths. The rate of significant bleeding (26.9%) was similar to that reported in a recent meta-analysis, suggesting that our results are generalisable to other centres performing TBLC [4]. However, the risk of bleeding remains a concern with TBLC, and our findings regarding the association between fentanyl dosage and bleeding risk may have important implications for the management of patients undergoing this procedure.

This study had several limitations that should be considered when interpreting the results. First, the retrospective nature of the study may have introduced selection bias and limited the ability to control for confounding factors. A prospective study design would enable more robust data collection and analysis. Second, the single-centre setting may not reflect the experience and outcomes of other institutions with different patient populations, procedural protocols and operator experience. The small sample size ($n=119$) may not provide a comprehensive representation of the complications and outcomes associated with TBLC. However, our post-hoc power analysis demonstrated high statistical power for our main findings, suggesting that the study was adequately powered to detect the observed effects despite the relatively small sample size. Nevertheless, larger prospective studies would be valuable to confirm these findings and potentially identify rarer complications or more subtle effects. Third, we were unable to adjust for operator experience as a potential confounding factor in the association between fentanyl dosage and bleeding risk. Procedures were performed by various doctors, and we lacked detailed records linking specific operators to each procedure. Since proficiency in TBLC requires specialized training beyond years of medical experience, it's possible that more experienced teams both administered higher doses of fentanyl and performed the procedures more safely, independent of fentanyl's pharmacological effects. This confounding could contribute to the observed association between higher fentanyl dosage and reduced bleeding risk. Future prospective studies with detailed tracking of operator experience and standardized sedation protocols are needed to clarify this relationship. Finally, the relatively short follow-up period (30 days) may not capture delayed complications and long-term outcomes associated with TBLC. Extending the follow-up period would provide a more comprehensive assessment of the safety and efficacy of this diagnostic modality.

In conclusion, our study demonstrated that TBLC is a safe and effective diagnostic tool for ILDs, with a high success rate in obtaining adequate lung tissue for histopathological analysis. However, the significant proportion of unclassifiable cases highlights the need for further refinement of the TBLC technique and standardisation of histopathological interpretation to improve diagnostic accuracy. The learning-curve analysis using the CUSUM method provides valuable insights into the development of procedural efficiency and competency for bronchoscopists performing TBLC and emphasises the importance of ongoing performance monitoring to ensure the maintenance of skills over time. Finally, our novel finding of an association between fentanyl dosage and bleeding risk, possibly related to its ability to minimise patient movement and attenuate haemodynamic responses, suggests that optimising sedation protocols may be an important strategy for reducing complications during TBLC. However, due to the inability to adjust for operator experience, this association should be interpreted with caution. Future studies should focus on validating this association and exploring the mechanisms underlying the protective effect of fentanyl on bleeding risk during TBLC.

Abbreviations

TBLC	Transbronchial lung cryobiopsy
ILD	Interstitial lung disease
SLB	Surgical lung biopsy

CUSUM	Cumulative sum
HRCT	High-resolution computed tomography
FVC	Forced vital capacity
FEV1	Forced expiratory volume in 1 s
UCIP	Unclassifiable interstitial pneumonia
OR	Odds ratio
CI	Confidence interval

Acknowledgements
The authors would like to thank Enago (www.enago.jp) for the professional English language review. We acknowledge the use of Anthropic's AI assistant, Claude, for aiding in the drafting and editing of this manuscript. However, the final content and conclusions remain the sole responsibility of the authors.

Author contributions
Shota Kaburaki: Conceptualization (lead); Data Curation (lead); Formal Analysis (lead); Investigation (lead); Methodology (lead); Resources (equal); Visualization (lead); Writing – Original Draft (lead). Toru Tanaka: Data Curation (equal); Investigation (equal); Supervision (equal); Writing – Review & Editing (equal). Koichiro Kamio: Project Administration (equal); Supervision (equal); Writing – Review & Editing (equal). Yosuke Tanaka: Writing – Review & Editing (equal). Kazuo Kasahara: Writing – Review & Editing (equal). Masahiro Seike: Conceptualization (equal); Methodology (equal); Project Administration (lead); Resources (lead); Supervision (lead); Writing – Original Draft (supporting); Writing – Review & Editing (lead).

Funding
None.

Data availability
The data that support the findings of this study are available from the corresponding author upon reasonable request.

Declarations

Competing interests
The authors declare no competing interests.

Conflict of interest
None of the authors has a conflict of interest to declare.

Human/Animal Ethics approval declaration
The Nippon Medical School Hospital Institutional Review Board approved this study (approval number B-2024-875) and waived informed consent due to its retrospective nature. Clinical trial number: not applicable. This study was conducted in accordance with the principles of the Declaration of Helsinki.

Consent to participate
Informed consent was waived by the Nippon Medical School Hospital Institutional Review Board due to the retrospective nature of this study.

Clinical Trial Registration
Clinical trial number: not applicable.

Received: 11 August 2024 / Accepted: 21 October 2024
Published online: 29 October 2024

References

1. Durheim MT, Kim S, Gulack BC, Burfeind WR, Gaissert HA, Kosinski AS, et al. Mortality and respiratory failure after thoracoscopic lung biopsy for interstitial lung disease. Ann Thorac Surg. 2017;104:465–70.
2. Han Q, Luo Q, Xie J-X, Wu L-L, Liao L-Y, Zhang X-X, et al. Diagnostic yield and postoperative mortality associated with surgical lung biopsy for evaluation of interstitial lung diseases: a systematic review and meta-analysis. J Thorac Cardiovasc Surg. 2015;149:1394–e4011.
3. Koslow M, Edell ES, Midthun DE, Mullon JJ, Kern RM, Nelson DR, et al. Bronchoscopic Cryobiopsy and forceps Biopsy for the diagnostic evaluation of diffuse parenchymal lung disease in clinical practice. Mayo Clin Proc Innov Qual Outcomes. 2020;4:565–74.
4. Kheir F, Uribe Becerra JP, Bissell B, Ghazipura M, Herman D, Hon SM, et al. Transbronchial Lung Cryobiopsy in patients with interstitial lung disease: a systematic review. Ann Am Thorac Soc. 2022;19:1193–202.
5. DiBardino DM, Haas AR, Lanfranco AR, Litzky LA, Sterman D, Bessich JL. High complication rate after introduction of Transbronchial Cryobiopsy into Clinical Practice at an Academic Medical Center. Ann Am Thorac Soc. 2017;14:851–7.
6. Lin P-L, Zheng F, Shin M, Liu X, Oh D, D'Attilio D. CUSUM learning curves: what they can and can't tell us. Surg Endosc. 2023;37:7991–9.
7. Almeida LM, Lima B, Mota PC, Melo N, Magalhães A, Pereira JM, et al. Learning curve for transbronchial lung cryobiopsy in diffuse lung disease. Rev Port Pneumol. 2017. https://doi.org/10.1016/j.rppnen.2017.09.005.
8. Ravaglia C, Bonifazi M, Wells AU, Tomassetti S, Gurioli C, Piciucchi S, et al. Safety and Diagnostic Yield of Transbronchial Lung Cryobiopsy in diffuse Parenchymal Lung diseases: a comparative study versus video-assisted thoracoscopic lung biopsy and a systematic review of the literature. Respiration. 2016;91:215–27.
9. Xiao Y, Liang M, Chen M, Li Z, Xia T, Yue X, et al. Evaluating the learning curve of high intensity focus ultrasound for breast fibroadenoma by CUSUM analysis: a multi-center study. Int J Hyperth. 2022;39:1238–44.
10. Davidsen JR, Skov IR, Louw IG, Laursen CB. Implementation of transbronchial lung cryobiopsy in a tertiary referral center for interstitial lung diseases: a cohort study on diagnostic yield, complications, and learning curves. BMC Pulm Med. 2021;21:67.
11. Wahidi MM, Jain P, Jantz M, Lee P, Mackensen GB, Barbour SY, et al. American College of Chest Physicians consensus statement on the use of topical anesthesia, analgesia, and sedation during flexible bronchoscopy in adult patients. Chest. 2011;140:1342–50.
12. José RJ, Shaefi S, Navani N. Sedation for flexible bronchoscopy: current and emerging evidence. Eur Respir Rev. 2013;22:106–16.
13. Greig JH, Cooper SM, Kasimbazi HJ, Monie RD, Fennerty AG, Watson B. Sedation for fibre optic bronchoscopy. Respir Med. 1995;89:53–6.
14. Stolz D, Chhajed PN, Leuppi JD, Brutsche M, Pflimlin E, Tamm M. Cough suppression during flexible bronchoscopy using combined sedation with midazolam and hydrocodone: a randomised, double blind, placebo controlled trial. Thorax. 2004;59:773–6.
15. Houghton CM, Raghuram A, Sullivan PJ, O'Driscoll R. Pre-medication for bronchoscopy: a randomised double blind trial comparing alfentanil with midazolam. Respir Med. 2004;98:1102–7.
16. Choi B-H, Lee Y-C. Effective Bolus dose of Sufentanil to Attenuate Cardiovascular responses in Laryngoscopic double-lumen endobronchial intubation. Anesth Pain Med. 2016;6:e33640.
17. Hashemian AM, Zamani Moghadam Doloo H, Saadatfar M, Moallem R, Moradifar M, Faramarzi R, et al. Effects of intravenous administration of fentanyl and lidocaine on hemodynamic responses following endotracheal intubation. Am J Emerg Med. 2018;36:197–201.
18. Adachi YU, Satomoto M, Higuchi H, Watanabe K. Fentanyl attenuates the hemodynamic response to endotracheal intubation more than the response to laryngoscopy. Anesth Analg. 2002;95:233–7. table of contents.

Publisher's note