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Perioperative comparison of uniportal versus multiportal video-assisted thoracoscopic surgery for complex segmentectomy of the lower lung lobe

Xinyu Wang^{1†}, Yuming Wang^{2†}, Min cao^{1†}, Yujie Fu¹, Wenbiao Pan¹, Qing Ye¹, Xiaojing Zhao^{1*} and Zhiyong Sun^{1*}

Abstract

Background Resection of basal segmentectomy through uniportal video-assisted thoracoscopic surgery (U-VATS) is technically challenging for thoracic surgeons. Compared with multiportal VATS (M-VATS), the safety and feasibility of U-VATS for complex segmentectomy of lower lung lobe need further validation. In this study, we aimed to compare the perioperative outcomes of U-VATS with M-VATS in the treatment of complex segmentectomy of lower lung lobe for stage IA lung cancer.

Methods We conducted a retrospective cohort study of 168 patients (116 U-VATS and 52 M-VATS) undergoing complex lower lobe segmentectomy for stage IA NSCLC from January 2021 to May 2023. The demographics of the enrolled patients were collected and propensity score matching (PSM) was used to reduce the heterogeneity of baseline characteristics. Perioperative outcomes were compared between the two groups.

Results After matching, 50 cases were yielded in each group. There was no 30-day postoperative mortality and conversion to open in both groups. The U-VATS exhibited shorter postoperative hospital stays (P=0.034) and a trend toward reduced postoperative drainage (P=0.081) compared to the M-VATS group. Pain score on postoperative day 2 in the U-VATS group was lower than M-VATS group (P=0.004). There were no significant differences in resection margins, operation time and postoperative complications between the two groups.

Conclusions U-VATS provides comparable perioperative safety and efficacy to M-VATS for complex lower lobe segmentectomy, with advantages in accelerated recovery and reduced postoperative pain. U-VATS complex segmentectomy of lower lung lobe is a safe and feasible technique for experienced thoracic surgeons, which deserves support and popularity.

Keywords Uniportal VATS, Multiportal VATS, Complex segmentectomy, Lower lung lobe

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Introduction

Lobectomy has been a standard surgical procedure for patients with peripheral T1N0 non-small cell lung cancer (NSCLC) based on the prospective, multi-institutional randomized trial conducted by the Lung Cancer Study Group (LCSG) in 1995 [1]. High-resolution computer tomography (HRCT), as an alternative to chest x-ray, has detected more lung ground-glass nodules (GGNs) [2]. Compared with lung solid nodules, GGNs exhibit more indolent biological behaviours [3]. The Japan Clinical Oncology Group (JCOG) has released the outcomes of a series of randomized trials, including JCOG0804 and JCOG0802, proving that sublobar resection is superior than lobectomy in overall survival for patients with clinical stage IA lung cancer with tumor diameter ≤ 2 cm [4, 5]. Therefore, the indications for sublobar resection are now extended for small-sized early lung cancer (tumor diameter ≤ 2 cm).

Since Gonzalez Rivas first reported uniportal video-assisted thoracoscopic surgery (U-VATS) lobectomy and segmentectomy [6, 7], U-VATS has developed rapidly in recent decades. Compared with multiportal VATS (M-VATS), U-VATS is technically more complex because of limited space and angle of single-incision [8]. With improvements in surgical skills, U-VATS segmentectomy is gradually recognized and accepted by thoracic surgeons. A meta-analysis conducted by Xiang and colleagues found that U-VATS segmentectomy had a shorter postoperative hospital stay, lower postoperative pain scores, fewer days of chest tube drainage and a smaller wound than M-VATS segmentectomy [9].

VATS segmentectomy could be categorized into simple and complex segmentectomy based on the surgical procedures and intersegmental planes. Simple segmentectomy such as S6 resection, or left lingula segments (L4+5) resection, et al., which could be completed by creating only one intersegmental plane. Complex segmentectomy, for instance, S3 resection or S9 resection, which needs more complicated procedures by creating multiple intersegmental planes [10]. Instrument conflict and visualization limitation via one incision not only heighten the technical complexity but also pose significant challenges to surgical safety for complex segmentectomies.

Although several studies have compared uniportal and multiportal approaches in thoracoscopic complex segmentectomy, the majority of reported cases involve upper lobe resections, with lower lobe complex segmentectomies remaining underrepresented in the current literature [8, 11]. Complex segmentectomy of lower lung lobe appears more technically challenging than other segmentectomies due to the deeper hilar structures, more prevalent variations of vessels and bronchi and more complex anatomy of intersegmental planes [12, 13]. To be best of our knowledge, there is no exclusive study

concerning the comparison the uniportal and multiportal VATS for complex segmentectomy of lower lung lobe. Following our previous study confirming the feasibility of thoracoscopic complex segmentectomy in the lower lobe [14], we have further investigated the impact of surgical incisions on lower lobe complex segmentectomies. In this study, we aimed to compare the perioperative outcomes of U-VATS with M-VATS in the treatment of complex segmentectomy of lower lung lobe for stage IA lung cancer.

Methods

Patient selection

Figure 1 showed the patient selection scheme. We retrospectively analyzed consecutive patients who underwent video-assisted thoracoscopic segmentectomy of lower lung lobe between January 2021 and May 2023 at Shanghai Renji hospital. The inclusion criteria included: (I) peripheral GGNs located at the outer half of the lung parenchyma; (II) clinical stage IA with tumor diameter ≤ 2 cm; (III) general condition and respiratory function ad equate for segmentectomy; (IV) lung nodules located in lower lung lobe. In total, 386 cases of segmentectomy of lower lung lobe were included. Complex segmentectomy of lower lung lobe was defined as all segmentectomy other than S6 and S8+9+10 resection, including S7 (right), S8, S9, S10, S* (Subsuperior segment) and combined resection of basal segmentectomy (S8+9, S9+10, et al.). Complex segmentectomy of lower lung lobe was further screened by excluding the following the criteria: (I) simple segmentectomy of lower lung lobe (n=184); (II) concurrent bilateral surgery (n=8); (III) incomplete medical records (n=4); (IV) benign GGN confirmed by pathology (n=22). The demographics of patients such as age, sex, body mass index (BMI), tumor size on HRCT, histology type, smoking status, comorbidity, ratio of forced expiratory volume in 1s to forced vital capacity (FEV1%), ratio of diffusion lung capacity for carbon monoxide (DLCO%) and ratio of consolidation-totumor (CTR) were recorded. Written informed consent was obtained from all patients before surgery.

VATS segmentectomy procedure

Preoperative CT scan with a slice thickness of 1.0 mm was performed to evaluate the neighboring relationships of vessels and bronchi for All patients. Patients were positioned laterally following dual-lumen endotracheal intubation. U-VATS or M-VATS was decided by a combination of patient-specific factors and intraoperative situation. All operations were performed by four experienced operating surgeons (XJ Zhao, YJ Fu, Q Ye and WB Pan) with at least 5 years of experience in both U-VATS and M-VATS, assisted by two skilled thoracic surgeons. For the U-VATS group, a single 3.5–4.0 cm skin incision

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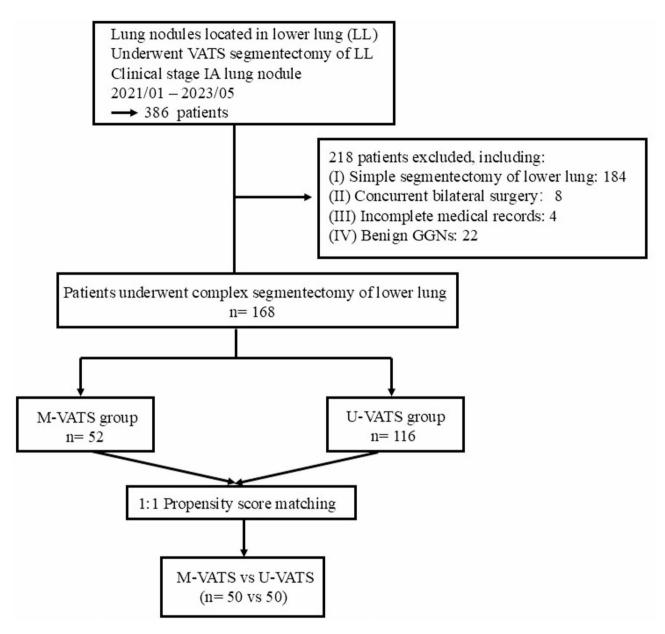


Fig. 1 Selection algorithm of the patients in this study

was made in the 4th or 5th intercostal space along the anterior axillary line. For M-VATS group, two additional 1.5 cm ports were inserted through the eighth intercostal space (midaxillary line and posterior axillary line) to assist the surgery. Figure 2 depicts the incision configurations and drain placement sites via U-VATS and M-VATS approaches.

All procedures were performed through an interlobar fissure approach or an inferior pulmonary ligament approach depend on the forms of interlobar fissures (complete or incomplete). Usually, S7 and S8 segmentectomy were performed via the interlobar fissure approach while S9 and S10 segmentectomy were performed via the inferior pulmonary ligament approach. After separating

the structures of target segment, blood vessels were ligated by the stapler (Endo-GIA, Covidien, USA, or ECHELON Flex, Ethicon Endo Surgery, USA) or hemo-lock while the bronchi were transected by the stapler. Intraoperative images of U-VATS and M-VATS segmentectomy of lower lung lobe were shown in Supplementary information.

The inflation-deflation method was used for delineating the intersegmental plane and subsequently resecting the target segment [15]. The resection margin of each nodule was examined by measuring the distance between lesion and resection margin. An extra wedge resection would be performed for the security of surgical margin if resection margin less than 2 cm or the diameter of the nodule.

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 $\textbf{Fig. 2} \ \ \textbf{The incision configurations via U-VATS and M-VATS approaches}$

After removing the specimens, mediastinal lymph node sampling or dissection was performed for primary lung cancer. Lymph node sampling was performed for GGNs with CTR≤0.25 while lymph node dissection was performed for GGNs with CTR>0.25. In this study, both groups received a consistent analgesic regimen. At the end of surgery, intercostal nerve blocks were performed by injecting 10 ml of 2% lidocaine at the T5-T8 intercostal levels. Two chest tubes were placed to drain the air (20 F chest tube, via main incision) and pleural effusion (12 F pigtail catheter, via 7th or 8th intercostal space on posterior axillary line) at the end of each surgery.

Postoperative management and evaluation of complications

All patients were cared by the same nursing team. Wound pain score and drainage volume were recorded every day until the patient was discharged. The 11-point numeric rating scale (NRS), which range from 0 (no pain) to 10 (worst pain), was used to reflect the wound pain intensity. NRS could be further categorized as: 0 'no pain', 1–3 'mild pain', 4–6 'moderate pain', and 7–10 'severe pain'. Postoperatively, all patients were administered parecoxib 40 mg intravenously every 12 h as routine analgesia. If

postoperative pain was moderate or severe (If NRS>3), an additional dose of a more potent rescue analgesic (e.g., tramadol 50 mg intravenously) was administered. Low-dose computer tomography (LDCT) was routinely experienced on postoperative day 2 (POD 2) to evaluate the pleural effusion or air. The traditional thoracic drainage system for chest tubes with a suction pressure of – 12 cmHev $_2$ O was routinely used after lung resection. The chest tubes were removed once there was no air leak and daily drainage of chest tube was < 300 mL.

Evaluation of perioperative outcomes

Perioperative outcomes included operation time, resection margins, postoperative drainage, postoperative hospital stay and postoperative complications (such as prolonged air leakage, pneumonia/atelectasis, hemoptysis and residual pleural space) were recorded. Prolonged air leakage was defined as air leakage continuing for 5 days after surgery [16]. Residual pleural space was defined as radiological pneumothorax larger than 20% [17].

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Statistical analyses

Statistical analyses were performed by SPSS 27.0 software (SPSS, Inc., Chicago, IL, USA). To minimize bias, 1:1 propensity score matching analysis (caliper = 0.2) was performed between the M-VATS and U-VATS group. Propensity scores were calculated by the following variables: age, sex, BMI, comorbidity, smoking status, FEV1%, DLCO%, tumour size and CTR. Continuous variables are presented as mean ± standard deviation (SD) while categorical variables are presented as numbers and percentages. Chi-square, Fisher exact or Student t test was used to compare differences between variables. Two-sided P values < 0.05 were considered statistically significant.

Results

Patient characteristics

The baseline characteristics of all eligible patients were shown in Table 1. A total of 168 patients in the two groups were finally included, including 52 patients in the M-VATS group and 116 patients in the U-VATS group. Before matching, M-VATS group had higher BMI $(23.3\pm2.8 \text{ vs. } 22.1\pm3.1, p=0.022)$, a tendency of older age $(60.3\pm12.2 \text{ vs. } 56.2\pm14.0, p=0.069)$ and larger tumor size $(13.0\pm3.8 \text{ vs. } 11.6\pm5.1, p=0.074)$ than U-VATS group. After matching, 50 cases were yielded in each

group. The two groups were more comparable in terms of baseline characteristics. The position and distribution of the complex segmentectomy of lower lung lobes were listed in Table 2.

Perioperative outcomes

Perioperative data were shown in Table 3. There were no 30-day postoperative mortality or conversion to open surgery occurred in either group. One patient in the U-VATS group was converted to M-VATS due to the severe pleural adhesion. The histology type in the M-VATS group included 29 (58.0%) invasive adenocarcinomas, 18 (36.0%) non-invasive adenocarcinomas and 3 (6.0%) other types (squamous cell carcinoma and metastatic carcinoma), while the corresponding numbers in the U-VATS group were 33 (66.0%), 16 (32.0%) and 1 (2.0%). Forty-nine cases (98.0%) in the M-VATS group and 50 cases (100.0%) in the U-VATS group had adequate resection margins by anticipated segmentectomy. The overall postoperative complication rate in the U-VATS group was 16.0% (8/50), comparable to that in the M-VATS group (18.0%, 9/50; p = 1.000). The U-VATS group had shorter postoperative hospital stays than the M-VATS group $(3.8 \pm 1.1 \text{ vs. } 4.3 \pm 1.5 \text{ days, } P = 0.034).$ The U-VATS group seemed to have less postoperative

Table 1 The demographic data of patients in uniportal and multiportal group

Values	Before matching			After matching		
	M-VATS	U-VATS	p Value	M-VATS	U-VATS	p Value
	(n = 52)	(n=116)		(n = 50)	(n=50)	
Age (years)	60.3 ± 12.2	56.2 ± 14.0	0.069	59.9 ± 12.3	60.0 ± 12.5	0.994
Sex			0.51			0.523
Male	16 (30.8%)	30 (25.9%)		15(30.0%)	18(36.0%)	
Female	36 (69.2%)	86 (74.1%)		35(70.0%)	32(64.0%)	
Pulmonary function						
FEV1(%)	82.6 ± 7.1	84.0 ± 6.8	0.231	82.8 ± 7.0	83.3 ± 6.7	0.695
DLCO(%)	82.4 ± 10.6	81.4 ± 7.4	0.526	81.6 ± 9.7	81.3 ± 7.1	0.866
Nodule size on HRCT (mm)	13.0 ± 3.8	11.6 ± 5.1	0.074	12.9 ± 3.9	12.6 ± 5.2	0.719
CTR (%)			0.763			0.839
0 to ≤ 25	21(40.4%)	44(37.9%)		21(42.0%)	20(40.0%)	
25 to ≤ 100	31(59.6%)	72(62.1%)		29(58.0%)	30(60.0%)	
BMI (kg/m ²)	23.3 ± 2.8	22.1 ± 3.1	0.022	23.2 ± 2.7	23.4 ± 3.2	0.717
Smoking status, n (%)			0.296			0.806
Current or ever	11 (21.2%)	17 (14.7%)		10(20.0%)	11(22.0%)	
Never	41 (78.8%)	99 (85.3%)		40(80.0%)	39(78.0%)	
Comorbidity			0.199			0.832
Yes	18(34.6%)	29(25.0%)		16(32.0%)	17(34.0%)	
No	34(65.4%)	87(75.0%)		34(68.0%)	33(66.0%)	

HRCT = High-resolution computer tomography

M-VATS = Multiportal video-assisted thoracoscopic surgery

U-VATS = Uniportal video-assisted thoracoscopic surgery

FEV1(%) = Ratio of forced expiratory volume in 1s to forced vital capacity

DLCO(%) = Ratio of diffusion lung capacity for carbon monoxide

BMI = Body mass index

CTR = Ratio of consolidation-to-tumor

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Table 2 The position and distribution of the complex segmentectomies of lower lung

Surgery types	M-VATS group	U-VATS group	
	(n=52)	(n=116)	
Right lower lobe	34 (65.4%)	62 (53.4%)	
S7	4	1	
S8	6	21	
S9	6	8	
S10	4	12	
S8+9	3	6	
S9+10	7	5	
S6+10a	1	1	
S6+9+10	1		
S6+9	1		
S7 + 10	1		
S7+8		1	
S8a		1	
S8b		1	
S9a		2	
S*+6b+8a		1	
S8a+9a+6b		1	
S9a + 10a		1	
Left lower lobe	18 (34.6%)	54 (46.6%)	
S8	1	23	
S9	1	6	
S10	7	12	
S8+9	2	8	
S9+10	7	2	
S*		1	
S6+8		1	
S6+10		1	

S*: Subsuperior segment

drainage than M-VATS group, although without statistical difference (504.7 ± 238.8 vs. 613.3 ± 363.5 ml, P=0.081). Pain score on POD2 of patients in the U-VATS group was lower than that in the M-VATS group (2.3 ± 1.0 vs. 3.1 ± 1.4 , P=0.004) while POD1 and POD3 showed no statistical difference (P=0.706, P=0.185). The operation time of the U-VATS group was 126.2 ± 26.4 min while that in the M-VATS group was 123.3 ± 22.5 min (P=0.550). In addition, postoperative complications including prolonged air leakage, pneumonia/ atelectasis, hemoptysis and residual pleural space were comparable between the two groups (all P>0.05).

Discussion

Uniportal VATS has been increasingly utilized in lung cancer resections since 2004, demonstrating safety and feasibility in selected patients with advantages in reducing hospitalized time, minimizing injury to the chest wall and intercostal nerves, as well as relieving postoperative pain [18–20]. In recent years, accumulating evidence indicate that U-VATS has comparable perioperative outcomes with M-VATS. With improvements of surgical

Table 3 Perioperative outcomes of uniportal and multiportal aroup

<u>9.04p</u>	NA 1/ATC	III VATC	
Values	M-VATS	U-VATS	p
	group (n = 50)	group (n = 50)	Value
Operation time (min)	123. 3 ± 22.5	126.2 + 26.4	0.55
Adequate resection margins	49 (98.0%)	50 (100.0%)	1
Histology type			0.491
Invasive adenocarcinoma	29 (58.0%)	33 (66.0%)	
Non-invasive	18 (36.0%)	16 (32.0%)	
adenocarcinoma			
Others	3 (6.0%)	1 (2.0%)	
Postoperative drainage (ml)	613.3 ± 363.5	504.7 ± 238.8	0.081
Postoperative hospital stay	4.3 ± 1.5	3.8 ± 1.1	0.034
(days)			
Postoperative pain (NRS)			
POD1	2.8 ± 1.1	2.9 ± 1.0	0.706
POD2	3.1 ± 1.4	2.3 ± 1.0	0.004
POD3	2.0 ± 0.8	1.8 ± 0.7	0.185
Postoperative complications	9 (18.0%)	8 (16.0%)	1
Prolonged air leakage	4 (8.0%)	2 (4.0%)	0.674
Pneumonia/Atelectasis	3 (6.0%)	4 (8.0%)	1
Hemoptysis (> 10 ml)	2 (4.0%)	2 (4.0%)	1
Residual pleural space	1 (2.0%)	2 (4.0%)	1

NRS = Numerical rating scale

POD = Postoperative day

skills, the indication of U-VATS has extended to lung segmentectomy. However, few studies have reported the values of U-VATS in basal segment resection. Several reasons may contribute to it. Firstly, basal segment has deeper intraparenchymal localization of the hilar structures [12]. Secondly, the anatomic variation of lower pulmonary structure is more prevalent than that of upper lung, making it difficult to identify target blood vessels [13, 21]. Thirdly, the intersegmental planes of basal segment are more complex so that it is tough to tailor the intersegmental planes through one port.

One of the major challenges with U-VATS is whether it could guarantee the safety of surgery. In our study, there was no mortality and conversion to open surgery in the two groups. In addition, postoperative complications were comparable between the two groups, proving that U-VATS was safe and feasible. Another challenge with U-VATS is oncological adequacy [11]. Our results demonstrated that all cases in the U-VATS group had adequate resection margins, indicating oncological adequacy was not compromised by U-VATS.

Instrument maneuverability and conflict are another challenge for U-VATS. The confined single-incision workspace increases the risk of instrument collision, particularly during complex segmentectomies. To mitigate this, we employ curved instruments (for example, curved suction and curved tip articulating reload) to decrease the crashing and expose the space [22]. In

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addition, visualization limitation is also a challenge during U-VATS. Single-port access restricts the ability to triangulate the camera and instruments, potentially obscuring critical structures like intersegmental veins. We address this by utilizing 30° high-definition thoracoscopes and fixing the camera on the top of the incision to release more space for operating surgeon.

Previous studies have reported conflicting results regarding operative time, with some indicating that U-VATS reduces operative duration and others demonstrating no significant difference [8, 11]. In our study, the operation time of the U-VATS group was similar to M-VATS group $(126.2 \pm 26.4 \text{ min vs. } 123.3 \pm 22.5 \text{ min}).$ Frequent interference among instruments during the surgery and tailor of intersegmental planes are timeconsuming. The four thoracic surgeons who performed the operations in our center were experienced in both U-VATS and M-VATS, and we believed that skill differences among the surgeons were not a confounding factor. From our perspective and experience, the following suggestions may be helpful. First, the camera assistant should be a dedicated, trained professional to ensure effective cooperation with the surgeon. Second, when spatial or angular constraints hinder the safe passage of a stapler device around the vessel, employing Hem-o-Lock or silk thread ligation for the target segmental vessels becomes a safer and more effective alternative under these circumstances. Finally, a "periphery to center" strategy (tailoring from periphery to center) is beneficial for demarcating intersegmental planes with a stapler.

One significant strength of U-VATS is that it causes less surgical trauma and reduces postoperative pain. Hirai K et al. revealed that U-VATS lobectomy could reduce the postoperative wound pain and the number of days analgesic agents than M-VATS lobectomy [23]. Tian and colleagues found that U-VATS segmentectomy reduced postoperative pain at 24, 48, 72 h after surgery, compared with the M-VATS group, confirming that U-VATS could reduce pain during the early postoperative period in lung segmentectomy [24]. In this study, we also observed that the U-VATS group had lower postoperative pain scores than those in the M-VATS group, especially on POD2, which was consistent with previous studies. The potential explanation may be that U-VATS could reduce intercostal nerve injury, therefore causing less surgical trauma. The pain scores in the M-VATS group on POD2 are slightly higher than on POD1, which may be attributed to delayed inflammatory responses and prolonged anesthetic metabolism.

Existing literature has shown conflicting results regarding the effects of U-VATS verse M-VATS on chest tube drainage and hospital stay. Some researchers found that U-VATS had less chest tube drainage and shorter post-operative hospital stays [11, 25], while others reported

no significant differences [24]. These discrepancies might be attributed to differences in postoperative care strategy and discharge criteria. In our study, we found U-VATS had shorter postoperative hospital stays (P=0.034) and a tendency toward less postoperative drainage (P=0.081) than those in the M-VATS group. Less surgical trauma caused by U-VATS may be responsible for it.

This study has limitations inherent to its retrospective, single-center design, including potential selection bias and unmeasured confounders. Additionally, long-term oncologic outcomes require further investigation. Future multicenter randomized trials are needed to validate these findings.

Conclusions

In conclusion, U-VATS achieves perioperative outcomes comparable to M-VATS for complex lower lobe segmentectomy, with statistically significant advantages in postoperative recovery and pain management. For experienced thoracic surgeons, U-VATS represents a safe and feasible minimally invasive strategy, meriting broader clinical implementation.

Abbreviations

NSCLC Non-small cell lung cancer

HRCT High-resolution computer tomography LDCT Low-dose computer tomography

GGNs Ground-glass nodules
JCOG Japan Clinical Oncology Group
LCSG Lung Cancer Study Group

U-VATS Uniportal video-assisted thoracoscopic surgery
M-VATS Multiportal video-assisted thoracoscopic surgery

POD Postoperative day NRS Numeric rating scale

FEV1(%) Ratio of forced expiratory volume in 1s to forced vital capacity

DLCO(%) Ratio of diffusion lung capacity for carbon monoxide

BMI Body mass index

CTR Ratio of consolidation-to-tumor

Supplementary Information

The online version contains supplementary material available at https://doi.or q/10.1186/s12893-025-02944-3.

Supplementary Material 1: Supplementary Fig.1. Left S8+9 VATS segmentectomy by uniportal VATS. (A) preoperative CT scan, (B) Branches of pulmonary artery, (C) Branches of bronchus, and (D) Branches of pulmonary vein of the target segment from the view of uniportal VATS

Supplementary Material 2: Supplementary Fig.2. Right S8 VATS segmentectomy by multiportal VATS. (A) preoperative CT scan, (B) Branches of pulmonary artery, (C) Branches of bronchus, and (D) Branches of pulmonary vein of the target segment from the view of multiportal VATS

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No.

Author contributions

(I) Conception and design: Zhiyong Sun and Xiaojing Zhao; (II) Administrative support: Yujie Fu; (III) Provision of study materials or patients: Wenbiao Pan and Qing Ye; (IV) Collection and assembly of data: Xinyu Wang, Yuming Wang; (V) Data analysis and interpretation: Min Cao; (VI) Manuscript writing: Xinyu Wang; (VII) Manuscript revision and final approval of manuscript: All authors.

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Data availability

The data that support the findings of this study are available on request from the corresponding author.

Declarations

Ethics approval and consent to participate

This study was approved by the Ethics Committee of the Renji Hospital affiliated to Shanghai Jiao Tong University School of Medicine and was performed in accordance with the Declaration of Helsinki. All methods were carried out in accordance with relevant guidelines and regulations. Prior written informed consent was obtained from all study participants.

Consent for publication

Not applicable.

Competing interests

The authors declare no competing interests.

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