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ORIGINAL ARTICLE / ÖZGÜN MAKALE

Anterior mediastinal tumor surgery applying single-port thoracoscopy using the subxiphoid approach

Subksifoidal yaklaşım kullanılarak tek port torakoskopi uygulanan anterior mediastinal tümör cerrahisi

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ABSTRACT

Background: This study aims to investigate the effectiveness of application of single-port thoracoscopy using the subxiphoid approach in anterior mediastinal tumor surgery.

Methods: Between June 2014 and June 2016, a total of 108 patients (59 males, 49 females; mean age: 64.9±7.5 years; range, 45 to 79 years) with anterior mediastinal tumors were randomized into subxiphoid (experimental) or right chest (control) groups. Single-port thoracoscopy using the subxiphoid approach was performed in the subxiphoid group, while thoracoscopy using the right thoracic approach was performed in the control group. Pre- and postoperative pain stress indicators, Visual Analog Scale scores, quality of life scores, postoperative tumor recurrence, and five-year survival rates were compared between the groups.

Results: There were no mortality or serious complications in either group. The mean Visual Analog Scale pain scores on postoperative Days 1 and 7 were 6.5±0.8 and 2.9±0.8 in the subxiphoid group, respectively, compared to 7.2±0.8 and 3.4±0.8 in the control group ($p<0.05$ for all), respectively. The mean quality of life scores on postoperative Days 1 and 7 were 81.5±5.0 and 79.3±7.7, respectively, in the subxiphoid group compared to 72.4±4.3 and 71.3±4.8 in the control group, respectively ($p<0.05$ for all). Postoperative pain and pain mediator indexes were lower in the subxiphoid group ($p<0.05$ for all). The postoperative tumor recurrence rates were 3.70% and 20.37% in the subxiphoid and control groups, respectively ($p=0.008$). The five-year survival rates were 85.2% and 63.0% in the subxiphoid and control groups, respectively ($p=0.008$).

Conclusion: Single-port thoracoscopy using the subxiphoid approach is technically feasible, safe, and effective in performing surgery for anterior mediastinal tumors with an intact capsule and a tumor diameter of ≤5 cm.

Keywords: Anterior mediastinal tumor, surgery, thoracoscopy, tumor, xiphoid.

ÖZ

Amaç: Bu çalışmada anterior mediastinal tümör cerrahisinde subksifoidal yaklaşım kullanılarak tek port torakoskopi uygulamasının etkinliği araştırıldı.

Çalışma planı: Haziran 2014-Haziran 2016 tarihleri arasında, anterior mediastinal tümörlü toplam 108 hasta (59 erkek, 49 kadın; ort. yaş: 64.9±7.5 yıl; dağılım, 45-79 yıl) subksifoid (deneysel) veya sağ göğüs (kontrol) grubuna randomize edildi. Subksifoid grubuna subksifoidal yaklaşım ile tek port torakoskopi uygulanırken, kontrol grubuna sağ torasik yaklaşım ile torakoskopi yapıldı. Ameliyat öncesi ve sonrası ağrı stres göstergeleri, Görsel Analog Ölçeği skorları, yaşam kalitesi skorları, ameliyat sonrası tümör rekürensi ve beş yıllık sağkalım oranları gruplar arasında karşılaştırıldı.

Bulgular: Grupların hiçbirinde mortalite veya ciddi komplikasyon izlenmedi. Ameliyat sonrası 1 ve 7. günlerde ortalama Görsel Analog Ölçeği ağrı skorları subksifoid grubunda sırasıyla 6.5±0.8 ve 2.9±0.8 iken, kontrol grubunda sırasıyla 7.2±0.8 ve 3.4±0.8 idi (tümü için $p<0.05$). Ameliyat sonrası 1 ve 7. günlerde ortalama yaşam kalitesi skorları subksifoid grubunda sırasıyla 81.5±5.0 ve 79.3±7.7 iken, kontrol grubunda sırasıyla 72.4±4.3 ve 71.3±4.8 idi (tümü için $p<0.05$). Ameliyat sonrası ağrı ve ağrı mediatörleri indeksleri subksifoid grubunda daha düşüktü (tümü için $p<0.05$). Ameliyat sonrası tümör rekürens oranları subksifoid ve kontrol grubunda sırasıyla %3.70 ve %20.37 idi ($p=0.008$). Beş yıllık sağkalım oranları subksifoid ve kontrol grubunda sırasıyla %85.2 ve %63.0 idi ($p=0.008$).

Sonuç: Subksifoid yaklaşım ile tek port torakoskopi, kapsülü sağlam olan ve çapı ≤5 cm olan anterior mediastinal tümör cerrahisinde teknik olarak uygulanabilir, güvenli ve etkilidir.

Anahtar sözcükler: Anterior mediastinal tümör, cerrahi, torakoskopi, tümör, subksifoid.

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Anterior mediastinal tumors are the most common mediastinal tumors, and surgery still remains the primary treatment.[1] In addition to radiotherapy and chemotherapy for malignant lymphoma, early surgical intervention is recommended for other types of mediastinal tumors.[2]

With advancements in thoracoscopic technology, thoracoscopic thoracic wall incision is being gradually promoted in clinical practice.[3] However, this exposure remains unsatisfactory, particularly for patients with thymomas, and surgical treatment that cannot completely remove the fat in the thymus area can affect the prognosis of such patients. Postoperative radiotherapy is required to reduce the risk of tumor recurrence.[4] However, the intercostal surgical approach is limited by exposure of the upper pole of the thymus, contralateral phrenic nerve, and full length of the left brachiocephalic vein and postoperative intercostal neuralgia.[5] Recently, anterior mediastinal tumor resection using the subxiphoid approach has gained prominence, although it is still not fully developed.[6]

In the present study, we aimed to examine the clinical effectiveness of single-port thoracoscopic resection of anterior mediastinal tumors using the subxiphoid approach.

PATIENTS AND METHODS

This single-center, prospective, randomized-controlled study was conducted at The Affiliated Hospital of Putian University, Department of Thoracic and Cardiovascular Surgery between June 2014 and June 2016. Patients with anterior mediastinal tumors admitted to our clinic were reviewed. Inclusion criteria were as follows: direct enhancement on chest computed tomography (CT) suggesting an anterior mediastinum mass with an intact capsule; a maximum tumor diameter of ≤5.0 cm; and complete postoperative follow-up data. Exclusion criteria were as follows: tumor invasion of adjacent organs; conversion to thoracotomy; a body mass index (BMI) of ≥28 kg/m^2; and recurrence of mediastinal tumors after surgery. Using the New Drug Data Statistical Processing Software random table, a total of 108 patients (59 males, 49 females; mean age: 64.9±7.5 years; range, 45 to 79 years) were included. The patients were randomly assigned to the subxiphoid group (experimental) and the right chest group (control group) including 54 patients in each group.

Surgical method

Subxiphoid group

Directly enhanced chest CT images of the mediastinal masses are shown in Figure 1. In the subxiphoid group, each patient underwent general anesthesia with intravenous agents and single-lumen tracheal intubation. The patient was placed in the supine position with legs spread; the main knife was placed between the patient's legs, and the laparoscopic assistant was at the patient's right side. An anterior median incision (~5.0 cm in length) was created and extended beneath and close to the lower edge of the xiphoid process (Figure 1). We used an electric knife to cut the skin, subcutaneous tissue, and abdominal white line, and to bluntly free the extraperitoneal fat tissue and the posterior sheath of the abdominal wall. The index finger and oval forceps bluntly freed the space between the posterior sternum and the pericardium, a single-hole protective sleeve (trocar puncture needle, Kangji Medical Instrument Co., Ltd., China) was inserted, and the medical Solos Insufflator (Olympus 45L Medical Pneumoperitoneum, Jiangsu Anmao Medical Technology Co., Ltd., Jiangsu, China) was connected. Carbon dioxide (CO_2) was continuously insufflated, with the pressure adjusted to 10.0 mmHg.[7] The single-hole protective sleeve has four lumens into which we inserted a 10.0-mm 30° thoracoscope, a gas input pipe for intraperitoneal insufflation, an ultrasound knife, and gastric forceps for thoracoscopy. An exhaust pipe was connected to the chest bottle to reduce blurred vision during surgery.

First, the left mediastinal pleura was incised to open the left thoracic cavity and free the thymus and fat tissues within and along the left phrenic nerve until the left internal thoracic vein, left brachiocephalic vein, and left subclavian artery were exposed. The right mediastinal pleura was incised to open the right thoracic cavity and free the thymus and fat tissues inside and along the right phrenic nerve until the right internal thoracic vein, right innominate vein, superior vena cava, and the left brachiocephalic vein were exposed (Figure 1). The lower pole of the thymus tissue was freed along the anterior pericardium to the lower edge of the left innominate vein. Finally, the upper pole of the thymus tissue was removed, until the innominate artery, trachea, and inferior thyroid artery were exposed (Figure 1). Thymic veins with small diameters were cut using an ultrasonic knife. For thymic veins with large diameters (Figure 1), we used titanium clamps or Hem-o-Lok® (Teleflex, American) clips. Finally, the specimen bag was placed, and the anterior mediastinal mass and surrounding fat tissue were removed through the incision under the xiphoid process (Figure 2). Two 16-gauge negative pressure drainage tubes were indwelled postoperatively and drained through the incision under the xiphoid process (Figure 2).

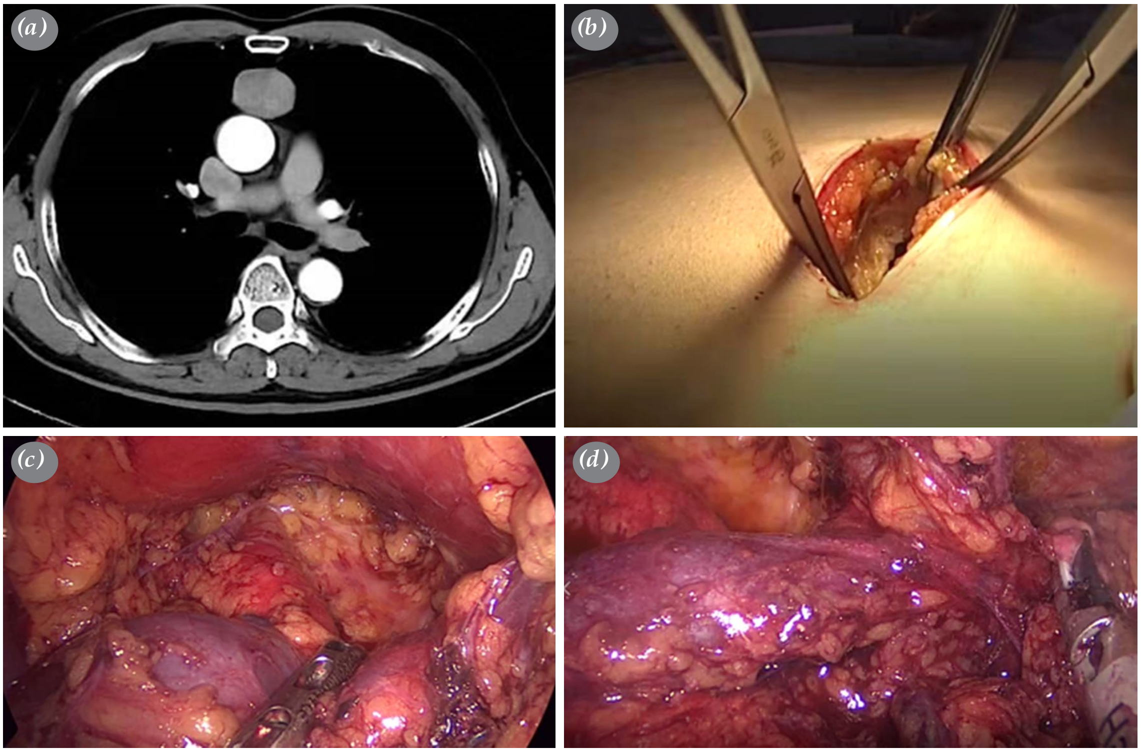

Figure 1. (a) Chest computed tomography images of the mediastinal masse. **(b)** An anterior median excision (~5.0 cm in length) is created and extended beneath the xiphoid process. **(c)** The innominate artery, trachea, and inferior thyroid artery are exposed. **(d)** The thymic veins are exposed.

Control group

Enhanced chest CT images of mediastinal masses are shown in Figure 3. In this group, the patient was in the supine position, with the right shoulder elevated at 45°. The mirror, main operating, and assistant operative holes were created in the fifth intercostal space on the right anterior axillary line and in the fifth and fourth intercostal spaces on the right

Figure 2. (a) The anterior mediastinal mass and surrounding fat tissue are removed through the incision beneath the xiphoid process. **(b)** Two 16-gauge negative pressure drainage tubes are indwelled postoperatively, both of which are drained through the incision under the xiphoid process.

midaxillary line, respectively, and were approximately 1.2, 1.0, and 0.5 cm in length, respectively (Figure 3). The anterior mediastinal mass and surrounding fat tissue were removed using an ultrasonic knife and electrocoagulation hook (Figure 3). Two closed chest drainage tubes were inserted postoperatively, followed by chest closure.

Outcome measures

Postoperative infections

Postoperative infections of the two groups were recorded, including lung, incision site, and urinary tract infections.

Pain mediator indexes and stress response indicators

Pre- and postoperative pain and stress were assessed before the operation and on postoperative Days 1 and 7. Fasting peripheral venous blood (3 mL) was collected in the morning. The serum from the samples was assayed by enzyme-linked immunoassay (ELISA) to detect pain mediators, including 5-hydroxytryptamine (5-HT), neuropeptide Y (NPY), adrenocorticotropic hormone (ACTH), and prostaglandin E2 (PGE2). Stress response indicators including interleukin (IL)-1β, IL-6, tissue necrosis factor-alpha (TNF-α), and C-reactive protein (CRP), were measured using an ELISA kit (Shanghai Runyu Biotechnology Co., Ltd, Shanghai, China).

Visual Analog Scale (VAS) scores

The VAS scores before surgery and on postoperative Days 1 and 7 were assessed.[8]

Karnofsky Performance Status (KPS) scores

Quality of life scores before surgery and on postoperative Days 1 and 7 using the KPS scale were assessed.

Follow-up

All patients were evaluated as follows: postoperative first year, once every three months; postoperative first and second years, once every six months; and postoperative third and fifth years, once yearly. A detailed medical history was taken, and patients underwent physical and direct enhancement examinations, including CT. The follow-up endpoint was mortality.

Statistical analysis

Statistical analysis was performed using the IBM SPSS for Windows version 25.0 software (IBM Corp., Armonk, NY, USA). Descriptive data were expressed in mean ± standard deviation (SD),

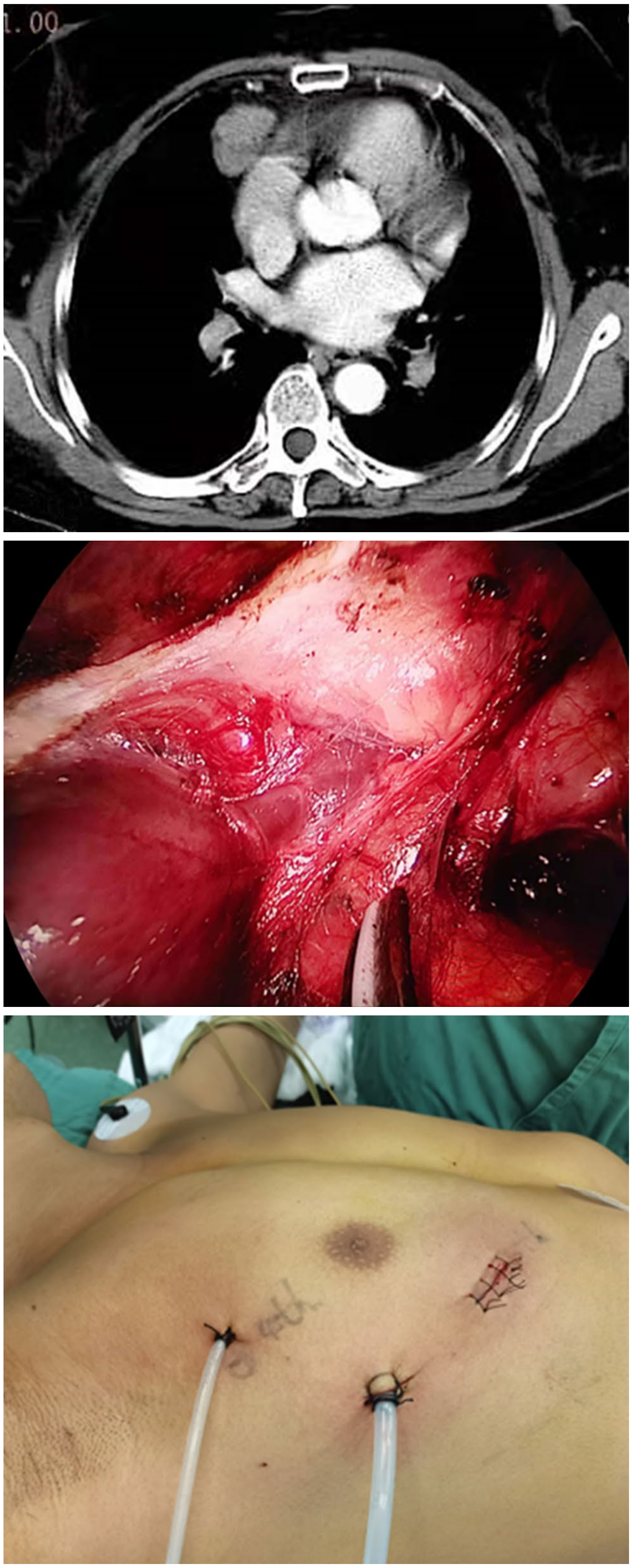

Figure 3. (a) Chest CT images of the mediastinal masses. **(b)** Thymic veins were exposed. **(c)** The mirror, main operating, and assistant operation holes were made at the fifth intercostal space on the right anterior axillary line, and at the fifth and forth intercostal space on the right midaxillary line.

median (min-max) or number and frequency, where applicable. Inter-group comparisons were performed using the independent samples t-test. Categorical data were compared using the chi-square test. The Kaplan-Meier curves were used for survival analysis. A p value of <0.05 was considered statistically significant.

RESULTS

Patients' characteristics and preoperative status

The characteristics of the patients in the subxiphoid and control groups are presented in Table 1. There were no significant differences in age, sex, tumor size, BMI, preoperative concurrent diseases, clinical symptoms, Masaoka staging of thymoma, or the pathological classification of thymoma between the two groups ($p>0.05$ for all).

Operative outcomes

All patients successfully underwent surgery and did not require conversion to thoracotomy. Postoperative pathology included thymic cyst (n=5), thymic hyperplasia (n=7), teratoma (n=6), hamartoma (n=9), thymic cancer (n=11), and types A (n=18), AB (n=12), B1 (n=14), B2 (n=13) and B3 (n=13) thymomas.

Intraoperative and postoperative conditions

The subxiphoid group had less postoperative pleural fluid drainage, shorter duration of catheter drainage, shorter postoperative hospital stays, lower incidence of postoperative complications, and lower postoperative tumor recurrence rates than the control group ($p<0.05$ for all) (Table 2).

Table 1. Clinical parameters of patients in the experimental and control groups

	Subxiphoid group (n=54)		Control group (n=54)			
Variables	n	Mean±SD	n	Mean±SD	χ^2/t-value	p
Age (year)		64.9±7.5		64.3±7.2	0.471	0.639
Sex					0.037	0.847
Male	30		29			
Female	24		25			
Tumor size (cm)		3.5±0.8		3.6±0.9	-0.657	0.512
BMI (kg/m^2)					0.339	0.560
<25	29		32			
25-28	25		22			
Preoperative concurrent diseases					0.389	0.823
Hypertension	23		20			
Diabetes	12		14			
Clinical symptoms					0.350	0.950
Ptosis	11		12			
Cough	11		10			
Chest pain	12		14			
No clinical symptoms	20		18			
Masaoka staging of thymoma					0.472	0.790
Stage I	24		23			
Stage II	18		16			
Pathological classification of thymoma					9.153	0.165
Type A	7		11			
Type AB	5		7			
Type B1	5		9			
Type B2	7		6			
Type B3	9		4			
Type C	9		2			

SD: Standard deviation; BMI: Body mass index.

Table 2. Intra- and postoperative characteristics of the experimental and control groups

	Subxiphoid group (n=54)			Control group (n=54)				
Variables	n	%	Mean±SD	n	%	Mean±SD	χ^2/t-value	*p*
Operation time (min)			134.5±60.1			129.4±46.3	0.502	0.617
Intraoperative blood loss (mL)			71.6±26.2			67.5±32.0	0.725	0.470
Postoperative pleural fluid drainage (mL)			384.4±118.4			443.9±110.9	-2.697	0.008
Duration of catheter drainage (h)			44.4±16.8			79.5±9.8	-13.400	<0.001
Postoperative hospital stay (day)			4.7±1.4			5.9±2.4	-3.073	0.003
Incidence of postoperative complications								
Pulmonary infection	4	7.41		12	22.22			
Incisional wound	2	3.70		6	11.11			
Urinary infection	1	1.85		4	7.41			
Postoperative tumor recurrence rate	2			11			7.083	0.008

SD: Standard deviation.

Comparison of pre- and postoperative pain mediator indexes

On postoperative Day 1, the levels of 5-HT, PGE2, ACTH, and NPY in the two groups were higher than the preoperative levels; however, the levels in the subxiphoid group were significantly lower than those in the control group ($p<0.05$). On postoperative Day 7, the 5-HT, PGE2, ACTH, and NPY levels of both groups of patients decreased in comparison with those from postoperative Day 1, but those of the

Table 3. Comparison of pre- and postoperative pain mediator indexes

	Subxiphoid group (n=54)	Control group (n=54)		
Variables	Mean±SD	Mean±SD	t-value	*p*
5-HT (ng/L)				
D1 (preoperative)	157.5±96.3	147.0±96.4	0.566	0.573
D1	214.5±107.7	272.1±116.0	-2.678	0.009
D7	184.8±100.9	228.4±73.4	-2.565	0.012
PGE2 (pg/mL)				
D1 (preoperative)	144.5±42.4	126.9±57.2	1.818	0.072
D1	230.4±36.7	277.8±63.1	-4.763	<0.001
D7	176.4±39.0	200.8±51.4	-2.782	0.006
ACTH (ng/mL)				
D1 (preoperative)	27.3±19.0	30.9±20.2	-0.967	0.336
D1	48.7±24.0	61.4±23.0	-2.808	0.006
D7	36.0±19.2	52.2±25.9	-3.698	<0.001
NPY (µg/L)				
D1 (preoperative)	127.0±33.0	116.9±33.4	1.578	0.118
D1	171.6±31.0	187.6±47.5	-2.075	0.040
D7	129.0±36.3	153.9±45.0	-3.163	0.002

SD: Standard deviation; 5-HT: 5-hydroxytryptamine; D1: First day after surgery; D7: Seventh day after surgery; PGE2: Prostaglandin E2; ACTH: Adrenocorticotropic hormone; NPY: Neuropeptide Y.

Table 4. Comparison of pre- and postoperative stress response indicators

	Subxiphoid group (n=54)	Control group (n=54)		
Variables	Mean±SD	Mean±SD	t-value	*p*
IL-1β (pg/mL)				
D1 (preoperative)	2.8±3.3	3.8±1.7	-1.924	0.057
D1	16.7±7.1	19.6±8.2	-1.995	0.049
D7	10.7±1.6	14.2±3.1	-7.385	<0.001
IL-6 (pg/mL)				
D1 (preoperative)	2.6±1.8	3.0±1.356	-1.231	0.221
D1	28.0±14.1	40.0±16.5	-4.076	<0.001
D7	8.6±4.5	12.6±3.9	-4.888	<0.001
TNF-α (ng/mL)				
D1 (preoperative)	8.2±5.6	7.3±3.3	1.045	0.298
D1	25.3±8.1	33.6±13.4	-3.888	<0.001
D7	10.2±4.7	18.2±9.0	-5.831	<0.001
C-reactive protein (mg/L)				
D1 (preoperative)	6.2±2.9	5.1±3.1	1.863	0.065
D1	44.7±44.2	69.6±75.6	-2.091	0.039
D7	11.4±10.0	27.1±18.6	-5.480	<0.001

SD: Standard deviation; IL-1β: Interleukin 1β; D1: First day after surgery; D7: Seventh day after surgery; IL-6: Interleukin 6; TNF-α: Tumor necrosis factor-alpha.

subxiphoid group remained significantly lower than those of the control group ($p<0.05$) (Table 3).

Comparison of pre- and postoperative stress response indicators

On postoperative Day 1, the levels of IL-1β, IL-6, TNF-α, and CRP in both groups were all higher compared to baseline, and the levels in the subxiphoid group were significantly lower than those in the control group ($p<0.05$ for all). On postoperative Day 7, the levels of IL-1β, IL-6, TNF-α, and CRP in the two groups decreased compared to those on postoperative Day 1; however, the levels in the subxiphoid group remained significantly lower than those of the control group ($p<0.05$) (Table 4).

Table 5. Comparison of VAS and KPS scores before and after surgery

	Subxiphoid group (n=54)	Control group (n=54)		
Variables	Mean±SD	Mean±SD	t-value	*p*
VAS scores				
D1 (preoperative)	0.5±0.5	0.5±0.5	-0.573	0.568
D1	6.5±0.8	7.2±0.8	-4.297	<0.001
D7	2.9±0.8	3.4±0.8	-3.222	0.002
KPS scores				
D1 (preoperative)	88.3±8.2	88.7±7.5	-0.245	0.807
D1	81.5±5.0	72.4±4.3	10.192	<0.001
D7	79.3±7.7	71.3±4.8	6.437	<0.001

SD: Standard deviation; VAS: Visual Analog Scale; KPS: Karnofsky Performance Status; D1: First day after surgery; D7: Seventh day after surgery.

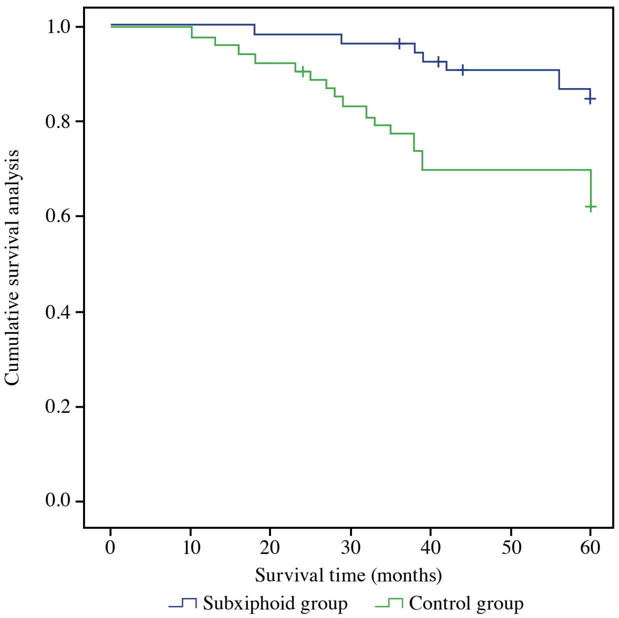

Figure 4. The five-year survival rate in the subxiphoid group (85.2%) is higher than in the control group (63.0%).

Comparison of VAS and KPS scores before and after surgery

On postoperative Day 1, the VAS scores of the two groups were higher than those before operation; the VAS scores of the subxiphoid group were significantly lower than those of the control group. The KPS scores were significantly lower in the subxiphoid group than the control group. On postoperative Day 7, the VAS scores were significantly lower, and the KPS scores were significantly higher in the subxiphoid group than the control group (Table 5).

Comparison of the five-year survival rate between the two groups of patients

A total of 108 patients in the two groups were followed; however, 11 patients were lost to follow-up (subxiphoid group, n=6; control group, n=5; rate, 10.2%). Recurrence of thymoma occurred in 13 patients. The five-year survival rate in the subxiphoid group was 85.2%, which was significantly higher than that the control group (63.0%) (χ^2=6.943, p=0.008) (Figure 4).

DISCUSSION

Mediastinal tumors have become an indication for thoracoscopic surgery.[9,10] This method enhances the postoperative quality of life of the patients. The transcostal approach for thoracoscopy has the advantages of reduced trauma, clear surgical vision, and quick postoperative recovery.[11] Regardless of if the three-hole or single-hole method is used, it is easy to compress the intercostal nerves and destroy the intercostal blood vessels, causing complications, such as postoperative incision pain and chest hemorrhage. Therefore, a simpler and more effective surgical method is required.

In 2012, Suda et al.[12] first reported a surgical method for thoracoscopic anterior mediastinal mass resection using the xiphoid process. The midline connection is the operating hole, which is then connected to the pneumoperitoneum to create an artificial pneumothorax for surgery. However, postoperative incision pain can restrict respiratory movement and increase norepinephrine secretion, aggravating the perioperative airway and inflammatory responses.[13]

After operation and five-year follow-up, our results confirm that single-port thoracoscopic surgery using the subxiphoid approach is safe and feasible. Pain scores on postoperative Days 1 and 7 were low, and none of the patients experienced intractable incision site pain.

We conducted this analysis to evaluate the single-port subxiphoid approach for surgery, as it has several major potential advantages: *(i)* single-port thoracoscopic approach decreases levels of pain mediators and pain-related stress response indicators compared to conventional three-port thoracoscopic surgery;[14] *(ii)* drainage tubes are placed in the posterior sternal space, avoiding compression of the intercostal nerves; and *(iii)* it avoids instrument friction against the costal arch tissue and compression of the nerve tissue at the lower edge of the costal arch.

Additionally, this approach *(i)* provides a clear surgical field of vision for thymomas, enabling more thorough removal of the adipose tissue; *(ii)* enhances procedural safety if the left innominate vein is damaged during surgery causing massive bleeding, as the thorax can be transferred immediately through the median sternum incision without the need to change positions; and *(iii)* is more conducive to removing the specimen, thereby avoiding the intercostal space, as there is no corresponding bony structure in the subxiphoid anatomy.

However, this surgical method also has the following shortcomings: *(i)* One small incision is made under the xiphoid process, and the entire operating space is narrow. Therefore, to perform this surgical technique, minimum 30 cases are required to overcome the learning curve to ensure the safety and effectiveness of the operation.[15] *(ii)* Surgical instruments must pass through the anterior pericardial space, which can easily stimulate and compress the heart, causing damage.[16] *(iii)* When the anterior mediastinal mass is large in diameter (>7 cm), the position is deep, or the substernal angle is less than 90°, intraoperative exposure is difficult and the surgical risk is greater. *(iv)* Once there is vascular injury, it is almost impossible to stop the bleeding using sutures, and the bleeding can only be stopped by local compression.

Single-port thoracoscopy under the xiphoid process has the following indications: *(i)* early thymoma; *(ii)* other benign anterior mediastinal tumors; *(iii)* anterior mediastinal tumor that does not significantly invade the surrounding organs or large blood vessels;[17] and *(iv)* anterior mediastinal mass with diameter <5.0 cm. Contraindications include *(i)* poor cardiopulmonary function and inability to tolerate surgery; *(ii)* malignant tumor of the anterior mediastinum with obvious invasion of surrounding organs or large blood vessels; and *(iii)* previous history of surgery for an anterior mediastinal tumor.

The main limitations to this study are its single-center design and relatively small sample size. Therefore, further large-scale, randomized studies are needed to confirm these findings.

In conclusion, the single-port thoracoscopic surgery technique under the xiphoid process is safe, effective, and feasible for the management of anterior mediastinal tumors. It can significantly reduce postoperative incision pain and has potential clinical applications.

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Ethics Committee Approval: The study protocol was approved by the The Affiliated Hospital of Putian University Ethics Committee (date: 18.06.2020, no: 2020028). The study was conducted in accordance with the principles of the Declaration of Helsinki.

Patient Consent for Publication: A written informed consent was obtained from each patient.

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

Author Contributions: Study conception and design, manuscript preparation: H.C., B.X.; Data collection: H.C., B.X., Q.Z., P.C.; Analysis and interpretation of results: Q.Z., M.C., J.Y.; All authors reviewed the results and approved the final version of the manuscript.

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