**Supplement 1**

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**This supplement contains the following items:**

1. Original protocol, final protocol, summary of revision.

2. Original statistical analysis plan, final statistical analysis plan, summary of revision.

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# Original study protocol

**Impact of epidural anesthesia-analgesia on recurrence-free survival in patients after surgery for lung cancer: Study protocol for a randomized controlled trial**

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## 1. Introduction

With increasing incidence and mortality, lung cancer has become the most common incident cancer and the leading cause of cancer death.1 At present, timely surgical resection remains the front-line therapy for non-small cell lung cancer (NSCLC).2 However, even after surgical resection, cancer recurrence or metastasis occurs in a significant proportion of patients. For example, in an 11-year follow-up study after surgery for early stage lung cancer (T1 to T2, N0 to N1 NSCLC), Kelsey et al.3 reported a 5-year rate of local and/or distant recurrence of 36%. Cancer recurrence/metastasis accounts for the majority of treatment failure and cancer death.4 In an early study of patients after surgery for N1 stage NSCLC, Sawyer et al.5 reported a 5-year mortality of 68%.

The development of cancer recurrence/metastasis after surgery depends largely on the balance between the ability of the residual cancer cells to implant, proliferate and attract new blood vessels and the anti-metastatic immune activity of the body.6,7 The balance may be shifted towards cancer progression by several perioperative factors. Firstly, surgery may promote the release of cancer cells into blood circulation.8,9 Secondly, cancer resection may reduce anti-angiogenic factors produced by the primary cancer10 but increase pro-angiogenic factors;11 these may promote cancer growth both locally and at distant sites. Thirdly, surgery-related stress response may inhibit cell-mediated immunity and promote cancer growth. Several studies demonstrated that perioperative stress may inhibit innate immunity, especially natural killer (NK) cells, which are known to play a major role in elimination of circulating cancer cells.12,13

It is increasingly recognized that anesthetic management may affect long-term outcome after cancer surgery.14,15 For example, experimental studies showed that inhalational anesthetics inhibit the toxicity of interferon-enhanced NK cells and the cytokine release from NK cells and NK cell-related cells;16,17 they can upregulate the expression of hypoxia inducible factors (HIFs),18-25 and high level HIFs are associated with poor cancer outcomes.26 Opioids are also found to inhibit cellular and humoral immune function. In animal studies, clinically relevant dose opioids can inhibit NK cell activity, and promote tumor growth and angiogenesis;27,28 although conflicting evidence also exists.29 On the other hand, local anesthetic drugs have effect of membrane stabilization and can inhibit invasion and proliferation of cancer cells in experimental studies.30,31 Furthermore, neuraxial anesthesia (such as epidural block) can effectively block the transmission of noxious stimuli into central neural system; it can, thus, decrease the consumption of general anesthetics and opioids, and avoid the over-activation of the sympathetic system and weaken the neural-endocrine stress response.32,33 Studies confirmed that use of thoracic epidural anesthesia is associated with lower concentrations of stress hormones, less release of endogenous opioids, and milder immunosuppression after surgery.33,34 Theoretically, use of neuraxial anesthesia during lung cancer surgery may help to relieve postoperative immunosuppression and reduce cancer recurrence/metastasis.

Clinical evidences mainly came from retrospective studies. Biki et al.35 analyzed data of patients after radical prostatectomy and found that use of epidural anesthesia was associated with a reduced risk of recurrence by about 57%. Exadaktylos et al.36 reported that women who received paravertebral block during breast cancer surgery had lower recurrence and metastasis rate at 36 months. Whereas in patients undergoing colorectal cancer surgery, the advantages of epidural anesthesia appear only in some subgroups.37,38 Results from prospective studies are still limited. Four trials are secondary analyzes of previously conducted prospective randomized controlled trials.39-42 To be noted, none of the above studies was performed in patients undergoing lung cancer surgery.

We hypothesize that, for patients undergoing lung cancer surgery, combined use of epidural anesthesia-analgesia with general anesthesia may reduce recurrence/metastasis and improve long-term survival, possibly by preserving immune function following surgery.

## 2. Study objectives

The main purpose of this study was to investigate whether “combined epidural-general anesthesia plus postoperative epidural analgesia” compared with “general anesthesia plus postoperative intravenous analgesia” can reduce rate of recurrence/metastasis in patients after surgery for lung cancer.

## 3. Study design

***3.1 Type of the study***

This is a two-center, randomized controlled trial with two parallel arms. The flow chart of the study is shown in Figure 1.

***3.2 Sample size estimation***

In a cohort study of patients after complete resection for NSCLC, Taylor and colleagues reported a recurrence rate of 33% at a median follow-up of 2 years after surgery.43 In a pilot study of our own, the 1-year recurrence rate after lung cancer surgery was 48% lower in patients who were given combined epidural-general anesthesia compared with those given general anesthesia alone. We expect that patient recruitment and follow-up will be completed in about 24 months, respectively. With the 2-sided significance level set at 0.05 and power at 80%, 360 patients (180 per group) are required to detect the difference. Considering a dropout rate of about 8% and an epidural failure rate of 2%, we plan to enroll 400 patients.

***3.3 Trial setting***

3.3.1 This two-center trial is conducted in Peking University First Hospital and Peking University Cancer Hospital in Beijing, China.

3.3.2 The study is coordinated by the Department of Anesthesiology and Critical Care Medicine of Peking University First Hospital and the Department of Anesthesiology of Peking University Cancer Hospital. Departments of Thoracic Surgery, Medical Imagining, and Pathology of the two Hospitals participate in the study.

## 4. Participants

Potential participants are screened by the qualified investigators the day before surgery. For those who will undergo surgery on Monday, screening will be performed on the previous Friday.

***4.1 Inclusion criteria***

4.1.1 Age between 18 and 80 years.

4.1.2 Planning to undergo lung cancer surgery.

4.1.3 Agreed to receive patient-controlled analgesia after surgery.

***4.2 Exclusion criteria***

Patients are excluded if they meet any of the following criteria:

4.2.1 Distant metastasis, malignant tumor in other organs, or chemo-/radio- or other anti-cancer therapy before surgery.

4.2.2 Comorbid with autoimmune diseases, or glucocorticoid/immunosuppressant therapy within 1 year.

4.2.3 Previous history of schizophrenia, epilepsy or Parkinson disease, or unable to complete preoperative assessment due to severe dementia, language barrier, or end-stage disease, or other psychological disorders related to the development of cancer.

4.2.4 Severe hepatic disease (Child-Pugh classification C), renal failure (serum creatinine >442 μmol/L or receiving renal replacement therapy), or American Society of Anesthesiologist (ASA) physical status classification ≥IV.

4.2.5 History of anesthesia and/or surgery within 1 year.

4.2.6 Contradictions to epidural anesthesia, including spinal deformity, coagulation disorder, local infection, history of spinal trauma/surgery.

4.2.7 Allergic to any medication during the study.

***4.3 Criteria of drop-out***

4.3.1 Study intervention is not administered successfully (due to failed epidural puncture, failed epidural catheterization, epidural catheter obstruction, blood appear in epidural catheter, inadequate epidural analgesia, dislodged epidural catheter, etc.).

4.3.2 Intervention interrupted by the investigators/anesthesiologists (due to severe adverse events).

4.3.3 Critical situations requiring early termination (potential coagulation disorder induced by massive bleeding, severe nausea and vomiting, or others).

The causes of protocol deviation should be recorded and corrected when possible. The cases will be followed up according to the study protocol and included in the intention-to-treat analysis.

4.3.4 Withdraw consent after intervention started.

The situation should be recorded. The primary therapeutic effects recorded in the last time will be regarded as the final results. The cases will be included in the intention-to-treat analysis.

***4.4 Criteria of elimination***

4.4.1 Withdraw consents before intervention.

4.4.2 Surgery cancelled.

4.4.3 No research record.

The causes of elimination should be explained. The case will be excluded from the intention-to-treat analysis. The case report forms will be preserved for reference.

***4.5 Criteria of study interruption***

Study will be interrupted in the following situations:

4.5.1 Severe safety problem occurred during the study.

4.5.2 Serious mistake found in the protocol.

4.5.3 Fund or management problem of the investigators.

4.5.4 Study cancelled by the administrative authority.

Study interruption may be transient or permanent. All recorded case report forms will be preserved for reference in case of study interruption.

## 5. Randomization and masking

***5.1 Randomization***

5.1.1 Random numbers are generated with a block size of 4 in a 1:1 ratio using the SAS 9.2 software package by a biostatistician who does not participate in the statistical analysis. The generated random numbers are sealed in consecutively numbered opaque envelopes, and kept by a study coordinator. Allocation is concealed until shortly before induction of anesthesia.

5.1.2 For each recruited patient, the study coordinator will distribute the randomization result to the anesthesiologists according to the sequence of recruited patients, and to coordinate between investigators.

5.1.3 For each recruited patient, an anesthesiologist will be designated for anesthesia and postoperative pain management according to the result of randomization.

5.1.4 Study intervention (combined epidural-general anesthesia plus postoperative epidural analgesia or general anesthesia alone plus postoperative intravenous analgesia) will be provided according to the randomization results by anesthesiologists who do not participate in the outcome assessments.

5.1.5 The results of randomization will be concealed and stored by the study coordinator until the end of the study.

***5.2 Masking***

5.2.1 Epidural block will be performed before anesthesia induction. Participants, anesthesiologists and other health-care team members are aware of study group assignment.

5.2.2 Investigators who are responsible for patient recruitment do not participate in anesthesia and perioperative care and postoperative in-hospital or long-term follow-up.

5.2.3 Anesthesiologists who are responsible for anesthesia and perioperative care are not involved in postoperative follow-up and outcome assessments.

5.2.4 Investigators who are designated for postoperative in-hospital follow‐ups do not participate in in-hospital care; investigators who are designated for long-term follow-ups and outcome assessments are not involved in in-hospital follow-up and are blinded to group assignment and perioperative management.

5.2.5 Statistical analysis will be performed by statisticians from the Department of Biostatistics of Peking University First Hospital.

***5.3 Emergency unmasking***

Because of the non-blinded design, emergency unblinding will not be necessary.

## 6. Study intervention

***6.1 General management***

6.1.1 For each participant, an attending anesthesiologist and an assistant anesthesiologist (usually a resident) are designated for anesthesia and perioperative care.

6.1.2 Intraoperative monitoring includes electrocardiogram, non-invasive blood pressure, pulse oxygen saturation, end-tidal concentrations of inhalational anesthetics and carbon dioxide, nasopharyngeal temperature, urine output and mechanical ventilation parameters (volume tidal, respiratory rate, etc.). Invasive arterial pressure (IAP) and central venous pressure (CVP) are monitored when necessary.

6.1.3 For all enrolled patients, video assisted thoracoscopic surgery is performed by a team consisting of thoracic surgeons, anesthesiologists, radiologists, and specialized nurses. The type of surgery is decided by surgeons according to patients’ conditions.

***6.2 General anesthesia and postoperative intravenous analgesia (GA group)***

6.2.1 Induction of anesthesia

General anesthesia is induced with propofol, sufentanil and rocuronium. Midazolam is administered when considered necessary. For patients with expected difficult airway, endotracheal intubation can be facilitated by succinylcholine or awake intubation can be performed. A double-lumen endobronchial tube or a bronchial blocker is used to facilitate one-lung ventilation.

6.2.2 Maintenance of anesthesia

General anesthesia is maintained with intravenous infusion of propofol and/or inhalation of sevoflurane (with or without nitrous oxide). Opioids (remifentanil and/or sufentanil) and muscle relaxant (rocuronium and/or cisatracurium) are administrated to maintain analgesia and muscle relaxation. The target is to maintain BIS between 40 to 60. A mixture of oxygen and air/nitrous oxide is provided during two-lung ventilation and also during one-lung ventilation as long as the pulse oxygen saturation is higher than 93%.

6.2.3 Analgesia after surgery

Patient-controlled intravenous analgesia (PCIA) is provided for up to 3 days after surgery, which is established with 0.5 mg/ml morphine and programmed to deliver 2-ml boluses with a lock-out interval of 6-10 minutes and a background infusion rate at 1 ml/h. For patients with low body weight or poor general condition, doses can be decreased and upper dose limit can be set for the patient-controlled pump. For patients whose intravenous analgesia pump has to be decreased or stopped, the reasons, administered dose and subsequent management should be recorded.

***6.3 Epidural-general anesthesia and postoperative epidural analgesia (EGA group)***

6.3.1 Epidural block

Epidural catheterization is performed before anesthesia induction. The intervertebral space for epidural puncture is selected by the attending anesthesiologists at T5-T8 levels. An epidural catheter is inserted using a standard technique. After negative aspiration of blood and cerebrospinal fluid, a test dose of 3-4 mL of 2% lidocaine is injected through the catheter to confirm the position of the catheter and the effect of neuraxial block.

6.3.2 Induction and maintenance of anesthesia

General anesthesia is induced and maintained with same medications in the same way as in GA group (see 6.2.1 and 6.2.2). In addition, 0.375% ropivacaine is administered via the indwelling epidural catheter by intermittent bolus injection or continuous infusion until the end of surgery. The target is to maintain BIS between 40 to 60.

6.3.3 Analgesia after surgery

Patient-controlled epidural analgesia (PCEA) is provided for up to 3 days after surgery, which is established with a mixture of 0.12% ropivacaine and 0.5 μg·ml-1 sufentanil and programmed to deliver 2-ml boluses with a lock-out interval of 20 minutes and a background infusion rate at 4 ml/h. For patients with low body weight or poor general condition, doses can be decreased and upper dose limit can be set for the patient-controlled pump. For patients whose epidural analgesia pump has to be decreased or stopped, the reasons, administered dose and subsequent management should be recorded.

***6.4 Remedial measures***

6.4.1 Failure of epidural block

6.4.1.1 For patients in the EGA group, epidural failure includes the following conditions: (1) failed multiple attempts (usually more than 5 times) of epidural puncture by a senior anesthesiologist; (2) patients refuse further epidural puncture attempts, or attending anesthesiologists consider further attempts is not beneficial; (3) no dermatomes with sensory loss appear after testing dose of epidural lidocaine (usually 10-20 min are required), and judged as failed epidural block by the attending anesthesiologists.

6.4.1.2 For patients in the EGA group with failed epidural block, anesthesia and postoperative analgesia will be performed as patients in the GA group (clause 6.2.1 to 6.2.3).

6.4.2 Inadequate anesthesia

6.4.2.1 For patients in the GA group, inadequate anesthesia is managed by increasing intravenous and/or inhalational anesthetics.

6.4.2.2 For patients in the EGA group, inadequate anesthesia is managed with additional local anesthetics administered through the epidural catheter, and/or by increasing intravenous and/or inhalational anesthetics.

6.4.3 Unsatisfied postoperative analgesia

6.4.3.1 For patients in the GA group, the PCIA pump setting will be adjusted (increased) and/or supplemental analgesics (such as opioids, non-steroidal anti-inflammatory drugs, and others) will be administered. The above measures will be recorded.

6.4.3.2 For patients in the EGA group, the PCEA pump setting will be adjusted (increased), supplemental analgesics (such as opioids, non-steroidal anti-inflammatory drugs, and others) will be administered, or an intravenous analgesia pump will be provided. The above measures will be recorded.

***6.5 Allowed and prohibited medications***

6.5.1 For patients of both groups, no premedication (usually include anticholinergics and sedatives) is administered. Dexmedetomidine can be administered at the discretion of anesthesiologists. Anticholinergics are prohibited unless being used for the treatment of bradycardia, in which case atropine will be administered.

6.5.2 During anesthesia, vasopressors (such as ephedrine, phenylephrine, dopamine, adrenaline and norepinephrine), antihypertensives (such as urapidil and nicardipine), atropine and esmolol can be used to maintain hemodynamic stable. The target is to maintain the fluctuation of blood pressure and heart rate within 30% from baseline (average level in the ward before surgery). Low-dose glucocorticoids (usually 5-10 mg dexamethasone) and 5-hydroxytryptamine 3 (5-HT3) receptor antagonists can be used to prevent postoperative nausea and vomiting.

6.5.3 For patients admitted to the ICU with endotracheal intubation, propofol and/or midazolam can be administered for sedation; the target is to maintain the Richmond Agitation-Sedation Scale44,45 (RASS, score ranges from –5 [unarousable] to +4 [combative] and 0 indicates alert and calm) from -2 to +1. For patients admitted to the ICU without endotracheal intubation, sedatives should not be administered unless otherwise needed. Other sedatives are not allowed.

6.5.4 Other perioperative management are provided according to routine practice.

## 7. Data collection

***7.1 Baseline data***

7.1.1 Demographic data, including gender, date of birth, educational levels, and body weight index.

7.1.2 Primary diagnosis, i.e., diagnosis of lung cancer according to the International Classification of Disease-10th edition, including location and clinical tumor-node-metastasis (TNM) stage.

7.1.3 Medical comorbidity, including diagnosis, duration, severity, and medical therapy.

7.1.4 Personal history, including chronic smoking, smoking index, excessive alcohol consumption, food and drug allergy, previous anesthesia/surgery, and exposure to harmful substances (such as coal, dust, asbestos and other chemicals).

7.1.5 Cancer history in first-degree family members.

7.1.6 Results of main laboratory tests, including blood/urine/stool routine, blood biochemistry, coagulation function, and cancer biomarkers.

7.1.7 Results of imaging diagnostic examinations, including chest X-ray, chest computed tomography (CT) scan/enhanced CT scan, cranial magnetic resonance imaging (MRI)/CT scan/enhancement CT scan, positron emission tomography (PET)-CT scan, abdominal ultrasound examination/CT scan, skeleton emission computed tomography (ECT) scan, and others if available.

7.1.8 Pathological diagnosis of biopsy specimen if performed.

7.1.9 Results of instrumental examinations, including electrocardiogram, echocardiogram, and lung function examination.

7.1.10 American Society of Anesthesiologists classification and New York Heart Association classification.

7.1.11 Serum concentrations of cortisol, interleukin-6 (IL-6) and IL-8. Blood samples are obtained in the morning (7:00-8:00 am) of the day before surgery.

***7.2 Intraoperative data***

For each participant, intraoperative data will be recorded by the anesthesiologists in charge of anesthesia.

7.2.1 Anesthesia method, types and doses of anesthetics and other medications used during anesthesia, duration of anesthesia.

7.2.2 Fluid balance (including fluid infusion, estimated blood loss, and urine output) and transfusion of blood products.

7.2.3 Surgical information, including location, type and duration of surgery, and results of frozen pathological examination if available.

7.2.4 Data of vital signs and arterial blood gas results.

7.2.5 Occurrence of adverse events and/or severe adverse event.

***7.3 In-hospital follow-ups after surgery***

7.3.1 Postoperative follow-ups will be performed by investigators who do not participate the intraoperative management. Postoperative follow-ups are performed twice daily (8:00-10:00 am, 18:00-20:00 pm) during the first 7 postoperative days and at hospital discharge (up to 30 days after surgery). Data in the Anesthesia Information Medical System and the Electronic Medical Record System will be achieved.

7.3.2 For patients who are admitted to ICU after surgery, the worst Acute Physiology and Chronic Health Evaluation II score (APACHE II; score ranges from 0 to 71, with higher score indicating more severe disease)46 during the first 24 hours after surgery, the percentage with endotracheal intubation, and the length of ICU stay are recorded.

7.3.3 The intensity of pain both at rest and with coughing are assessed twice daily (8:00-10:00 am, 18:00-20:00 pm) during the first 3 postoperative days with numeric rating scale (NRS; an 11-point scale where 0=no pain and 10=the worst pain). The final status of PCIA/PCEA pump use (completed use, dose reduction, early termination, change to other analgesic method) is recorded. The use of supplemental analgesics and other medications (including antiemetics) are recorded.

7.3.4 Serum concentrations of cortisol, interleukin-6 (IL-6) and IL-8. Blood samples are obtained in the morning (7:00-8:00 am) of postoperative days 1 and 3.

7.3.5 Delirium is assessed twice daily (8:00-10:00 am, 18:00-20:00 pm) during the first 7 postoperative days with the Confusion Assessment Method for the Intensive Care Unit (CAM-ICU).47,48 Immediately before assessing delirium, sedation or agitation will be assessed using the Richmond Agitation-Sedation Scale (RASS).44,45 For deeply sedated or unarousable patients (RASS –4 or –5), delirium is not assessed and the patient is recorded as comatose. For patients with a RASS from -3 to +4, delirium is assessed with the CAM-ICU. Investigators for postoperative delirium assessment will be trained by psychiatrists to use the CAM-ICU before the trial is commenced.

7.3.6 The occurrence of postoperative complications during hospital stay (up to 30 days) are recorded. Postoperative complications are generally defined as newly occurred medical conditions that are harmful for patients’ recovery and required therapeutic intervention, i.e., grade II or higher on the Clavien-Dindo classification.49

7.3.7 Duration of chest-tube drainage and length of stay in hospital after surgery.

7.3.8 All-cause in-hospital mortality after surgery (up to 30 days).

7.3.9 Results postoperative pathological examination are collected and included pathological type, maximal diameter, lymph node invasion, metastasis, and results of immune-histochemical analysis (if available). The TNM stage is classified according to the 7th edition International Association for the Study of Lung Cancer (IASLC) and the American Joint Committee on Cancer (AJCC) TNM classification.50

***7.4 Long-term follow-ups after surgery***

7.4.1 Long-term follow-ups will be performed by investigators who do not participate in in-hospital care and follow-ups, and are blinded to study group assignment.

7.4.2 Long-term follow-ups are performed every 3 months during the first year after surgery, every 6 months during the second year after surgery, and every year thereafter. The interval is shortened in cases with advanced-stage cancer. Patients are informed of the follow-up schedule before hospital discharge and are encouraged to cooperate with the investigators. Long-term follow-ups are performed via clinic visit, telephone interview and message contact until 2 years after surgery.

7.4.3 Data collection during long-term follow-ups includes the following:

7.4.3.1 Postoperative therapy for the primary surgical disease. For patients with confirmed lung cancer, post-surgical anti-cancer therapy may include chemotherapy, radiotherapy, targeted therapy, re-surgery, and others.

7.4.3.2 Results of postoperative re-examinations, which usually include chest radiography, computed tomography scan, positron emission tomography scan, sputum cytology, and measurement of tumor biomarkers.51-54 Cancer recurrence is defined as reappearance of the same cancer in the ipsilateral thorax, including lung and/or mediastinal/hilar lymph nodes; cancer metastasis is defined as reappearance of the same cancer in other part of the body, except the ipsilateral thorax. The development of cancer recurrence and/or metastasis will be diagnosed by surgeons (and/or radiologists when necessary) according to the results of re-examination. For patients with confirmed cancer recurrence and/or metastasis, the diagnostic evidence, date of earliest diagnoses and subsequent therapies are documented.

7.4.3.3 For patients who die during the follow-up period after surgery, the exact dates of death are recorded (consistent with death certificate). The causes of death are documented. Cancer-specific death is defined as death fully attributable to a specific cancer, i.e., the cancer for which the surgery is performed during the trial.55 Cancer-specific death usually occur after cancer recurrence and/or metastasis, with other causes including stroke, myocardial infarction or accident excluded.

## 8. Outcomes and assessment

***8.1 Primary outcome***

The primary outcome is rate of recurrence/metastasis after surgery.

***8.2 Secondary outcomes***

8.2.1 Pain intensity at postoperative days 1-3.

8.2.2 Serum concentrations of cortisol, IL-6 and IL-8 in the morning of postoperative days 1 and 3.

8.2.3 Incidence of postoperative delirium within the first 7 postoperative days.

8.2.4 Incidence of postoperative complications during hospital stay (up to 30 days after surgery).

8.2.5 Duration of chest-tube drainage.

8.2.6 Length of stay in hospital after surgery.

8.2.7 In-hospital all-cause mortality (up to 30 days after surgery).

8.2.8 Rate of all-cause mortality after surgery.

8.2.9 Rate of cancer-specific death after surgery.

## 9. Adverse events

***9.1 Definition***

An adverse event indicates any unpredictable, unfavorable medical event that is associated with any medical intervention and occurs during the study period. It can be related to the study intervention or otherwise. It can manifest as any uncomfortable signs (including abnormal laboratory findings), symptoms or transient morbidity.

***9.2 Predicted adverse events in this study***

9.2.1 Epidural puncture related adverse events

Adverse events related to epidural puncture/catheterization include accidental dural puncture, nerve injury, failed epidural puncture/catheterization, epidural catheter obstruction, blood appear in epidural catheter, epidural catheter dislodgement, local/epidural hematoma, local/epidural infection, etc.

9.2.2 Intraoperative adverse events related to epidural and/or general anesthesia may include the following:

|  |  |
| --- | --- |
| Local anesthetic intoxication | Caused by unexpected intravascular injection of large-dose local anesthetics, usually manifested as convulsion, twitch, loss of consciousness, arrhythmia, circulatory collapse, etc. |
| Total spinal anesthesia | Caused by unexpected intraspinal injection of large-dose local anesthetics, usually manifested as loss of sensation in all spinal innervated area, hypotension, loss of consciousness, apnea, etc. |
| New-onset arrythmia | Atrial/ventricular premature beat, supraventricular tachycardia, atrial fibrillation, etc. |
| Cardiac events | Acute coronary syndrome, ventricular fibrillation (require emergent cardiopulmonary resuscitation), etc. |
| High airway pressure | Peak airway pressure >30 cmH2O after readjusting endobronchial tube position by fiberscope re-examination, adequate muscle relaxation, and/or sputum suction. |
| CPAP to non-ventilated lung | Use of CPAP (continuous positive airway pressure) device to non-ventilated lung during one-lung ventilation to relieve desaturation (pulse oxygen saturation <88%). |
| Bradycardia | Heart rate <45 beats per minute. |
| Tachycardia | Heart rate >100 beats per minute. |
| Hypotension | Systolic blood pressure <90mmHg or a decrease of >30% from baseline value before surgery. |
| Hypertension | Systolic blood pressure >180 mmHg or an increase of >30% from baseline value before surgery. |
| Teeth injury | Caused by the procedure of laryngoscope exposure and endotracheal intubation. |
| Prophylaxis or prophylactic shock | Caused by medications during anesthesia, usually manifested as skin rash, bronchospasm, hypotension, shock, etc. |
| Others | --- |

9.2.2 Postoperative adverse events

9.2.2.1 Adverse events related to epidural analgesia include epidural catheter obstruction, inadequate epidural analgesia, epidural catheter dislodgement, epidural hematoma, epidural abscess, sequelae of nerve injury, post-dural puncture headache, pruritus, leg weakness (muscle strength of less than grade 5), etc.

9.2.2.2 Other adverse events include nausea, vomiting, postoperative hypotension (systolic blood pressure <90 mmHg or a decrease of >30% from baseline), postoperative hypertension (systolic blood pressure >180 mmHg or an increase of >30% from baseline), postoperative bradycardia (heart rate <45 bpm), postoperative tachycardia (heart rate >100 bpm), inadequate analgesia, etc.

9.2.3 In the present study, adverse events will be monitored from the beginning of anesthesia (epidural puncture or induction of general anesthesia) until 72 hours after surgery, i.e., end of patient-controlled analgesia.

***9.3 Management***

9.3.1 Therapies will be provided according to patients’ condition and routine practice. Generally, hypotension is managed with lighting anesthesia, intravenous fluid and vasopressors; hypertension is managed with deepening anesthesia and antihypertensive drugs; bradycardia is managed with lighting anesthesia and atropine; tachycardia is managed with deepening anesthesia, intravenous fluid and esmolol when necessary.

9.3.2 The study intervention (epidural anesthesia and/or postoperative epidural/intravenous analgesia) can be stopped temporarily or permanently if considered necessarily by the attending anesthesiologist or surgeons. The time and reasons of study intervention interruption will be recorded.

9.3.3 In case that the patient-controlled analgesia (for study intervention) is terminated early, other analgesics will be provided.

***9.4 Record***

9.4.1 Any adverse event should be documented, including occurrence, type/diagnosis, time of diagnosis, management, duration of persistence, and sequelae.

9.4.2 Any adverse event should be followed up until it is completely resolved or therapy terminated.

## 10. Severe adverse events

***10.1 Definition***

A severe adverse event indicates any unpredictable medical events that lead to death, threat of life, prolonged length of hospital stays, persistent disability or dysfunction, or other severe events.

***10.2 Monitoring severe adverse events***

In the present study, severe adverse events will be monitored from the beginning of anesthesia (epidural puncture or induction of general anesthesia) until 72 hours after surgery, i.e., end of patient-controlled analgesia.

***10.3 Management***

In case of any severe adverse events, the study intervention will be stopped and treatment will be initiated immediately according to routine practice.

***10.4 Record and report***

10.3.1 In case of any severe adverse event, apart from active treatment and record as above, the principal investigator and the Clinical Research Ethics Committee of Peking University First Hospital will be informed within 24 hours in report form.

10.3.2 In case of study intervention related death, immediately stop the clinical trial, report the event to the Ethics Committee as soon as possible, record in detail and carefully preserve the related documents.

10.3.3 Any severe adverse event must be followed up until it is completely resolved or when therapy is ended.

## 11. The rule of unmasking

11.1 After the follow-up of all cases have been completed, the data of case report forms have been checked as correct, and the data entry have been finished and checked without problems, the database will be locked.

11.2 After the database is locked, the first unmasking will be conducted. And the database will be sent to the statisticians for statistical analysis.

11.3 When the statistical analysis was completed, a second unmasking will be performed to determine which group is the intervention group or the control group.

## 12. Data management

12.1 All data collected during the study period will be recorded in the case report form (CRF) by designated investigators. The investigators are trained to obey the predefined data-collection rule and complete the CRF as soon as possible after the follow-up is performed.

12.2 To ensure the correctness of database, data entry will be double checked by two personnel. Any queries during data entry will be resolved by the investigators or the principal investigator.

12.3 After data entry is completed and database is checked without problems, the database will be locked and sent for statistical analysis. The database will be analyzed by the statistician personnel.

## 13. Statistical analysis

***13.1 General principles***

13.1.1 Baseline data and outcomes will be described according the data type and distribution. Continuous variables with normal or approximate normal distribution will be expressed as mean ± standard deviation (SD), while continuous variables with non-normal distribution will be expressed as median (minimum, maximum; or interquartile range, IQR). Categorical variables will be presented as number of cases (percentage). Time-to-events variables will be presented as mean/median time (95% CI).

13.1.2 For each hypothesis, two-tailed tests will be used in all statistical analysis, and p <0.05 will be considered statistically significant.

13.1.3 The primary and secondary outcomes will be analyzed in an intention-to-treat population, i.e., all patients are analyzed in the group to which they are randomized and received at least part of study intervention. If epidural anesthesia and/or analgesia fail, these patients will be converted to general anesthesia and intravenous analgesia but will be analyzed in the group to which they are randomized. Also, we will do per-protocol analysis for the primary endpoint, in this case, drop-out patients will be excluded.

***13.2 Patient recruitment and drop-out status***

The status of patient recruitment and drop-out will be summarized and listed. Comparison of the overall elimination/drop-out rate between the two groups will be performed with chi-square test.

***13.3 Demographics and baseline characteristics***

13.3.1 Demographics and baseline data will be presented.

13.3.2 For between-group differences, continuous variables will be analyzed using independent-samples t test or Mann-Whitney U test; categorical variables will be analyzed using the chi-square test, continuity correction chi-square test or Fisher exact test.

***13.4 Intra- and postoperative variables***

Numeric variables will be analyzed using the independent-samples t test or Mann-Whitney U test; categorical variables will be analyzed using the chi-square test, continuity correction chi-square test or Fisher exact test. Missing data will not be replaced.

***13.5 Outcome analysis***

13.5.1 Evaluation of primary endpoint

13.5.1.1 Cancer recurrence/metastasis after surgery will be analyzed using a Kaplan-Meier estimator with difference between groups assessed by log-rank test; a Cox proportional hazards model will be used to adjust for potential confounding factors. Effect size will be expressed as hazard ratio and its 95% CI. Patients who are lost to follow-up will be censored at the time of last contact.

13.5.2 Evaluation of secondary outcomes

13.5.2.1 Discrete variables (NRS pain scores) will be analyzed with the Mann-Whitney U test. Median difference and 95% CI will be calculated with the Hodges-Lehmann estimator. Missing data will not be replaced.

13.5.2.2 Numeric variables (serum concentrations of cortisol, IL-6 and IL-8) will be analyzed using the independent-samples t test or Mann-Whitney U test. Mean/median difference and 95% CI will be calculated. Median difference and 95% CI will be calculated with the Hodges-Lehmann estimator. Missing data will not be replaced.

13.5.2.3 Categorical variables (incidence of postoperative delirium, incidence of postoperative complication, in-hospital mortality) will be analyzed using the chi-square test, continuity correction chi-squared test or Fisher’s exact test. The estimated risks ratio and 95% CI will be provided. Missing data will not be replaced.

13.5.2.4 Time-to-event variables (duration of chest-tube drainage, length of stay in hospital) will be analyzed using a Kaplan-Meier estimator with difference between groups assessed by log-rank test. The estimated hazard ratio and 95% CI will be provided. Patients who are lost to follow-up will be censored at the time of last contact.

13.5.2.5 Long-term mortality (all-cause death after surgery, cancer-specific death after surgery) will be analyzed using a Kaplan-Meier estimator with difference between groups assessed by log-rank test; Cox proportional hazards models will be used to adjust for potential confounding factors. The estimated hazard ratio and 95% CI will be provided. For cancer-specific death, patients who die of a cause other than the specific cancer will be censored at the time of death. Patients who are lost to follow-up will be censored at the time of last contact.

***13.6 Safety analysis***

13.3.1 Describe the occurrence of adverse events in each group.

13.3.2 Describe the management of adverse events when appropriate.

13.3.3 Describe the occurrence of severe adverse events.

13.3.4 The rates of adverse events and/or managements between the two groups will be compared with chi-square test, continuity correction chi-square test or Fisher exact test. The estimated risks ratio (RR) and 95% CI will be provided.

13.3.5 Missing data will not be replaced.

## 14. Quality control and quality assurance

14.1.1 An investigator training program will be designed by the principle investigator. A study coordinator will be designated to organize and implement the training program, and to record and preserve the related documents.

14.1.2 The training program includes the following:

14.1.2.1 For anesthesiologists, they should be familiar with the protocol, the procedures of radical lung cancer surgery, the procedures of anesthesia and perioperative care, and the case report form.

14.1.2.2 For in-hospital follow-up investigators, they should be familiar with the potential adverse events or severe adverse event, postoperative complications, as well as the case report form.

14.1.2.3 For long-term outcome follow-up investigators, they are trained to communicate with participants and collect data regarding cancer progression and/or living status regularly (every 3 months during the first year, every 6 months during the second year, and every year thereafter); they are required to closely communicate with thoracic surgeons regarding the diagnosis of cancer outcomes. Furthermore, they do not participate in perioperative care and in-hospital follow-ups, have no knowledge of study group allocation, and are not permitted to communicate with health-care providers, participants and other investigators regarding study group allocation.

14.1.2.4 Other training contents that must be completed before trial initiation include the Good Clinical Practice principles, the standard operating procedures of the study, the working plan of the study, the data recording format, the outcome assessment, the instruction for the case report form, the skills of communicating with subjects, and other matters needing attention during the study (screening path, allowed and prohibited medications, etc.).

14.1.3 The instruments, equipment, assessment scales and definitions involved in the trial should be used according to executive standards. Investigators should stick to the preselected tools/definitions in order to achieve uniform evaluation and assessment.

14.1.4 Investigators who are responsible to record data must be authorized by the principle investigator in advance. Data sources include, but not limits to, electronic health records, anesthesia medical information system, medical equipment, as well as interviews before surgery and during postoperative (long-term) follow-ups.

14.1.5 The investigator should fill in the case report forms timely and completely. All observed and assessed results as well as abnormal findings during the study period should be verified and recorded to ensure the reliability of data.

14.1.6 Dataset management and statistical analysis are performed by professional biostatistics personnel.

14.1.7 All conclusions should be based on original data.

## 15. Ethical requirements and patient informed consent

***15.1 Ethics Committee***

The study protocol must be approved by the Clinical Research Ethics Committee of Peking University First Hospital before the study can be started. The investigators must strictly follow the Helsinki Declaration and China's relevant clinical trial management regulations. The principal investigator is responsible to report the status and the progress of the study to the Ethics Committee.

***15.2 Written informed consent***

For each potential participant, investigators are responsible to fully explain the purpose, procedures and possible risks of this study in a written form manner. The investigators must let every potential participant know that he/she has the right to withdraw consent from the study at any time. Every potential participant must be given a written informed consent. Every participant or the authorized surrogate of the participant must sign the consent before he/she can be enrolled in the study. The written informed consents will be kept as a part of the clinical trial documents.

## 16. Study termination

16.1 In case that intervention-related severe adverse events or serious quality problem occur during the study period, the study will be stopped. A report will be sent to the Ethics Committee. Restart of the study will need an approval from the Ethics Committee.

16.2 The study will be terminated after accomplishment of required patient recruitment and data collection. Decision will be made by the principal investigator.

## 17. Document preservation

Investigators and sponsors should properly keep clinical trial documents and data for 5 years in accordance with Good Clinic Practice requirements.

## 18. Conflict of interest

The investigators declare no conflicts of interest.

Patients planned for lung cancer surgery

Screening according to inclusion/exclusion criteria

Randomization

N = 400

General anesthesia and postoperative intravenous analgesia (GA group)

In-hospital follow-up

Combined epidural-general anesthesia and postoperative epidural analgesia (EGA group)

Estimated epidural failure rate of 2%

In-hospital follow-up

Informed consent

**Enrollment**

ITT analysis (N= )

PP analysis (N= )

ITT analysis (N= )

PP analysis (N= )

Long-term follow-up

Estimated dropout rate of 4%

Long-term follow-up

Estimated dropout rate of 4%

**Allocation**

**Follow-up**

**Analysis**

**Figure 1.** Planned flow chart. ITT=intention-to-treat; PP=per-protocol.

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# Final study protocol

**Impact of epidural anesthesia-analgesia on recurrence-free survival in patients after surgery for lung cancer: Study protocol for a randomized controlled trial**

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## 1. Introduction

With increasing incidence and mortality, lung cancer has become the most common incident cancer and the leading cause of cancer death.[1](#_ENREF_6_1) At present, timely surgical resection remains the front-line therapy for non-small cell lung cancer (NSCLC).[2](#_ENREF_6_2) However, even after surgical resection, cancer recurrence or metastasis occurs in a significant proportion of patients. For example, in an 11-year follow-up study after surgery for early stage lung cancer (T1 to T2, N0 to N1 NSCLC), Kelsey et al.[3](#_ENREF_6_3) reported a 5-year rate of local and/or distant recurrence of 36%. Cancer recurrence/metastasis accounts for the majority of treatment failure and cancer death.[4](#_ENREF_6_4) In an early study of patients after surgery for N1 stage NSCLC, Sawyer et al.[5](#_ENREF_6_5) reported a 5-year mortality of 68%.

The development of cancer recurrence/metastasis after surgery depends largely on the balance between the ability of the residual cancer cells to implant, proliferate and attract new blood vessels and the anti-metastatic immune activity of the body.[6](#_ENREF_6_6),[7](#_ENREF_6_7) The balance may be shifted towards cancer progression by several perioperative factors. Firstly, surgery may promote the release of cancer cells into blood circulation.[8](#_ENREF_6_8),[9](#_ENREF_6_9) Secondly, cancer resection may reduce anti-angiogenic factors produced by the primary cancer[10](#_ENREF_6_10) but increase pro-angiogenic factors;[11](#_ENREF_6_11) these may promote cancer growth both locally and at distant sites. Thirdly, surgery-related stress response may inhibit cell-mediated immunity and promote cancer growth. Several studies demonstrated that perioperative stress may inhibit innate immunity, especially natural killer (NK) cells, which are known to play a major role in elimination of circulating cancer cells.[12](#_ENREF_6_12),[13](#_ENREF_6_13)

It is increasingly recognized that anesthetic management may affect long-term outcome after cancer surgery.[14](#_ENREF_6_14),[15](#_ENREF_6_15) For example, experimental studies showed that inhalational anesthetics inhibit the toxicity of interferon-enhanced NK cells and the cytokine release from NK cells and NK cell-related cells;[16](#_ENREF_6_16),[17](#_ENREF_6_17) they can upregulate the expression of hypoxia inducible factors (HIFs),[18-25](#_ENREF_6_18) and high level HIFs are associated with poor cancer outcomes.[26](#_ENREF_6_26) Opioids are also found to inhibit cellular and humoral immune function. In animal studies, clinically relevant dose opioids can inhibit NK cell activity, and promote tumor growth and angiogenesis;[27](#_ENREF_6_27),[28](#_ENREF_6_28) although conflicting evidence also exists.[29](#_ENREF_6_29) On the other hand, local anesthetic drugs have effect of membrane stabilization and can inhibit invasion and proliferation of cancer cells in experimental studies.[30](#_ENREF_6_30),[31](#_ENREF_6_31) Furthermore, neuraxial anesthesia (such as epidural block) can effectively block the transmission of noxious stimuli into central neural system; it can, thus, decrease the consumption of general anesthetics and opioids, and avoid the over-activation of the sympathetic system and weaken the neural-endocrine stress response.[32](#_ENREF_6_32),[33](#_ENREF_6_33) Studies confirmed that use of thoracic epidural anesthesia is associated with lower concentrations of stress hormones, less release of endogenous opioids, and milder immunosuppression after surgery.[33](#_ENREF_6_33),[34](#_ENREF_6_34) Theoretically, use of neuraxial anesthesia during lung cancer surgery may help to relieve postoperative immunosuppression and reduce cancer recurrence/metastasis.

Clinical evidences mainly came from retrospective studies. Biki et al.[35](#_ENREF_6_35) analyzed data of patients after radical prostatectomy and found that use of epidural anesthesia was associated with a reduced risk of recurrence by about 57%. Exadaktylos et al.[36](#_ENREF_6_36) reported that women who received paravertebral block during breast cancer surgery had lower recurrence and metastasis rate at 36 months. Whereas in patients undergoing colorectal cancer surgery, the advantages of epidural anesthesia appear only in some subgroups.[37](#_ENREF_6_37),[38](#_ENREF_6_38) In a recent meta-analysis, Pei et al.[39](#_ENREF_6_39) concluded that epidural anesthesia combined with general anesthesia might be associated with improvement in prognosis of patients with operable prostate cancer; however, no obvious relationship between the improvement in prognosis of colorectal cancer and combined epidural-general anesthesia were detected.

Results from prospective studies are still limited. A meta-analysis included four post-hoc analyzes of randomized trials with a total of 746 participants.[40](#_ENREF_6_40) The authors concluded that evidence regarding the benefit of regional anesthesia on cancer recurrence is inadequate, and further prospective randomized controlled trials are needed. To be noted, none of the above studies was performed in patients undergoing lung cancer surgery.

We hypothesize that, for patients undergoing lung cancer surgery, combined use of epidural anesthesia-analgesia with general anesthesia may reduce recurrence/metastasis and improve long-term survival, possibly by preserving immune function following surgery.

## 2. Study objectives

The main purpose of this study was to investigate whether “combined epidural-general anesthesia plus postoperative epidural analgesia” compared with “general anesthesia plus postoperative intravenous analgesia” can improve recurrence-free survival in patients after surgery for lung cancer.

## 3. Study design

***3.1 Type of the study***

This is a single-center, randomized controlled trial with two parallel arms. The flow chart of the study is shown in Figure 1.

***3.2 Sample size estimation***

In a cohort study of patients after complete resection for NSCLC, Taylor and colleagues reported a recurrence rate of 33% at a median follow-up of 2 years after surgery.[41](#_ENREF_6_41) In a pilot study of our own, the 1-year recurrence rate after lung cancer surgery was 48% lower in patients who were given combined epidural-general anesthesia compared with those given general anesthesia alone. We expect that patient recruitment and follow-up will be completed in about 24 months, respectively. With the 2-sided significance level set at 0.05 and power at 80%, 360 patients (180 per group) are required to detect the difference. Considering a dropout rate of about 8% and an epidural failure rate of 2%, we plan to enroll 400 patients.

***3.3 Trial setting***

3.3.1 This single-center trial is conducted in Peking University First Hospital in Beijing, China.

3.3.2 The study is coordinated by the Department of Anesthesiology and Critical Care Medicine of Peking University First Hospital. Departments of Thoracic Surgery, Medical Imagining, and Pathology of Peking University First Hospital participate in the study.

## 4. Participants

Potential participants are screened by the qualified investigators the day before surgery. For those who will undergo surgery on Monday, screening will be performed on the previous Friday.

***4.1 Inclusion criteria***

4.1.1 Age between 18 and 80 years.

4.1.2 Planning to undergo lung cancer surgery.

4.1.3 Agreed to receive patient-controlled analgesia after surgery.

***4.2 Exclusion criteria***

Patients are excluded if they meet any of the following criteria:

4.2.1 Distant metastasis, malignant tumor in other organs, or chemo-/radio- or other anti-cancer therapy before surgery.

4.2.2 Comorbid with autoimmune diseases, or glucocorticoid/immunosuppressant therapy within 1 year.

4.2.3 Previous history of schizophrenia, epilepsy or Parkinson disease, or unable to complete preoperative assessment due to severe dementia, language barrier, or end-stage disease, or other psychological disorders related to the development of cancer.

4.2.4 Severe hepatic disease (Child-Pugh classification C), renal failure (serum creatinine >442 μmol·L-1 or receiving renal replacement therapy), or American Society of Anesthesiologist (ASA) physical status classification ≥IV.

4.2.5 History of anesthesia and/or surgery within 1 year.

4.2.6 Contradictions to epidural anesthesia, including spinal deformity, coagulation disorder, local infection, history of spinal trauma/surgery.

4.2.7 Allergic to any medication during the study.

***4.3 Criteria of drop-out***

4.3.1 Study intervention is not administered successfully (due to failed epidural puncture, failed epidural catheterization, epidural catheter obstruction, blood appear in epidural catheter, inadequate epidural analgesia, dislodged epidural catheter, etc.).

4.3.2 Intervention interrupted by the investigators/anesthesiologists (due to severe adverse events).

4.3.3 Critical situations requiring early termination (potential coagulation disorder induced by massive bleeding, severe nausea and vomiting, or others).

The causes of protocol deviation should be recorded and corrected when possible. The cases will be followed up according to the study protocol and included in the intention-to-treat analysis.

4.3.4 Withdraw consent after intervention started.

The situation should be recorded. The primary therapeutic effects recorded in the last time will be regarded as the final results. The cases will be included in the intention-to-treat analysis.

***4.4 Criteria of elimination***

4.4.1 Withdraw consents before intervention.

4.4.2 Surgery cancelled.

4.4.3 No research record.

The causes of elimination should be explained. The case will be excluded from the intention-to-treat analysis. The case report forms will be preserved for reference.

***4.5 Criteria of study interruption***

Study will be interrupted in the following situations:

4.5.1 Severe safety problem occurred during the study.

4.5.2 Serious mistake found in the protocol.

4.5.3 Fund or management problem of the investigators.

4.5.4 Study cancelled by the administrative authority.

Study interruption may be transient or permanent. All recorded case report forms will be preserved for reference in case of study interruption.

## 5. Randomization and masking

***5.1 Randomization***

5.1.1 Random numbers are generated with a block size of 4 in a 1:1 ratio using the SAS 9.2 software package by a biostatistician who does not participate in the statistical analysis. The generated random numbers are sealed in consecutively numbered opaque envelopes, and kept by a study coordinator. Allocation is concealed until shortly before induction of anesthesia.

5.1.2 For each recruited patient, the study coordinator will distribute the randomization result to the anesthesiologists according to the sequence of recruited patients, and to coordinate work between investigators.

5.1.3 For each recruited patient, an anesthesiologist will be designated for anesthesia and postoperative pain management according to the result of randomization.

5.1.4 Study intervention (combined epidural-general anesthesia plus postoperative epidural analgesia or general anesthesia alone plus postoperative intravenous analgesia) will be provided according to the randomization results by anesthesiologists who do not participate in the outcome assessments.

5.1.5 The results of randomization will be concealed and stored by the study coordinator until the end of the study.

***5.2 Masking***

5.2.1 Epidural block will be performed before anesthesia induction. Participants, anesthesiologists and other health-care team members are aware of study group assignment.

5.2.2 Investigators who are responsible for patient recruitment do not participate in anesthesia and perioperative care and postoperative in-hospital or long-term follow-up.

5.2.3 Anesthesiologists who are responsible for anesthesia and perioperative care are not involved in postoperative follow-up and outcome assessments.

5.2.4 Investigators who are designated for postoperative follow‐ups do not participate in in-hospital care; investigators who are designated for long-term follow-ups and outcome assessments are not involved in in-hospital follow-up and are blinded to group assignment and perioperative management.

5.2.5 Statistical analysis will be performed by statisticians from the Department of Biostatistics of Peking University First Hospital.

***5.3 Emergency unmasking***

Because of the non-blinded design, emergency unblinding will not be necessary.

## 6. Study intervention

***6.1 General management***

6.1.1 For each participant, an attending anesthesiologist and an assistant anesthesiologist (usually a resident) are designated for anesthesia and perioperative care.

6.1.2 Intraoperative monitoring includes electrocardiogram, non-invasive blood pressure, pulse oxygen saturation, end-tidal concentrations of inhalational anesthetics and carbon dioxide, nasopharyngeal temperature, urine output and mechanical ventilation parameters (volume tidal, respiratory rate, etc.). Invasive arterial pressure (IAP) and central venous pressure (CVP) are monitored when necessary.

6.1.3 For all enrolled patients, video assisted thoracoscopic surgery is performed by a team consisting of thoracic surgeons, anesthesiologists, radiologists, and specialized nurses. The type of surgery is decided by surgeons according to patients’ conditions.

***6.2 General anesthesia and postoperative intravenous analgesia (GA group)***

6.2.1 Induction of anesthesia

General anesthesia is induced with propofol, sufentanil and rocuronium. Midazolam is administered when considered necessary. For patients with expected difficult airway, endotracheal intubation can be facilitated by succinylcholine or awake intubation can be performed. A double-lumen endobronchial tube or a bronchial blocker is used to facilitate one-lung ventilation.

6.2.2 Maintenance of anesthesia

General anesthesia is maintained with intravenous infusion of propofol and/or inhalation of sevoflurane (with or without nitrous oxide). Opioids (remifentanil and/or sufentanil) and muscle relaxant (rocuronium and/or cisatracurium) are administrated to maintain analgesia and muscle relaxation. The target is to maintain BIS between 40 to 60. A mixture of oxygen and air/nitrous oxide is provided during two-lung ventilation and also during one-lung ventilation as long as the pulse oxygen saturation is higher than 93%.

6.2.3 Analgesia after surgery

Patient-controlled intravenous analgesia (PCIA) is provided for up to 3 days after surgery, which is established with 0.5 mg·ml-1 morphine and programmed to deliver 2-ml boluses with a lock-out interval of 6-10 minutes and a background infusion rate at 1 ml·h-1. For patients with low body weight or poor general condition, doses can be decreased and upper dose limit can be set for the patient-controlled pump. For patients whose intravenous analgesia pump has to be decreased or stopped, the reasons, administered dose and subsequent management should be recorded.

***6.3 Epidural-general anesthesia and postoperative epidural analgesia (EGA group)***

6.3.1 Epidural block

Epidural catheterization is performed before anesthesia induction. The intervertebral space for epidural puncture is selected by the attending anesthesiologists at T5-T8 levels. An epidural catheter is inserted using a standard technique. After negative aspiration of blood and cerebrospinal fluid, a test dose of 3-4 mL of 2% lidocaine is injected through the catheter to confirm the position of the catheter and the effect of neuraxial block.

6.3.2 Induction and maintenance of anesthesia

General anesthesia is induced and maintained with same medications in the same way as in GA group (see 6.2.1 and 6.2.2). In addition, 0.375% ropivacaine is administered via the indwelling epidural catheter by intermittent bolus injection or continuous infusion until the end of surgery. The target is to maintain BIS between 40 to 60.

6.3.3 Analgesia after surgery

Patient-controlled epidural analgesia (PCEA) is provided for up to 3 days after surgery, which is established with a mixture of 0.12% ropivacaine and 0.5 μg·ml-1 sufentanil and programmed to deliver 2-ml boluses with a lock-out interval of 20 minutes and a background infusion rate at 4 ml·h-1. For patients with low body weight or poor general condition, doses can be decreased and upper dose limit can be set for the patient-controlled pump. For patients whose epidural analgesia pump has to be decreased or stopped, the reasons, administered dose and subsequent management should be recorded.

***6.4 Remedial measures***

6.4.1 Failure of epidural block

6.4.1.1 For patients in the EGA group, epidural failure includes the following conditions: (1) failed multiple attempts (usually more than 5 times) of epidural puncture by a senior anesthesiologist; (2) patients refuse further epidural puncture attempts, or attending anesthesiologists consider further attempts is not beneficial; (3) no dermatomes with sensory loss appear after testing dose of epidural lidocaine (usually 10-20 min are required), and judged as failed epidural block by the attending anesthesiologists.

6.4.1.2 For patients in the EGA group with failed epidural block, anesthesia and postoperative analgesia will be performed as patients in the GA group (6.2.1 to 6.2.3).

6.4.2 Inadequate anesthesia

6.4.2.1 For patients in the GA group, inadequate anesthesia is managed by increasing intravenous and/or inhalational anesthetics.

6.4.2.2 For patients in the EGA group, inadequate anesthesia is managed with additional local anesthetics administered through the epidural catheter, and/or by increasing intravenous and/or inhalational anesthetics.

6.4.3 Unsatisfied postoperative analgesia

6.4.3.1 For patients in the GA group, the PCIA pump setting will be adjusted (increased) and/or supplemental analgesics (such as opioids, non-steroidal anti-inflammatory drugs, and others) will be administered. The above measures will be recorded.

6.4.3.2 For patients in the EGA group, the PCEA pump setting will be adjusted (increased), supplemental analgesics (such as opioids, non-steroidal anti-inflammatory drugs, and others) will be administered, or an intravenous analgesia pump will be provided. The above measures will be recorded.

***6.5 Allowed and prohibited medications***

6.5.1 For patients of both groups, no premedication (usually include anticholinergics and sedatives) is administered. Dexmedetomidine can be administered at the discretion of anesthesiologists. Anticholinergics are prohibited unless being used for the treatment of bradycardia, in which case atropine will be administered.

6.5.2 During anesthesia, vasopressors (such as ephedrine, phenylephrine, dopamine, adrenaline and norepinephrine), antihypertensives (such as urapidil and nicardipine), atropine and esmolol can be used to maintain hemodynamic stable. The target is to maintain the fluctuation of blood pressure and heart rate within 30% from baseline (average level in the ward before surgery). Low-dose glucocorticoids (usually 5-10 mg dexamethasone) and 5-hydroxytryptamine 3 (5-HT3) receptor antagonists can be used to prevent postoperative nausea and vomiting.

6.5.3 For patients admitted to the ICU with endotracheal intubation, propofol and/or midazolam can be administered for sedation; the target is to maintain the Richmond Agitation-Sedation Scale[42](#_ENREF_6_42),[43](#_ENREF_6_43) (RASS, score ranges from –5 [unarousable] to +4 [combative] and 0 indicates alert and calm) from -2 to +1. For patients admitted to the ICU without endotracheal intubation, sedatives should not be administered unless otherwise needed. Other sedatives are not allowed.

6.5.4 Other perioperative management are provided according to routine practice.

## 7. Data collection

***7.1 Baseline data***

7.1.1 Demographic data, including gender, date of birth, educational levels, and body weight index.

7.1.2 Primary diagnosis, i.e., diagnosis of lung cancer according to the International Classification of Disease-10th edition, including location and clinical tumor-node-metastasis (TNM) stage.

7.1.3 Medical comorbidity, including diagnosis, duration, severity, and medical therapy.

7.1.4 Personal history, including chronic smoking, smoking index, excessive alcohol consumption, food and drug allergy, previous anesthesia/surgery, and exposure to harmful substances (such as coal, dust, asbestos and other chemicals).

7.1.5 Cancer history in first-degree family members.

7.1.6 Results of main laboratory tests, including blood/urine/stool routine, blood biochemistry, coagulation function, and cancer biomarkers.

7.1.7 Results of imaging diagnostic examinations, including chest X-ray, chest computed tomography (CT) scan/enhanced CT scan, cranial magnetic resonance imaging (MRI)/CT scan/enhancement CT scan, positron emission tomography (PET)-CT scan, abdominal ultrasound examination/CT scan, skeleton emission computed tomography (ECT) scan, and others if available.

7.1.8 Pathological diagnosis of biopsy specimen if performed.

7.1.9 Results of instrumental examinations, including electrocardiogram, echocardiogram, and lung function examination.

7.1.10 American Society of Anesthesiologists classification and New York Heart Association classification.

***7.2 Intraoperative data***

For each participant, intraoperative data will be recorded by the anesthesiologists in charge of anesthesia.

7.2.1 Anesthesia method, types and doses of anesthetics and other medications used during anesthesia, duration of anesthesia.

7.2.2 Fluid balance (including fluid infusion, estimated blood loss, and urine output) and transfusion of blood products.

7.2.3 Surgical information, including location, type and duration of surgery, and results of frozen pathological examination if available.

7.2.4 Data of vital signs and arterial blood gas results.

7.2.5 Occurrence of adverse events and/or severe adverse events.

***7.3 In-hospital follow-ups after surgery***

7.3.1 Postoperative follow-ups will be performed by investigators who do not participate the intraoperative management. Postoperative follow-ups are performed daily (8:00-10:00 am) during the first 3 postoperative days and at hospital discharge (up to 30 days after surgery). Data in the Anesthesia Information Medical System and the Electronic Medical Record System will be achieved.

7.3.2 For patients who are admitted to ICU after surgery, the worst Acute Physiology and Chronic Health Evaluation II score (APACHE II; score ranges from 0 to 71, with higher score indicating more severe disease)[44](#_ENREF_6_44) during the first 24 hours after surgery, the percentage with endotracheal intubation, and the length of ICU stay are recorded.

7.3.3 The intensity of pain both at rest and with coughing are assessed daily (8:00-10:00 am) during the first 3 postoperative days with numeric rating scale (NRS; an 11-point scale where 0=no pain and 10=the worst pain). The final status of PCIA/PCEA pump use (completed use, dose reduction, early termination, change to other analgesic method) is recorded. The use of supplemental analgesics and other medications (including antiemetics) are recorded.

7.3.4 The occurrence of postoperative complications during hospital stay (up to 30 days) are recorded. Postoperative complications are generally defined as newly occurred medical conditions that are harmful for patients’ recovery and required therapeutic intervention, i.e., grade II or higher on the Clavien-Dindo classification.[45](#_ENREF_6_45)

7.3.5 Duration of chest-tube drainage and length of stay in hospital after surgery.

7.3.6 All-cause in-hospital mortality after surgery (up to 30 days).

7.3.7 Results postoperative pathological examination are collected and included pathological type, maximal diameter, lymph node invasion, metastasis, and results of immune-histochemical analysis (if available). The TNM stage is classified according to the 8th edition International Association for the Study of Lung Cancer (IASLC) and the American Joint Committee on Cancer (AJCC) TNM classification.[46](#_ENREF_6_46)

7.3.8 Immunohistochemical staining of CD8 and FOXP3 molecules are performed in excised lung adenocarcinoma specimens. Number of CD8+ and FOXP3+ T cells are counted per mm2 tumor area (sub-study, performed in part of enrolled patients).

7.3.9 Peripheral blood samples are collected at the beginning of surgery, at the end of surgery and 24 hours after surgery. Percentages of NK- and T-cells are measured by flow cytometry (sub-study, performed in part of enrolled patients).

***7.4 Long-term follow-ups after surgery***

7.4.1 Long-term follow-ups will be performed by investigators who do not participate in in-hospital care and follow-ups, and are blinded to study group assignment.

7.4.2 Long-term follow-ups are performed every 3 months during the first year after surgery, every 6 months during the second year after surgery, and every year thereafter. The interval is shortened in cases with advanced-stage cancer. Patients are informed of the follow-up schedule before hospital discharge and are encouraged to cooperate with the investigators. Long-term follow-ups are performed via clinic visit, telephone interview and message contact until 2 years after surgery of the last enrolled patient.

7.4.3 Data collection during long-term follow-ups includes the following:

7.4.3.1 Postoperative therapy for the primary surgical disease. For patients with confirmed lung cancer, post-surgical anti-cancer therapy may include chemotherapy, radiotherapy, targeted therapy, re-surgery, and others.

7.4.3.2 Results of postoperative re-examinations, which usually include chest radiography, computed tomography scan, positron emission tomography scan, sputum cytology, and measurement of tumor biomarkers.[47-50](#_ENREF_6_47) Cancer recurrence is defined as reappearance of the same cancer in the ipsilateral thorax, including lung and/or mediastinal/hilar lymph nodes; cancer metastasis is defined as reappearance of the same cancer in other part of the body, except the ipsilateral thorax. The development of cancer recurrence and/or metastasis will be diagnosed by surgeons (and/or radiologists when necessary) according to the results of re-examination. For patients with confirmed cancer recurrence and/or metastasis, the diagnostic evidence, date of earliest diagnoses and subsequent therapies are documented.

7.4.3.3 For patients who die during the follow-up period after surgery, the exact dates of death are recorded (consistent with death certificate). The causes of death are documented. Cancer-specific death is defined as death fully attributable to a specific cancer, i.e., the cancer for which the surgery is performed during the trial.[51](#_ENREF_6_51) Cancer-specific death usually occur after cancer recurrence and/or metastasis, with other causes including stroke, myocardial infarction or accident excluded.

7.4.3.4 For 1-year survivors, self-reported physical activities are assessed by estimating metabolic equivalents (MET; 1 MET = 3.5 ml·min-1·kg-1 resting oxygen consumption) for activity in daily life. Full physical recovery was defined as performance of moderate-intensity activity (3-5.9 METs) for at least 150 minutes a week or higher.[52](#_ENREF_6_52),[53](#_ENREF_6_53) Examples of metabolic equivalents values of moderate-intensity physical activities are illustrated as below.

Metabolic equivalents (METs) value of common moderate-intensity physical activities

|  |  |
| --- | --- |
| Activity | Approximate METs |
| **Walking** |  |
| Walking 4.8 km·h-1 | 3.0 |
| Walking at very brisk pace (6.4 km·h-1) | 5.0 |
| **Household and occupation** |  |
| Cleaning, heavy — washing windows, car, cleaning garage | 3.0 |
| Sweeping floors or carpet, vacuuming, mopping | 3.0-3.5 |
| Stair climbing, 3 floors (at least) | 4.0 |
| Carrying home products, upstairs | 5.0 |
| **Leisure time and sports** |  |
| Volleyball — non-competitive | 3.0-4.0 |
| Table tennis | 4.0 |
| Badminton — recreational | 4.5 |
| Basketball — shooting around | 4.5 |
| Tennis doubles | 5.0 |

7.4.3.5 The occurrence of chronic pain will be assessed with Brief Pain Inventory (BPI), neuropathic pain screening questionnaire (ID pain), and McGill Pain Questionnaire (MPQ) at the end of the 3rd and 6th months after surgery (sub-study, performed in part of enrolled patients).

## 8. Outcomes and assessment

***8.1 Primary outcome***

The primary outcome is recurrence-free survival after surgery, which is defined as time from surgery to cancer recurrence/metastasis or all-cause death, whichever occurs first.

***8.2 Secondary outcomes***

8.2.1 Percentage of ICU admission after surgery.

For patients admitted to the ICU, the worst APACHE II score within 24 h, the percentage with endotracheal intubation, the duration of mechanical ventilation, and the length of ICU stay are provided.

8.2.2 Incidence of postoperative complications during hospital stay (up to 30 days after surgery).

8.2.3 Duration of chest-tube drainage.

8.2.4 Length of stay in hospital after surgery.

8.2.5 In-hospital all-cause mortality (up to 30 days after surgery).

8.2.6 Engagement in activity in 1-year survivors.

8.2.7 Overall survival after surgery, which is defined as time from surgery to all-cause death.

8.2.8 Cancer-specific survival after surgery, which is defined as time from surgery to cancer-specific death, i.e., death fully attributable to lung cancer for which surgery is performed. Patients who die of a cause other than the specific lung cancer are “censored” at the time of death.

***8.3 Other predefined outcomes***

8.3.1 Pain intensity after surgery.

8.3.2 Recurrence-free survival after surgery in patients with confirmed cancer. Recurrence-free survival is defined as the time from surgery to the date of cancer recurrence/metastasis or all-cause death, whichever occurs first.

8.3.3 Overall survival after surgery in patients with confirmed cancer. Overall survival is defined as the time from surgery to all-cause death.

8.3.4 Cancer-specific survival after surgery in patients with confirmed cancer. Cancer-specific death indicates death fully attributable to lung cancer for which surgery is performed. Patients who die of a cause other than the specific lung cancer are “censored” at the time of death.

8.3.5 Number of CD8+ and FOXP3+ T cells per mm2 tumor area (sub-study, in part of enrolled patients).

8.3.6 Percentage of NK- and T-cells in peripheral blood (sub-study, in part of enrolled patients).

8.3.7 Rate of chronic pain at 3 and 6 months after surgery (sub-study, performed in part of enrolled patients).

## 9. Adverse events

***9.1 Definition***

An adverse event indicates any unpredictable, unfavorable medical event that is associated with any medical intervention and occurs during the study period. It can be related to the study intervention or otherwise. It can manifest as any uncomfortable signs (including abnormal laboratory findings), symptoms or transient morbidity.

***9.2 Predicted adverse events in this study***

9.2.1 Epidural puncture related adverse events

Adverse events related to epidural puncture/catheterization include accidental dural puncture, nerve injury, failed epidural puncture/catheterization, epidural catheter obstruction, blood appear in epidural catheter, epidural catheter dislodgement, local/epidural hematoma, local/epidural infection, etc.

9.2.2 Intraoperative adverse events related to epidural and/or general anesthesia may include the following:

|  |  |
| --- | --- |
| Local anesthetic intoxication | Caused by unexpected intravascular injection of large-dose local anesthetics, usually manifested as convulsion, twitch, loss of consciousness, arrhythmia, circulatory collapse, etc. |
| Total spinal anesthesia | Caused by unexpected intraspinal injection of large-dose local anesthetics, usually manifested as loss of sensation in all spinal innervated area, hypotension, loss of consciousness, apnea, etc. |
| New-onset arrythmia | Atrial/ventricular premature beat, supraventricular tachycardia, atrial fibrillation, etc. |
| Cardiac events | Acute coronary syndrome, ventricular fibrillation (require emergent cardiopulmonary resuscitation), etc. |
| High airway pressure | Peak airway pressure >30 cmH2O after readjusting endobronchial tube position by fiberscope re-examination, adequate muscle relaxation, and/or sputum suction. |
| CPAP to non-ventilated lung | Use of CPAP (continuous positive airway pressure) device to non-ventilated lung during one-lung ventilation to relieve desaturation (pulse oxygen saturation <88%). |
| Bradycardia | Heart rate <45 beats per minute. |
| Tachycardia | Heart rate >100 beats per minute. |
| Hypotension | Systolic blood pressure <90 mmHg or a decrease of >30% from baseline value before surgery. |
| Hypertension | Systolic blood pressure >180 mmHg or an increase of >30% from baseline value before surgery. |
| Teeth injury | Caused by the procedure of laryngoscope exposure and endotracheal intubation. |
| Prophylaxis or prophylactic shock | Caused by medications during anesthesia, usually manifested as skin rash, bronchospasm, hypotension, shock, etc. |
| Others | --- |

9.2.2 Postoperative adverse events

9.2.2.1 Adverse events related to epidural analgesia include epidural catheter obstruction, inadequate epidural analgesia, epidural catheter dislodgement, epidural hematoma, epidural abscess, sequelae of nerve injury, post-dural puncture headache, pruritus, leg weakness (muscle strength of less than grade 5), etc.

9.2.2.2 Other adverse events include nausea, vomiting, postoperative hypotension (systolic blood pressure <90 mmHg or a decrease of >30% from baseline), postoperative hypertension (systolic blood pressure >180 mmHg or an increase of >30% from baseline), postoperative bradycardia (heart rate <45 bpm), postoperative tachycardia (heart rate >100 bpm), inadequate analgesia, etc.

9.2.3 In the present study, adverse events will be monitored from the beginning of anesthesia (epidural puncture or induction of general anesthesia) until 72 hours after surgery, i.e., end of patient-controlled analgesia.

***9.3 Management***

9.3.1 Therapies will be provided according to patients’ condition and routine practice. Generally, hypotension is managed with lighting anesthesia, intravenous fluid and vasopressors; hypertension is managed with deepening anesthesia and antihypertensive drugs; bradycardia is managed with lighting anesthesia and atropine; tachycardia is managed with deepening anesthesia, intravenous fluid and esmolol when necessary.

9.3.2 The study intervention (epidural anesthesia and/or postoperative epidural/intravenous analgesia) can be stopped temporarily or permanently if considered necessarily by the attending anesthesiologist or surgeons. The time and reasons of study intervention interruption will be recorded.

9.3.3 In case that the patient-controlled analgesia (for study intervention) is terminated early, other analgesics will be provided.

***9.4 Record***

9.4.1 Any adverse event should be documented, including occurrence, type/diagnosis, time of diagnosis, management, duration of persistence, and sequelae.

9.4.2 Any adverse event should be followed up until it is completely resolved or therapy terminated.

## 10. Severe adverse events

***10.1 Definition***

A severe adverse event indicates any unpredictable medical events that lead to death, threat of life, prolonged length of hospital stays, persistent disability or dysfunction, or other severe events.

***10.2 Monitoring severe adverse events***

In the present study, severe adverse events will be monitored from the beginning of anesthesia (epidural puncture or induction of general anesthesia) until 72 hours after surgery, i.e., end of patient-controlled analgesia.

***10.3 Management***

In case of any severe adverse events, the study intervention will be stopped and treatment will be initiated immediately according to routine practice.

***10.4 Record and report***

10.4.1 In case of any severe adverse event, apart from active treatment and record as above, the principal investigator and the Clinical Research Ethics Committee of Peking University First Hospital will be informed within 24 hours in report form.

10.4.2 In case of study intervention related death, immediately stop the clinical trial, report the event to the Ethics Committee as soon as possible, record in detail and carefully preserve the related documents.

10.4.3 Any severe adverse event must be followed up until it is completely resolved or when therapy is ended.

## 11. The rule of unmasking

11.1 After the follow-up of all cases have been completed, the data of case report forms have been checked as correct, and the data entry have been finished and checked without problems, the database will be locked.

11.2 After the database is locked, the first unmasking will be conducted. And the database will be sent to the statisticians for statistical analysis.

11.3 When the statistical analysis was completed, a second unmasking will be performed to determine which group is the intervention group or the control group.

## 12. Data management

12.1 All data collected during the study period will be recorded in the case report form (CRF) by designated investigators. The investigators are trained to obey the predefined data-collection rule and complete the CRF as soon as possible after the follow-up is performed.

12.2 To ensure the correctness of database, data entry will be double checked by two personnel. Any queries during data entry will be resolved by the investigators or the principal investigator.

12.3 After data entry is completed and database is checked without problems, the database will be locked and sent for statistical analysis. The database will be analyzed by the statistician personnel.

## 13. Statistical analysis

***13.1 General principles***

13.1.1 Baseline data and outcomes will be described according the data type and distribution. Continuous variables with normal or approximate normal distribution will be expressed as mean ± standard deviation (SD), while continuous variables with non-normal distribution will be expressed as median (minimum, maximum; or interquartile range, IQR). Categorical variables will be presented as number of cases (percentage). Time-to-events variables will be presented as mean/median time (95% CI).

13.1.2 For each hypothesis, two-tailed tests will be used in all statistical analysis, and *P* <0.05 will be considered statistically significant. For the treatment-by-covariate interaction in predefined subgroup analyzes, a *P* <0.10 will be defined as statistically significant.

13.1.3 The primary and secondary outcomes will be analyzed in an intention-to-treat population, i.e., all patients are analyzed in the group to which they are randomized and received at least part of study intervention. If epidural anesthesia and/or analgesia fail, these patients will be converted to general anesthesia and intravenous analgesia but will be analyzed in the group to which they are randomized. Also, we will do per-protocol analysis for the primary endpoint, in this case, drop-out patients will be excluded.

***13.2 Patient recruitment and drop-out status***

The status of patient recruitment and drop-out will be summarized and listed. Comparison of the overall elimination/drop-out rate between the two groups will be performed with chi-square test.

***13.3 Demographics and baseline characteristics***

13.3.1 Demographics and baseline data will be presented.

13.3.2 Between-group differences are compared using the absolute standardized differences (ASDs), which are defined as the absolute difference in means, mean ranks or proportions divided by the pooled standard deviation and calculated with the formula published by Austin.[54](#_ENREF_6_54) Baseline variables with an ASD ≥0.196 (ASD =) will be considered imbalanced between the two groups and adjusted for in all analyzes if considered necessary.

***13.4 Intra- and postoperative variables***

Numeric variables will be analyzed using the independent-samples t test or Mann-Whitney U test; categorical variables will be analyzed using the chi-square test, continuity correction chi-square test or Fisher exact test. Missing data will not be replaced.

***13.5 Outcome analysis***

13.5.1 Evaluation of primary endpoint

13.5.1.1 Recurrence-free survival will be analyzed using a Kaplan-Meier estimator with difference between groups assessed by log-rank test; a Cox proportional hazard model will be used to adjust for predetermined factors including age, sex, chronic smoking, American Society of Anesthesiologist (ASA) physical status classification, TNM stage, and postoperative anti-cancer therapy. These factors are preselected according to clinical importance. Effect size will be expressed as hazard ratio and its 95% CI. Patients who are lost to follow-up will be censored at the time of last contact.

13.5.1.2 The differences of the primary outcome in predefined subgroups will be tested using the Cox proportional hazard models adjusted for preselected factors of age, sex, chronic smoking, American Society of Anesthesiologists physical status, TNM stage, and postoperative anti-cancer therapy. Treatment-by-covariate interactions will be assessed separately for each predefined factor, adjusting for the same variables. The hazard ratio (and 95% CI) for each subgroup and the *P* values of treat-by-covariate interactions will be displayed in a forest plot.

13.5.2 Evaluation of secondary and other outcomes

13.5.2.1 Long-term survival variables (overall survival, cancer-specific survival) will be analyzed using a Kaplan-Meier estimator with differences between groups assessed by log-rank test; Cox proportional hazards models will be used to adjust for predetermined factors including age, sex, chronic smoking, ASA physical status classification, TNM stage, and postoperative anti-cancer therapy. The estimated hazard ratio and 95% CI will be provided. For cancer-specific survival, patients who die of a cause other than the specific cancer will be censored at the time of death. Patients who are lost to follow-up will be censored at the time of last contact.

13.5.2.2 Long-term survival variables in the subgroup of cancer patients (recurrence-free survival, overall survival, and cancer-specific survival) will be analyzed using a Kaplan-Meier estimator with differences between groups assessed by log-rank test; Cox proportional hazards models will be used to adjust for predetermined factors including age, sex, chronic smoking, ASA physical status classification, TNM stage, and postoperative anti-cancer therapy. The estimated hazard ratio and 95% CI will be provided. For cancer-specific survival, patients who die of a cause other than the specific cancer will be censored at the time of death. Patients who are lost to follow-up will be censored at the time of last contact.

13.5.2.3 Other time-to-event variables (duration of chest-tube drainage, length of stay in hospital) will be analyzed using a Kaplan-Meier estimator with differences between groups assessed by log-rank test. The estimated hazard ratio and 95% CI will be provided. Patients who are lost to follow-up will be censored at the time of last contact.

13.5.2.4 Categorical variables (percentage of ICU admission after surgery, incidence of postoperative complication, in-hospital mortality, activity engagement in 1-year survivor, and rate of chronic pain [sub-study]) will be analyzed using the chi-square test, continuity correction chi-squared test or Fisher’s exact test. The estimated risks ratio and 95% CI will be provided. Missing data will not be replaced.

13.5.2.5 Discrete variables (NRS pain scores, numbers of infiltrated CD8+ and FOXP3+ T cells [sub-study]) will be analyzed with the Mann-Whitney U test. Median difference and 95% CI will be calculated with the Hodges-Lehmann estimator. Missing data will not be replaced.

13.5.2.6 Numeric variables (percentages of NK- and T-cells in peripheral blood [sub-study]) will be analyzed using the independent-samples t test or Mann-Whitney U test. Mean/median difference and 95% CI will be calculated. Median difference and 95% CI will be calculated with the Hodges-Lehmann estimator. Missing data will not be replaced.

***13.6 Safety analysis***

13.3.1 Describe the occurrence of adverse events in each group.

13.3.2 Describe the management of adverse events when appropriate.

13.3.3 Describe the occurrence of severe adverse events.

13.3.4 The rates of adverse events and/or managements between the two groups will be compared with chi-square test, continuity correction chi-square test or Fisher exact test. The estimated risks ratio (RR) and 95% CI will be provided.

13.3.5 Missing data will not be replaced.

## 14. Quality control and quality assurance

14.1.1 An investigator training program will be designed by the principle investigator. A study coordinator will be designated to organize and implement the training program, and to record and preserve the related documents.

14.1.2 The training program includes the following:

14.1.2.1 For anesthesiologists, they should be familiar with the protocol, the procedures of radical lung cancer surgery, the procedures of anesthesia and perioperative care, and the case report form.

14.1.2.2 For in-hospital follow-up investigators, they should be familiar with the potential adverse events or severe adverse event, postoperative complications, as well as the case report form.

14.1.2.3 For long-term outcome follow-up investigators, they are trained to communicate with participants and collect data regarding cancer progression and/or living status regularly (every 3 months during the first year, every 6 months during the second year, and every year thereafter); they are required to closely communicate with thoracic surgeons regarding the diagnosis of cancer outcomes. Furthermore, they do not participate in perioperative care and in-hospital follow-ups, have no knowledge of study group allocation, and are not permitted to communicate with health-care providers, participants and other investigators regarding study group allocation.

14.1.2.4 Other training contents that must be completed before trial initiation include the Good Clinical Practice principles, the standard operating procedures of the study, the working plan of the study, the data recording format, the outcome assessment, the instruction for the case report form, the skills of communicating with subjects, and other matters needing attention during the study (screening path, allowed and prohibited medications, etc.).

14.1.3 The instruments, equipment, assessment scales and definitions involved in the trial should be used according to executive standards. Investigators should stick to the preselected tools/definitions in order to achieve uniform evaluation and assessment.

14.1.4 Investigators who are responsible to record data must be authorized by the principle investigator in advance. Data sources include, but not limits to, electronic health records, anesthesia medical information system, medical equipment, as well as interviews before surgery and during postoperative (long-term) follow-ups.

14.1.5 The investigator should fill in the case report forms timely and completely. All observed and assessed results as well as abnormal findings during the study period should be verified and recorded to ensure the reliability of data.

14.1.6 Dataset management and statistical analysis are performed by professional biostatistics personnel.

14.1.7 All conclusions should be based on original data.

## 15. Ethical requirements and patient informed consent

***15.1 Ethics Committee***

The study protocol must be approved by the Clinical Research Ethics Committee of Peking University First Hospital before the study can be started. The investigators must strictly follow the Helsinki Declaration and China's relevant clinical trial management regulations. The principal investigator is responsible to report the status and the progress of the study to the Ethics Committee.

***15.2 Written informed consent***

For each potential participant, investigators are responsible to fully explain the purpose, procedures and possible risks of this study in a written form manner. The investigators must let every potential participant know that he/she has the right to withdraw consent from the study at any time. Every potential participant must be given a written informed consent. Every participant or the authorized surrogate of the participant must sign the consent before he/she can be enrolled in the study. The written informed consents will be kept as a part of the clinical trial documents.

***15.3 Privacy and confidentiality***

15.3.1 During the study period, the collected data from participants are labelled with special recruitment numbers and acronyms of names.

15.3.2 All personal information of the participants will be kept confidential and cannot be copied. The filing cabinets storing the study documents will be locked. Apart from the study investigators, only authorized inspectors from the Clinical Research Ethics Committee of Peking University First Hospital are allowed to access the information after obtaining consents from the participants.

15.3.3 Results of the study will be published as scientific articles, but all personal data (including name and age, etc.) are strictly confidential.

## 16. Study termination

16.1 In case that intervention-related severe adverse events or serious quality problem occur during the study period, the study will be stopped. A report will be sent to the Ethics Committee. Restart of the study will need an approval from the Ethics Committee.

16.2 The study will be terminated after accomplishment of required patient recruitment and data collection. Decision will be made by the principal investigator.

## 17. Document preservation

Investigators and sponsors should properly keep clinical trial documents and data for 5 years in accordance with Good Clinic Practice requirements.

## 18. Conflict of interest

The investigators declare no conflicts of interest.

Patients planned for lung cancer surgery

Screening according to inclusion/exclusion criteria

Randomization

N = 400

General anesthesia and postoperative intravenous analgesia (GA group)

In-hospital follow-up

Combined epidural-general anesthesia and postoperative epidural analgesia (EGA group)

Estimated epidural failure rate of 2%

In-hospital follow-up

Informed consent

**Enrollment**

ITT analysis (N= )

PP analysis (N= )

ITT analysis (N= )

PP analysis (N= )

Long-term follow-up

Estimated dropout rate of 4%

Long-term follow-up

Estimated dropout rate of 4%

**Allocation**

**Follow-up**

**Analysis**

**Figure 1.** Planned flow chart. ITT=intention-to-treat; PP=per-protocol.

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# Summary of changes from the original study protocol approved by the ethics committee

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Number** | **Version** | **Date of version** | **Drafter, Reviser/**  **Chairman** | **Contents of revision** | **Reasons for revision** |
| 1 | 1.1 | Nov. 24, 2013 | Z-ZX, D-XW/Y-NH | (This was the first version approved by the Ethics Committee). | --- |
| 2 | 1.2 | Dec. 8, 2015 | Z-ZX, D-XW/Y-NH | 1. Closed one participating center (Peking University Cancer Hospital) (clause 3.3.1). | 1. No patient was enrolled since trial initiation in this participating center. |
|  |  |  |  | 2. Deleted research contents: “Serum concentration of cortisol, interleukin-6 (IL-6) and IL-8… the day before surgery” (original clause 7.1.11) and “… postoperative days 1 and 3” (original clause 7.3.4). Deleted these variables in secondary outcomes (original clause 8.2.2) and statistical analysis (original clause 13.5.2.2). | 2. To improve patients’ compliance. |
|  |  |  |  | 3. Deleted research contents: “Delirium is assessed twice daily (8:00-10:00 am, 18:00-20:00 pm) during the first 7 postoperative days …” (original clause 7.3.5). Deleted this variable in secondary outcomes (original clause 8.2.3) and statistical analysis (original clause 13.5.2.3). | 3. To reduce workload of the investigators. |
|  |  |  |  | 4. Reduced frequency of postoperative follow-up: “Postoperative follow-ups are performed daily (8:00-10:00 am) during the first 3 postoperative days …” (clause 7.3.1); “The intensity of pain both at rest and with coughing are assessed daily (8:00-10:00 am) during the first 3 postoperative days …” (clause 7.3.3). | 4. To reduce workload of the investigators. |
|  |  |  |  | 5. Added research contents (sub-study, performed in part of enrolled patients): “… Number of CD8+ and FOXP3+ T cells are counted per mm2 tumor area” (clause 7.3.8); “… Percentages of NK- and T-cells are measured by flow cytometry” (clause 7.3.9). Added these variables in “Other predefined outcomes” (clauses 8.3.5 and 8.3.6). Added methods of statistical analysis for these variables (clauses 13.5.2.5 and 13.5.2.6). | 5. To explored the effects of epidural anesthesia-analgesia on immune function in patients after lung cancer surgery. |
|  |  |  |  | 6. Added research contents: “For 1-year survivors, self-reported physical activities are assessed by estimating metabolic equivalents …” (clause 7.4.3.4). Added this variable in “Secondary outcomes” (clause 8.2.6). Added method of statistical analysis for this variable (clause 13.5.2.4). | 6. To investigate the effect of epidural anesthesia-analgesia on physical status in 1-year survivors. |
|  |  |  |  | 7. Added research contents (sub-study, performed in part of enrolled patients): “The occurrence of chronic pain will be assessed … at the end of the 3rd and 6th months after surgery” (clause 7.4.3.5). Added these variables in “Other predefined outcomes” (clause 8.3.7). Added methods of statistical analysis for these variables (clause 13.5.2.4). | 7. To investigate the effect of epidural anesthesia-analgesia on the development of chronic pain after surgery. |
| 3 | 1.3 | May 12, 2017 | Z-ZX, D-XW/Y-MY | We applied to extend the duration of patient recruitment. | We did not finish recruiting 400 patients in 24 months. |
| 4 | 1.4 | Sep. 22, 2017 | Z-ZX, D-XW/R-HY | 1. Re-clarified that “Long-term follow-ups are performed … until 2 years after surgery of the last enrolled patient” (clause 7.4.2). | 1. All enrolled patients will be followed up until a same time-point. |
|  |  |  |  | 2. Changed primary outcome from “rate of recurrence/metastasis after surgery” (original clause 8.1) to “recurrence-free survival after surgery, which is defined as time from surgery to cancer recurrence/metastasis or all-cause death, whichever occurs first” (clause 8.1). Clarified the method of statistical analysis for the primary outcome (clause 13.5.1). | 2. Some patients did not come/go back to hospital for re-examination and died. Recurrence-free survival is, therefore, a more proper primary outcome. |
|  |  |  |  | 3. In “Secondary outcomes” section, changed “rate of all-cause mortality after surgery” (original clause 8.2.8) to “overall survival after surgery, …” (clause 8.2.7); changed “rate of cancer-specific death after surgery” (original clause 8.2.9) to “cancer-specific survival after surgery, …” (clause 8.2.8). Clarified the method of statistical analysis for these variables (clause 13.5.2.1). | 3. “Overall survival” and “cancer-specific survival” have same meanings as previous terms but are more commonly used to express long-term outcomes after cancer surgery. |

# Original statistical analysis plan

Original statistical analysis plan as reported in the original trial protocol.

**1. Sample size calculation**

In a cohort study of patients after complete resection for NSCLC, Taylor and colleagues reported a recurrence rate of 33% at a median follow-up of 2 years after surgery.1 In a pilot study of our own, the 1-year recurrence rate after lung cancer surgery was 48% lower in patients who were given combined epidural-general anesthesia compared with those given general anesthesia alone. We expect that patient recruitment and follow-up will be completed in about 24 months, respectively. With the 2-sided significance level set at 0.05 and power at 80%, 360 patients (180 per group) are required to detect the difference. Considering a dropout rate of about 8% and an epidural failure rate of 2%, we plan to enroll 400 patients.

**2. Outcomes**

***2.1 Primary Outcome***

The primary outcome is rate of recurrence/metastasis after surgery.

***2.2 Secondary Outcome***

2.2.1 Pain intensity at postoperative days 1-3.

2.2.2 Serum concentrations of cortisol, IL-6 and IL-8 in the morning of postoperative days 1 and 3.

2.2.3 Incidence of postoperative delirium within the first 7 postoperative days.

2.2.4 Incidence of postoperative complications during hospital stay (up to 30 days after surgery).

2.2.5 Duration of chest-tube drainage.

2.2.6 Length of stay in hospital after surgery.

2.2.7 In-hospital all-cause mortality (up to 30 days after surgery).

2.2.8 Rate of all-cause mortality after surgery.

2.2.9 Rate of cancer-specific death after surgery.

# 3. Statistical analysis

***3.1 General principles***

3.1.1 Baseline data and outcomes will be described according the data type and distribution. Continuous variables with normal or approximate normal distribution will be expressed as mean ± standard deviation (SD), while continuous variables with non-normal distribution will be expressed as median (minimum, maximum; or interquartile range, IQR). Categorical variables will be presented as number of cases (percentage). Time-to-events variables will be presented as mean/median time (95% CI).

3.1.2 For each hypothesis, two-tailed tests will be used in all statistical analysis, and *P* <0.05 will be considered statistically significant.

3.1.3 The primary and secondary outcomes will be analyzed in an intention-to-treat population, i.e., all patients are analyzed in the group to which they are randomized and received at least part of study intervention. If epidural anesthesia and/or analgesia fail, these patients will be converted to general anesthesia and intravenous analgesia but will be analyzed in the group to which they are randomized. Also, we will do per-protocol analysis for the primary endpoint, in this case, drop-out patients will be excluded.

***3.2 Patient recruitment and drop-out status***

The status of patient recruitment and drop-out will be summarized and listed. Comparison of the overall elimination/drop-out rate between the two groups will be performed with chi-square test.

***3.3 Demographics and baseline characteristics***

3.3.1 Demographics and baseline data will be presented.

3.3.2 For between-group differences, continuous variables will be analyzed using independent-samples t test or Mann-Whitney U test; categorical variables will be analyzed using the chi-square test, continuity correction chi-square test or Fisher exact test.

***3.4 Intra- and postoperative variables***

Numeric variables will be analyzed using the independent-samples t test or Mann-Whitney U test; categorical variables will be analyzed using the chi-square test, continuity correction chi-square test or Fisher exact test. Missing data will not be replaced.

***3.5 Outcome analysis***

3.5.1 Evaluation of primary endpoint

3.5.1.1 Cancer recurrence/metastasis after surgery will be analyzed using a Kaplan-Meier estimator with difference between groups assessed by log-rank test; a Cox proportional hazards model will be used to adjust for potential confounding factors. Effect size will be expressed as hazard ratio and its 95% CI. Patients who are lost to follow-up will be censored at the time of last contact.

3.5.2 Evaluation of secondary outcomes

3.5.2.1 Discrete variables (NRS pain scores) will be analyzed with the Mann-Whitney U test. Median difference and 95% CI will be calculated with the Hodges-Lehmann estimator. Missing data will not be replaced.

3.5.2.2 Numeric variables (serum concentrations of cortisol, IL-6 and IL-8) will be analyzed using the independent-samples t test or Mann-Whitney U test. Mean/median difference and 95% CI will be calculated. Median difference and 95% CI will be calculated with the Hodges-Lehmann estimator. Missing data will not be replaced.

3.5.2.3 Categorical variables (incidence of postoperative delirium, incidence of postoperative complication, in-hospital mortality) will be analyzed using the chi-square test, continuity correction chi-squared test or Fisher’s exact test. The estimated risks ratio and 95% CI will be provided. Missing data will not be replaced.

3.5.2.4 Time-to-event variables (duration of chest-tube drainage, length of stay in hospital) will be analyzed using a Kaplan-Meier estimator with difference between groups assessed by log-rank test. The estimated hazard ratio and 95% CI will be provided. Patients who are lost to follow-up will be censored at the time of last contact.

3.5.2.5 Long-term mortality (all-cause death after surgery, cancer-specific death after surgery) will be analyzed using a Kaplan-Meier estimator with difference between groups assessed by log-rank test; Cox proportional hazards models will be used to adjust for potential confounding factors. The estimated hazard ratio and 95% CI will be provided. For cancer-specific death, patients who die of a cause other than the specific cancer will be censored at the time of death. Patients who are lost to follow-up will be censored at the time of last contact.

***3.6 Safety analysis***

3.6.1 Describe the occurrence of adverse events in each group.

3.6.2 Describe the management of adverse events when appropriate.

3.6.3 Describe the occurrence of severe adverse events.

3.6.4 The rates of adverse events and/or managements between the two groups will be compared with chi-square test, continuity correction chi-square test or Fisher exact test. The estimated risks ratio and 95% CI will be provided.

3.6.5 Missing data will not be replaced.

**4. References**

1 Taylor MD, Nagji AS, Bhamidipati CM, et al. Tumor recurrence after complete resection for non-small cell lung cancer. Ann Thorac Surg 2012; 93: 1813-20; discussion 20-1.

# Final statistical analysis plan

Final statistical analysis plan was completed prior to outcome analysis.

**1. Sample size calculation**

In a cohort study of patients after complete resection for NSCLC, Taylor and colleagues reported a recurrence rate of 33% at a median follow-up of 2 years after surgery.1 In a pilot study of our own, the 1-year recurrence rate after lung cancer surgery was 48% lower in patients who were given combined epidural-general anesthesia compared with those given general anesthesia alone. We expect that patient recruitment and follow-up will be completed in about 24 months, respectively. With the 2-sided significance level set at 0.05 and power at 80%, 360 patients (180 per group) are required to detect the difference. Considering a dropout rate of about 8% and an epidural failure rate of 2%, we plan to enroll 400 patients.

**2. Outcomes**

***2.1 Primary outcome***

The primary outcome is recurrence-free survival after surgery, which is defined as time from surgery to cancer recurrence/metastasis or all-cause death, whichever occurs first.

***2.2 Secondary outcome***

2.2.1 Percentage of ICU admission after surgery.

For patients admitted to the ICU, the worst APACHE II score within 24 h, the percentage with endotracheal intubation, the duration of mechanical ventilation, and the length of ICU stay are provided.

2.2.2 Incidence of postoperative complications during hospital stay (up to 30 days after surgery).

2.2.3 Duration of chest-tube drainage.

2.2.4 Length of stay in hospital after surgery.

2.2.5 In-hospital all-cause mortality (up to 30 days after surgery).

2.2.6 Engagement in activity in 1-year survivors.

2.2.7 Overall survival after surgery, which is defined as time from surgery to all-cause death.

2.2.8 Cancer-specific survival after surgery, which is defined as time from surgery to cancer-specific death, i.e., death fully attributable to lung cancer for which surgery is performed. Patients who die of a cause other than the specific lung cancer are “censored” at the time of death.

***2.3 Other pre-specified outcome***

2.3.1 Pain intensity after surgery.

2.3.2 Recurrence-free survival after surgery in patients with confirmed cancer. Recurrence-free survival is defined as the time from surgery to the date of cancer recurrence/metastasis or all-cause death, whichever occurs first.

2.3.3 Overall survival after surgery in patients with confirmed cancer. Overall survival is defined as the time from surgery to all-cause death.

2.3.4 Cancer-specific survival after surgery in patients with confirmed cancer. Cancer-specific death indicates death fully attributable to lung cancer for which surgery is performed. Patients who die of a cause other than the specific lung cancer are “censored” at the time of death.

2.3.5 Number of CD8+ and FOXP3+ T cells per mm2 tumor area (sub-study, in part of enrolled patients).

2.3.6 Percentage of NK- and T-cells in peripheral blood (sub-study, in part of enrolled patients).

2.3.7 Rate of chronic pain at 3 and 6 months after surgery (sub-study, in part of enrolled patients).

**3. Statistical analysis**

***3.1 General principles***

3.1.1 Baseline data and outcomes will be described according the data type and distribution. Continuous variables with normal or approximate normal distribution will be expressed as mean ± standard deviation (SD), while continuous variables with non-normal distribution will be expressed as median (minimum, maximum; or interquartile range, IQR). Categorical variables will be presented as number of cases (percentage). Time-to-events variables will be presented as mean/median time (95% CI).

3.1.2 For each hypothesis, two-tailed tests will be used in all statistical analysis, and *P* <0.05 will be considered statistically significant. For the treatment-by-covariate interaction in predefined subgroup analyzes, a *P* <0.10 will be defined as statistically significant.

3.1.3 The primary and secondary outcomes will be analyzed in an intention-to-treat population, i.e., all patients are analyzed in the group to which they are randomized and received at least part of study intervention. If epidural anesthesia and/or analgesia fail, these patients will be converted to general anesthesia and intravenous analgesia but will be analyzed in the group to which they are randomized. Also, we will do per-protocol analysis for the primary endpoint, in this case, drop-out patients will be excluded.

***3.2 Patient recruitment and drop-out status***

The status of patient recruitment and drop-out will be summarized and listed. Comparison of the overall elimination/drop-out rate between the two groups will be performed with chi-square test.

***3.3 Demographics and baseline characteristics***

3.3.1 Demographics and baseline data will be presented.

3.3.2 Between-group differences are compared using the absolute standardized differences (ASDs), which are defined as the absolute difference in means, mean ranks or proportions divided by the pooled standard deviation and calculated with the formula published by Austin.[2](#_ENREF_4_54) Baseline variables with an ASD ≥0.196 (ASD =) will be considered imbalanced between the two groups and adjusted for in all analyzes if considered necessary.

***3.4 Intra- and postoperative variables***

Numeric variables will be analyzed using the independent-samples t test or Mann-Whitney U test; categorical variables will be analyzed using the chi-square test, continuity correction chi-square test or Fisher exact test. Missing data will not be replaced.

***3.5 Outcome analysis***

3.5.1 Evaluation of primary endpoint

3.5.1.1 Recurrence-free survival after surgery will be analyzed using a Kaplan-Meier estimator with difference between groups assessed by log-rank test; a Cox proportional hazard model will be used to adjust for predetermined factors including age, sex, chronic smoking, American Society of Anesthesiologist (ASA) physical status classification, TNM stage, and postoperative anti-cancer therapy. These factors are preselected according to clinical importance. Effect size will be expressed as hazard ratio and its 95% CI. Patients who are lost to follow-up will be censored at the time of last contact.

3.5.1.2 The differences of the primary outcome in predefined subgroups will be tested using the Cox proportional hazard models adjusted for preselected factors of age, sex, chronic smoking, American Society of Anesthesiologists physical status, TNM stage, and postoperative anti-cancer therapy. Treatment-by-covariate interactions will be assessed separately for each predefined factor, adjusting for the same variables. The hazard ratio (and 95% CI) for each subgroup and the *P* values of treat-by-covariate interactions will be displayed in a forest plot.

3.5.2 Evaluation of secondary and other outcomes

3.5.2.1 Long-term survival variables (overall survival, cancer-specific survival) will be analyzed using a Kaplan-Meier estimator with differences between groups assessed by log-rank test; Cox proportional hazards models will be used to adjust for predetermined factors including age, sex, chronic smoking, ASA physical status classification, TNM stage, and postoperative anti-cancer therapy. The estimated hazard ratio and 95% CI will be provided. For cancer-specific survival, patients who die of a cause other than the specific cancer will be censored at the time of death. Patients who are lost to follow-up will be censored at the time of last contact.

3.5.2.2 Long-term survival variables in the subgroup of cancer patients (recurrence-free survival, overall survival, and cancer-specific survival) will be analyzed using a Kaplan-Meier estimator with differences between groups assessed by log-rank test; Cox proportional hazards models will be used to adjust for predetermined factors including age, sex, chronic smoking, ASA physical status classification, TNM stage, and postoperative anti-cancer therapy. The estimated hazard ratio and 95% CI will be provided. For cancer-specific survival, patients who die of a cause other than the specific cancer will be censored at the time of death. Patients who are lost to follow-up will be censored at the time of last contact.

3.5.2.3 Other time-to-event variables (duration of chest-tube drainage, length of stay in hospital) will be analyzed using a Kaplan-Meier estimator with differences between groups assessed by log-rank test. The estimated hazard ratio and 95% CI will be provided. Patients who are lost to follow-up will be censored at the time of last contact.

3.5.2.4 Categorical variables (percentage of ICU admission after surgery, incidence of postoperative complication, in-hospital mortality, engagement in activity in 1-year survivors, and rate of chronic pain [sub-study]) will be analyzed using the chi-square test, continuity correction chi-squared test or Fisher’s exact test. The estimated risks ratio and 95% CI will be provided. Missing data will not be replaced.

3.5.2.5 Discrete variables (NRS pain scores, numbers of infiltrated CD8+ and FOXP3+ T cells [sub-study]) will be analyzed with the Mann-Whitney U test. Median difference and 95% CI will be calculated with the Hodges-Lehmann estimator. Missing data will not be replaced.

3.5.2.6 Numeric variables (percentages of NK- and T-cells in peripheral blood [sub-study]) will be analyzed using the independent-samples t test or Mann-Whitney U test. Mean/median difference and 95% CI will be calculated. Median difference and 95% CI will be calculated with the Hodges-Lehmann estimator. Missing data will not be replaced.

***3.6 Safety analysis***

3.6.1 Describe the occurrence of adverse events in each group.

3.6.2 Describe the management of adverse events when appropriate.

3.6.3 Describe the occurrence of severe adverse events.

3.6.4 The rates of adverse events and/or managements between the two groups will be compared with chi-square test, continuity correction chi-square test or Fisher exact test. The estimated risks ratio (RR) and 95% CI will be provided.

3.6.5 Missing data will not be replaced.

**4. References**

1 Taylor MD, Nagji AS, Bhamidipati CM, et al. Tumor recurrence after complete resection for non-small cell lung cancer. Ann Thorac Surg 2012; 93: 1813-20; discussion 20-1.

2 Austin PC. An Introduction to Propensity Score Methods for Reducing the Effects of Confounding in Observational Studies. Multivariate Behav Res 2011; 46: 399-424.

# Summary of revision from the original statistical analysis plan

In “General principles” section, we added the significant level for treat-by-covariate interaction: “For the treatment-by-covariate interaction in predefined subgroup analyzes, a p<0.10 will be defined as statistically significant” (clause 3.1.2).

In “Demographics and baseline characteristics” section, we changed method to compare between-group differences: “Between-group differences are compared using the absolute standardized differences (ASDs), which are defined as the absolute difference in means, mean ranks or proportions divided by the pooled standard deviation and calculated with the formula published by Austin.[1](#_ENREF_4_54) Baseline variables with an ASD ≥0.196 (ASD =) will be considered imbalanced between the two groups and adjusted for in all analyzes if considered necessary” (clause 3.3.2).

In the “Evaluation of primary outcome” section, we changed primary outcome from “Cancer recurrence/metastasis after surgery …” (original clause 3.5.1.1) to “recurrence-free survival after surgery, …” (clause 3.5.1.1); because some patients did not come/go back to hospital for re-examination and died. Recurrence-free survival is, therefore, a more proper primary outcome. We clarified factors that will be included in the Cox proportional hazard model for adjustment: “…; a Cox proportional hazard model will be used to adjust for predetermined factors including age, sex, chronic smoking, American Society of Anesthesiologist (ASA) physical status classification, TNM stage, and postoperative anti-cancer therapy” (clause 3.5.1.1). We added subgroup analysis for the assessment of treatment-by-covariate interaction: “The differences of the primary outcome in predefined subgroups will be tested using the Cox proportional hazard models adjusted for preselected factors of age, sex, chronic smoking, American Society of Anesthesiologists physical status, TNM stage, and postoperative anti-cancer therapy. Treatment-by-covariate interactions will be assessed separately for each predefined factor, adjusting for the same variables. The hazard ratio (and 95% CI) for each subgroup and the *P* values of treat-by-covariate interactions will be displayed in a forest plot” (clause 3.5.1.2).

In the “Evaluation of secondary and other outcomes” section, we changed “Long-term mortality (all-cause death after surgery, cancer-specific death after surgery) …” (original clause 3.5.2.5) to “Long-term survival variables (overall survival, cancer-specific survival), …” (clause 3.5.2.1). These terms have same meanings but the latter ones are more commonly used to express long-term outcomes after cancer surgery. We clarified factors that will be included in Cox proportional hazard models for adjustment: “…; Cox proportional hazards models will be used to adjust for predetermined factors including age, sex, chronic smoking, ASA physical status classification, TNM stage, and postoperative anti-cancer therapy” (clause 3.5.2.1).

In the “Evaluation of secondary and other outcomes” section, we added methods to analyze long-term survival variables in the subgroup of cancer patients: “Long-term survival variables in the subgroup of cancer patients (recurrence-free survival, overall survival, and cancer-specific survival after surgery) will be analyzed using a Kaplan-Meier estimator with difference between groups assessed by log-rank test; Cox proportional hazards models will be used to adjust for predetermined factors including age, sex, chronic smoking, ASA physical status classification, TNM stage, and postoperative anti-cancer therapy. The estimated hazard ratio and 95% CI will be provided. For cancer-specific survival, patients who die of a cause other than the specific cancer will be censored at the time of death. Patients who are lost to follow-up will be censored at the time of last contact” (clause 3.5.2.2). To be noted, long-term survival variables in the subgroup of cancer patients are added after data collection, but before analysis.

In categorical variables of the “Evaluation of secondary and other outcomes” section, we deleted “incidence of postoperative delirium” but added “percentage of ICU admission after surgery, engagement in activity in 1-year survivors, and rate of chronic pain [sub-study]” (clause 3.5.2.4).

In discrete variables of the “Evaluation of secondary and other outcomes” section, we added “numbers of infiltrated CD8+ and FOXP3+ T cells [sub-study]” (clause 3.5.2.5).

In numeric variables of the “Evaluation of secondary and other outcomes” section, we deleted “serum concentrations of cortisol, IL-6 and IL-8” but added “percentages of NK- and T-cells in peripheral blood [sub-study]” (clause 3.5.2.6).

**Reference**

1 Austin PC. An Introduction to Propensity Score Methods for Reducing the Effects of Confounding in Observational Studies. Multivariate Behav Res 2011; 46: 399-424.