

## Peer Review File

Article Information: <https://dx.doi.org/10.21037/jtd-23-1794>

### Reviewer A

**Comment 1:** Abstract. Results: Patients undergoing resection of four or more lung13 subsegments had a continuously elevated pepsin concentration in saliva on postoperative day 3 and at discharge. Please add some statistically significant data to support the results.

**Reply1:** Thank you for your comment. We added statistically significant data to the result section in Abstract.

**Change in the text:** Line 10 to Line 12 in abstract.

**Comment 2:** Abstract. Conclusion: Resection of large volumes of lung may lead to changes in breathing patterns and result in GER. Abstract might be beneficial to include a sentence that briefly summarizes the key findings of the study. This can provide readers with a quick overview of the research.

**Reply 2:** Thank you for your kind comment. We added the following statement to the conclusion in abstract. “Lung resection resulted in elevated pepsin concentration in the saliva, which persisted in patients who received resections equivalent to or more than right middle lobectomy in volume.”

**Change in the text:** Line 14 to Line 16 in abstract

**Comment 3:** Keywords: lung resection, pepsin, GER. Please, add some keywords

**Reply 3:** Thank you for your comment. We added “Peptest”, “salivary”, “Frequency Scale for the Symptoms of GERD (FSSG)”.

**Comment 4:** Introduction. Patients with lung disease reportedly have a high incidence of GERD. Please, improve this paragraph and discuss some references:

- a) Circulating inflammatory cytokines and risk of idiopathic pulmonary fibrosis: a Mendelian randomization study. BMC Pulm Med. 2023 Oct 3;23(1):369. doi: 10.1186/s12890-023-02658-3.
- b) Epithelial-Mesenchymal Transition: A Major Pathogenic Driver in Idiopathic Pulmonary Fibrosis? Medicina (Kaunas). 2020 Nov 13;56(11):608. doi: 10.3390/medicina56110608.
- c) Genetic association analysis of dietary intake and idiopathic pulmonary fibrosis: a two-sample mendelian randomization study. BMC Pulm Med. 2024 Jan 5;24(1):15. doi: 10.1186/s12890-023-02831-8.

**Reply 4:** Thank you for your comment. We changed Introduction to one that cites the three papers as follow. “Interstitial lung disease and COPD frequently coexist in patients undergoing lung surgery for lung tumors. Understanding and preventing factors associated with disease progression and exacerbation may lead to improved prognosis in these patients. Studies on Idiopathic Pulmonary Fibrosis have reported higher inflammatory cytokines such as Interleukin-14 (1), Epithelial-Mesenchymal transition following injury due to defective alveolar type-II cells (2) and dietary intake such as alcohol and beef (3), to be associated with pathogenesis and disease progression.”

**Change in the text:** p1 Line 2 - 8 in text.

**Comment 5:** Further understanding of the incidence of and risk factors for GERD in patients who have undergone lung resection may lead to preventive strategies against GERD and silent aspiration, in turn improving the postoperative course of patients with underlying lung disease. Please, improve the description of study aim.

**Reply 5:** Thank you for your comment. We improved the description of study aim as follows. “The aim of this study is to reveal the incidence of GERD in patients who have undergone lung resection and to ascertain risk factors of GERD, such as underlying lung disease, operative site and type of resection.”

**Change in the text:** p2 Line 5 - 7 in text.

**Comment 6:** The primary endpoint of this study was the frequency of GER following lung resection. The secondary endpoints were the pepsin positivity rate in saliva by type of surgery and the relationship with underlying lung disease, PFT results, and the pepsin positivity rate. Statistical analysis was performed with GraphPad Prism (GraphPad Software, San Diego, CA, USA). A p value of <0.05 was considered statistically significant. Please, improve the description of statistical tests used to evaluate the data.

**Reply 6:** Thank you for your comment. We added the statistical tests used in this study as follow. “Paired t tests were performed for changes in FSSG and pepsin concentration in saliva preoperatively and at discharge. One-way repeated measure ANOVA was used for analysis in changes in pepsin concentration at all time points and Two-way repeated measure ANOVA for pepsin concentration according to resected number of subsegments. Dunnett’s multiple comparison test was performed when comparing each time point with the preoperative value.”

**Change in the text:** p 4 Line 3 - 8 in text

**Comment 7:** Results. Please, underline the most important statistically significant data to support the conclusions.

**Reply 7:** Thank you for your comment. I have underlined the most important data in the Results.

**Change in the text:** p 5 Line 3 - 9 in text

**Comment 8:** Discussion. The standard procedure for studying GER is 24-hour pH monitoring of the esophagus. A study on chronic cough following lung resection showed that patients with a nonproductive cough in the postoperative period had a high probability of GER on 24-hour pH monitoring (5). The discussion section needs to be improved. It could be interesting to clarify the results obtained and compare them with previous or similar articles.

**Reply 8:** The Discussion section has been changed to first discuss the association of GERD with structural changes after lung resection, followed by a discussion about previous studies on the incidence and risk factors for GER after lung resection. We have added the sentences in the second paragraph.

**Change in the text:** p 6 Line 5 – 9 in text.

## **Reviewer B**

**Comment 1:** Lack of a control group: The absence of a control group makes it difficult to accurately evaluate changes in salivary pepsin concentration induced by surgery. The wide range of surgical techniques, including different types of lung resection such as wedge resection and lobectomy, as well as the possibility of manipulation around the vagus nerve during lymph node dissection, makes it unclear the specific impact of lung resection alone.

**Reply 1:** Thank you for your valuable comment. We appreciate your understanding that this is a small, one-arm, prospective study in a single-institute, which is a limitation to this study. We would like to thank you for your understanding that we have included the following as a Limitation. “The main limitations of this study are that it was a small-scale study from a single institution with no control group and a wide range of surgical procedure. The method used to detect GER was also not a standard procedure. Further analysis of structural changes and manometric measurements may establish the causal relationship between lung resection and GERD.”

**Change in the text:** p 9 Line 17 - p 10 Line3

**Comment 2:** Inadequate assessment of silent aspiration: There is a lack of clear methodology for evaluating the occurrence of silent aspiration. It appears that evaluation is based solely on the pepsin test, considering its invasiveness. However, the reliability of the pepsin test as a diagnostic tool is not sufficiently addressed in this paper.

**Reply 2:** Thank you very much for your remarks. As you know, Peptest® is a simple test that has been used in recent years to aid in the diagnosis for GERD. As you pointed out, we have not described the reliability of the test, so we have added the following additional information, citing the literature. “The Peptest® is a device containing human monoclonal antibodies that detect and capture pepsin protein, with a lower limit of detection of 16 ng/mL and an upper limit of 500 ng/mL. In a study of 285 subjects who underwent 24-hour multichannel intraluminal

impedance/pH monitoring and upper gastrointestinal endoscopy, salivary pepsin detection with the Peptest® had a sensitivity of 73% and specificity of 88.3% for diagnosing GERD at a cut-off value of 76 ng/mL (15). Another recent study placed the cut-off value at 31.4ng/mL with a sensitivity of 86.7% and specificity of 66.0% (16). Although the diagnostic value of the Peptest® still remains controversial among authors (17), it is easy to perform and noninvasive, and it may be a useful device to screen patients at high risk of silent aspiration.”

**Change in the text:** p 8 Line 3 - 5

**Comment 3:** Lack of clarification of causality: There is insufficient detailed analysis to establish causality regarding the occurrence of GER induced by surgery, potentially neglecting other factors such as patients' baseline health status or the type of surgery. While it is understandable that GER may be a concern post-lung transplantation, it is unclear if it becomes a significant issue after general lung resection and the underlying mechanisms are not elucidated.

**Reply 3:** Thank you for your valuable comment. As the reviewer pointed out, it is unclear whether GERD after lung resection has a significant impact. However, some of the literature reports that structural changes are associated with GERD and a possible association with prolonged postoperative cough. In light of these, we have added the following sentence at the beginning of the Discussion, and have included additional sentences regarding future research to clarify the relationship between lung resection and GERD.” Studies concerning structural changes following lung resection have reported mediastinal shift and elevation of the diaphragm according to the resected lobe (8) and new hiatal hernias, especially after lobectomy (9). Hiatal hernia is associated with GERD and is also reported to be a risk factor for postoperative complications in patients undergoing lobectomy (10).” “Further analysis of structural changes and manometric measurements may establish the causal relationship between lung resection and GERD.”

**Change in the text:** p 6 Line 5 – 9, p 10 Line 1 - 3.

**Comment 4:** Insufficient data analysis: The study lacks detailed analysis to establish a causal relationship between lung resection and GERD occurrence, potentially overlooking confounding variables such as patients' baseline health status or surgical techniques used.

**Reply 4:** Thank you for your valuable comment. We accept that this is similar to your Comment 1. We appreciate your understanding that this is also a small, one-arm, prospective study from a single-institute, and we have described it as a limitation. “The main limitations of this study are that it was a small-scale study from a single institution with no control group and a wide range of surgical procedure. The method used to detect GER was also not a standard procedure. Further analysis of structural changes and manometric measurements may establish the causal relationship between lung resection and GERD.”

**Change in the text:** p 9 Line 17 - p 10 Line3

## Reviewer C

**Comment 1:** This is an interesting and relevant endeavor in order to seek out those thoracic patients at high risk of aspiration so that this may be managed peri-operatively.

**Reply 1:** Thank you for your input. We are hopeful that if we can detect potential aspiration patients at an early stage, we can reduce postoperative complications.

**Comment 2:** How did you calculate the number of subsegments resected with there being such variability in subsegments and also the inclusions of wedge resections?

**Reply 2:** Thanks for your comment. The number of resected subsegments was calculated based on there being 42 subsegments in the lung. For example, right upper lobectomy results in the loss of 6 subsegments, and right middle lobectomy results in the loss of 4 subsegments. A wedge resection will lose less than one segment, which means less than two or three subsegments.

**Comment 3:** I believe that the rationale for the cause of aspiration in these patients (higher intrathoracic pressure gradients) is reasonable and this would increase with increased volumes of lung resected and those with pre-existing COPD.

**Reply 3:** Thank you for your valuable opinion. We agree with the reviewer. We think that it is important to be very careful regarding lung volume reduction and COPD patients. And, we consider that further clinical studies are needed to prove that.