Peer review file
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Reviewer A

General reply: We thank reviewer A for carefully reviewing our manuscript. We revised several points according to the reviewer's comments, as described point-by-point below. Revisions are highlighted in yellow in the revised paper.

Comment 1: In the first paragraph, there is quite a lot pathological informations. This should be simplified
Reply: We reduced the information in the first paragraph and simplified it.

Comment 2: The authors have not acknowledged any limitations of the study when there are several. Reporting results wuthout acknowledging limitations can misguide the readers. Tuthors are strongly ceccomended to add a paragraph on the limitations and the strengths if their study.
Reply: We created a new paragraph describing the limitations of our study at the end of manuscript (**page 15, line 16– page 16, line 3**).

Comment 3: Did the authors note postoperative complications. Please add information.
Reply: We investigated postoperative complications according to the expression of PDH. There was no significant correlation between the presence of postoperative complications and survival. We included text in this issue in the discussion (**Table 2, Table 3**, **page 14, lines 15–17**).

Comment 4: How many patients have received an adjuvant chemiotherapy? What kind? Please add these informations and add chemiotherapy in uni and multivariate analysis.
Reply: We described adjuvant chemotherapy in the material and methods. In our institute, treatment with and the regimen of adjuvant chemotherapy was appropriately determined by the cancer board consisting of a thoracic surgeon, a radiologist, and an oncologist. Adjuvant chemotherapy did not significantly correlate with the expression of PDH. We performed univariate and multivariate analyses of the overall and disease-free survival, including information of adjuvant chemotherapy. We added information to the discussion for clarity (**page 7, lines 12–14, Tables 2 and 3, page 11 line 4 – page 12 line 11, page 14, lines 15–17**).

Reviewer B

Comment 1: The inclusion and exclusion are not so clear. Please clarify the detail of the patient disposition algorithm and the exclusion and inclusion criteria
Reply 1: We described the inclusion and exclusion criteria in the materials and methods. Patients who underwent R0 and curative treatment (more than lobectomy and mediastinal lymph node dissection) were enrolled in this study (**page 7, lines 5–10**).

Comment 2: Does all TNM stages were classified by the 8th edition? Please clarify.
Reply 2: In this study, we re-evaluated pathological information and re-staged it according to the 8th edition. We added information about the classification to the manuscript (**page 7, line 11**).

Comment 3: Does all patients with stage III were received curative-intent surgery?

Stage IIIb patients should be excluded from this study.
Reply 3: We excluded patients in pathological stage IIIB. We added information about pathological stage-related eligibility to the revised version of the manuscript (**on page 7, line 9**).

Comment 4: Please showed the detail of operation status, such as operation method, lymph node dissection status, etc. Dose all patients received anatomic resection and mediastinal lymph node dissection? Wedge resection needs to be excluded because of less N1 lymph node dissection.
Reply 4: For all patients, more than lobectomy and mediastinal lymph node dissection was performed. Patients with wedge resection were excluded. The surgical method was described in the materials and methods (**page7, lines 5–7**).

Comment 5: In this study, the study cohort seems only to include patients who received curative resection. The major survival issue is disease-free survival instead of overall survival. In addition, there were many clinical factors that may affect patients' overall survival. Please clarify the correlation between pyruvate dehydrogenase and disease-free survival.
Reply 5: We investigated the relationship between the expression of PDH and disease-free survival. The results are shown in the manuscript (**Figure 3 and Table 3, page 11, lines 4–8, page 12, lines 3–11**).

Comment 6: In subgrouping analysis, only patients who presented stage II had a statistical significant difference between pyruvate dehydrogenase positive group and pyruvate dehydrogenase negative group. However, the survival difference did not identify in stage I and III patients, please showed the subsequent analysis and discuss.
Reply 6: We added a discussion about the results of the subgrouping analysis. The results are probably due to fact that the expression of PDH indicates the potential of cancer malignancy, and the effect of surgery and PDH expression for prognosis may differ according to the pathological stage. For further analysis, it would be necessary to accumulate data including patients without surgery (**page 15, lines 3–9**).

Comment 7: Please clarify the limitations of this study.
Reply 7: We created a new paragraph highlighting the limitations of our study at the end of the manuscript (**page 15, line 16– page 16, line 3**).

Reviewer C

Submitted manuscript titled "Pyruvate Dehydrogenase represents a reliable Prognostic Predictor for Patients with Non-Small Cell Lung Cancer Resected via Curative Operation", by Ito et al investigated the prognostic role of PDH-E1α in NSCLC using immunohistochemistry. In survival analysis, PDH-E1α negative group was associated with poor prognosis. The author's study has some important findings, but the importance of the experiment and the adequacy of the method seems to be low.

Comment 1: When the PDH expression score was 4 or higher, it was considered positive. Why did you use 4 as a cutoff?
Reply 1: We determined the cutoff score of PDH expression as 4 because the number of patients being PDH positive and negative was divided into almost half when the PDH expression cutoff score was defined as 4.

Comment 2: The metabolic mechanisms between adenocarcinoma and squamous cell carcinoma are different. Therefore, it is necessary to perform survival analysis according to cell type.
Reply 2: As per the reviewer's comment, it may be better to perform the analysis according to the histological type. We performed subset analysis in patients with adenocarcinoma and with squamous cell carcinoma. However, there was no significant correlation in clinicopathological features or survival rates in each histological type (**page 10, lines 8–11, page 11, lines 9–11**).

Comment 3: In results, PDH negativity was associated with poor prognosis. Adenocarcinoma histology was also associated with poor prognosis. Therefore, PDH-negative frequency is predicted to be high in adenocarcinoma. However, the results showed that the PDH-negative frequency was lower in adenocarcinoma.
Reply 3: One of the reasons for the low frequency of PDH negative patients with adenocarcinoma might be the higher oxygen level in the tumor microenvironment in adenocarcinoma than in squamous cell carcinoma (**page 13, lines 8–14**).

Comment 4: The pyruvate dehydrogenase complex is composed of E1, E2, and E3 subunits. E1 is also composed of E1α and E1β subunits. As a result of PDH E1α alone, it is difficult to say that it is an important result, and studies on other subunits or related targets are needed.
Reply 4: In this study, the PDH E1α subunit played a key role in the PDH complex. As per the reviewer's comment, the analyses of other subunits, such as E1β, E2, and E3, might provide more detailed results, so that we would like to examine these subunits in future studies. We referred to this limitation in the revised manuscript (**page 15, line 18–page 16, line 1**).

Comment 5: Immunohistochemistry can only measure protein expression, not gene or mRNA expression. In addition to the results of immunohistochemistry, it is more reliable if there are other gene or mRNA results.
Reply 5: As per the reviewer's comment, it might be necessary to analyze the mRNA level. For patients in this study, surgery was performed more than ten years ago, so the evaluation of mRNA might be difficult. However, it is certain that analyzing mRNA would lead to more accurate results, which is why we would like to examine the mRNA level of the subunits in future. We referred to this limitation in the revised manuscript (**page 16, lines 1–3**).

Comment 6: Reproducibility is important in immunohistochemistry. Has the antibody used by the author been used in other papers?
Reply 6: The PDH E1α antibody, sc–377092, we used in this study was also used in other studies, e.g., Yang, Z et al. 2021. Oncol Lett. 21: 176. We cited this paper (**page 8, line 10**).

Comment 7: The authors investigated the expression of PDH-E1α, not the entire expression of PDH. PDH should be corrected to PDH-E1α in the paper.
Reply 7: We changed "PDH" to "PDH-E1α" in the text.