Peer Review File

Article information: https://dx.doi.org/10.21037/jtd-24-265

Reviewer A

Comment 1: However, their 'narrative review' manuscript wandered off topic in discussing of other cancers. Narrowing the review to the title is a needed revision starting point.

Reply 1: We appreciate the suggestions made by the reviewers and we will remove unrelated cancers. To focus on lung cancer, we modified corresponding paragraph and kept the most important parts relevant to lung cancer. without detriment to the review's structure. Other unrelated sentences were deleted or minimized. We condensed line 260-267 into one sentence. We also deleted sentence in line 290-292.

Changes in the text: line 260-267; line 295-297.

Comment 2: A differentiation between post-surgical tissue-informed and plasma only ctDNA assays.

Reply 2: Dear reviewers, we appreciate your suggestions for our review. After reviewing the full text, we realized the importance of this section for overall exposition, and therefore we add this differentiation to the INTRODUCTION section.

Changes in the text: line 81-88

Comment 3: Lines 236-237, The NADIM study did show baseline ctDNA was predictive of neoaduvant chemoIO RX.

Reply 3: Thank you very much for advice. Another reviewer also had questions in the same area. I carefully reread the reference and searched for subsequent studies by the same author as suggested. I reorganized the wording in that section to match the topic of the section and to be consistent with the conclusions of the study. The ctDNA can predict the prognosis but the appendix data showed no significant difference in clinical response like CR/PR/SD/PD. When talking about the pathological response, the author did not analyze relationship between ctDNA and MPR/pCR in all patients.

We modify the sentence into “Provencio's study in 2022 analyzed ctDNA after NAT, but and found it was not a significant predictor of NAT clinical response, while pathological response was not fully discussed.” The role of prediction in baseline ctDNA was showed in two studies, which listed in next section.

Changes in the text: line 286-288; line 300.

Comment 4: The discussion of cost, if discussed, should be more specific than 'prohibitively high' with current 2024 costs of commercial plasma NGS assays.

Reply 4: Thanks to the reviewers for this pertinent suggestion. We have found large variations in

the means and price of this test through inquiries and publicly available information. To make it easier for the reader to visualize the difference in expenses, we have shown the prices by referring to the medical prescription in HIS of our hospital. We assumed that the patient's expenses would be completely self-funded due to differences in health insurance coverage. However, it is evident that NGS-based assay and ctDNA-MRD tests are not covered by national health insurance, and almost all commercial insurances do not cover the costs of these tests.

We add “For example, the recommended enhanced CT scan and related tumor marker screening two cycles after NAT costs about 1400-1600 CNY (193.6-221.3 USD), while ctDNA detection for targeted driven gene costs about 5000-6000 CNY (691.6- 829.9 USD), ctDNA-MRD detection costs about 13000-16000 CNY (1383.1-2213.0 USD) in our hospital. and its inclusion would impose.” We also referred test costs in Europe.

Changes in the text: line 410-416,

Comment 5: The summary discussion is more the future challenges than supported by narrative review findings.

Reply 5: After your reminder, we noticed a deviation from the previous section of the summary. Therefore, we extensively revised the text to eliminate redundancy in the future outlook and to summarize the narrative overview.

We rewrote this paragraph: “Neoadjuvant therapy for lung cancer provides additional treatment options for patients. It is important to evaluate its efficacy accurately. The related study included ctDNA early on due to its unique advantages. Several studies have shown that ctDNA can predict the effectiveness of neoadjuvant therapy. Positive test results or low clearance rates may be associated with a higher recurrence rate and worse prognosis. At the same time, ctDNA has the potential to assist in the development of current treatment regimens and to assess step-up or step-down therapy. Many studies have used ctDNA as a secondary or primary assessment, and with the completion of appropriate studies, we will be able to explore the value of ctDNA more fully. Meanwhile, researchers believe that combining ctDNA testing with tissue samples is more appropriate for today's neoadjuvant treatment paradigm. Although value of ctDNA in lung cancer is recognized by clinicians, and it is the most effective biomarker in liquid biopsy, several issuesits accuracy, standardization and costs remain to be solved for its wider application in neoadjuvant therapy, its accuracy, standardization and costs remain to be solved for its wider application in neoadjuvant therapy. To better evaluate the validity and cost-effectiveness of this test, large-scale, multicenter clinical trials are necessary, including additional testing and subsequent analysis.” Thank you for generous comments.

Changes in the text: Line 427-455.

Comment 6: A more focused 'narrative review' is needed.

Reply 6: Thank you very much for your advice on the general structure of the article. We have cut

down unnecessary text to make this paper more like a narrative review. Thank you for generous comments.

Changes in the text: Line 166-169 Line 172-175 Line 200-203

Reviewer B

Comment 1: lines 236-237: "Provencio's study analyzed circulating tumor DNA (ctDNA), but found it was not a significant predictor of neoadjuvant therapy efficacy, however (69)" -> the authors could differentiate between MRD assays (with a sensitivity below 0.1% VAF), which are more suitable for the neoadjuvant/perioperative setting, and conventional NGS-based ctDNA assays (such as the one used in ref. 69 = NADIM trials and also the one used in the follow-up randomized study NADIM-2 by the same investigators, https://www.nejm.org/doi/full/10.1056/NEJMoa2215530), which are less sensitive and therefore less useful. A discussion of these issues is also given in a recent review, which could also be referenced here: https://pubmed.ncbi.nlm.nih.gov/38372058/

Reply 1: Dear reviewers, we are honored to receive your comments. After reading in detail the 2022 published study of NADIM-II and the 2023 follow-up study, we realized that ctDNA-based MRD assay has a great advantage, and although both technological routes (NGS assay vs. MRD assay) have value in recurrence prediction, it is clear that ctDNA-based MRD is more effective in the assessment of efficacy, recurrence and indication of escalation therapy. We have also read in detail the review that you provided to us for reference, especially its section on the comparison of the two and the listing of the corresponding clinical studies. We found it to be of great value to our review, and citing it.

We rewrote this sentence into" While no similar researches for NSCLC patients receiving NAT have been published yet. Reck's research did show clearance of ctDNA was necessary for pCR(100% negative predictive value), but not sufficient(40.5% positive predictive value). Conventional NGS-based assay can partially reflect the trend but have unsatisfactory accuracy. While the ctDNA- based MRD detection showed unique advantages like high sensitivity, which may be more suitable for NAT assessment."

Changes in the text: line 260-273; line 384-392

Comment 2: The text could be shortened at some passages, especially where other tumor types than lung cancer are addressed.

Reply 2: Thank you for your comments on the length focus, I removed some unnecessary paragraphs and sentences and confined the synthesis to the topic to reduce irrelevant content.

Changes in the text: In the full text of the review, according to the paragraphs, each of them is

partially adjusted and modified