Phase angle as a prognostic biomarker in metastatic colorectal cancer - a prospective trial

Kekez D, et al.

RESPONSE TO REVIEWER

We wish to thank the reviewer for the effort made in reviewing the manuscript. The manuscript was modified to comply with the comments.

The manuscript is generally well-structured, and the research appears to be methodologically sound. The study provides valuable insights into the role of PA as an objective pre-chemotherapy prognostic factor in mCRC. The findings that a high PA (≥4.60°) is associated with longer median progression-free survival are novel and could have significant implications for patient stratification and treatment planning.

1. The prospective observational design strengthens the validity of the findings. However, the single-institution design may limit the generalizability of the results. It would be beneficial if the authors could discuss this limitation and its potential impact on the findings.

Thank you for your comment.

Single-center studies can have limitations, such as potential biases due to including patients from a specific geographic area, which may not represent the broader

population. Additionally, treatment practices and resource availability can vary between institutions, potentially influencing study outcomes. Our study mitigated these concerns by including patients from across the country and adhering to international treatment guidelines for colorectal cancer.

The discussion was modified accordingly.

2. The use of ROC curve analysis to determine the PA cut-off value is appropriate. Nevertheless, the manuscript would be strengthened by additional sensitivity analyses to evaluate the robustness of this cut-off value.

The sensitivity is approximately 75% when considering the negative predictive value, indicating the efficacy of the phase angle in predicting rapid progressors (patients who do not respond to first-line chemotherapy).

However, the study design lacks sufficient statistical power to accurately determine the phase angle cut-off value for predicting favorable or unfavorable treatment outcomes. To achieve this, separate analyses for each patient subgroup, such as males and females and younger and older populations, would be necessary.

The phase angle value obtained through ROC analysis was utilized to demonstrate that the phase angle can serve as a prognostic indicator for survival in a prospective study involving a population of patients with the same diagnosis at the same disease stage.

The points above have been clarified and emphasized further.

3. The results indicate a significant difference in PFS between the high and low PA groups, but no difference in objective response rate (ORR). The authors should discuss potential reasons for this discrepancy and its clinical implications.

This discrepancy can stem from several potential causes. If we hypothesize that PA could indicate better organ physiology — such as improved organ function, enhanced nutrition, or reduced inflammation — this may affect the ability to maintain disease control rather than reflecting the tumor's inherent sensitivity to treatment. As a result, it could lead to a longer PFS compared to the ORR.

A higher PA might be linked to characteristics such as better vascularization, lower levels of hypoxia, or lower systemic inflammation. These factors can contribute to slower disease progression but may not necessarily lead to a better tumor shrinkage response. Similarly, patients with higher PA might better tolerate treatment due to their overall health, enabling them to sustain longer disease control. This could improve PFS without necessarily enhancing ORR.

Included in the manuscript.

4. The discussion is comprehensive, but it could be enhanced by comparing the findings with other studies that have used different methodologies or populations. Additionally, the authors might consider discussing the potential mechanisms underlying the association between PA and survival outcomes.

Thank you for your comment. We have included additional information and discussion.

Several studies have also demonstrated that low PA, is a significant predictor of adverse outcomes in patients with various chronic diseases. In a study of cirrhotic patients, a PA cutoff of ≤4.9° was independently associated with increased mortality. Patients with this low PA exhibited poorer metabolic health, worse nutritional status, and more rapid disease progression. In another study of COPD patients, low PA was significantly correlated with sarcopenia and malnutrition. Cutoff values of 4.75° and 4.25° for PA were identified as predictors of sarcopenia and malnutrition, respectively. A retrospective study of adult HIV patients found that low PA was an independent predictor of malnutrition. Cutoff values of 5.45° for men and 4.95° for women demonstrated high sensitivity and specificity in predicting malnutrition.

Also, PA reflects the integrity and function of cell membranes. Cell membranes exhibit high organization and fluidity in healthy individuals, allowing for efficient ion exchange and cellular communication. In cancer patients, particularly those with advanced disease, cell membrane integrity may be compromised due to factors such as inflammation, oxidative stress, and malnutrition. This disruption can lead to altered electrical properties of tissues, resulting in a lower PA. Cancer cells often exhibit altered metabolic pathways, including increased glycolysis and decreased oxidative phosphorylation. These metabolic changes can affect the electrical properties of cells and contribute to lower PhA values. Chronic inflammation and oxidative stress are common in cancer patients and can contribute to disease progression and poor outcomes. These processes can damage cell membranes, disrupt cellular metabolism, and impair immune function. Studies have shown an association between low PhA and elevated levels of inflammatory markers, suggesting that inflammation may play a role in the relationship between PhA and survival.

5. Figure 2: The Kaplan-Meier curves are informative, but the number of patients at risk for each time point should be included in the figure or the figure caption for better clarity.

The number of at-risk patients is included in the figure and indicated at each five-month interval.

6. No specific details on the randomization process and allocation concealment were provided in the manuscript. This is a very important part of prospective trials to ensure the reliability of the results and reduce bias.

This study focused on a homogenous population of patients with metastatic colorectal cancer before initiating first-line treatment. While traditional randomization was not employed, the investigated variable (PA values impacting treatment outcomes) was unknown, and previous studies in this area have been retrospective. Therefore, a key strength of our study lies in its inclusion of patients with uniformly staged disease, allowing for the control of disease characteristics and enhancing the robustness of our findings.

7. The manuscript utilized the Kaplan-Meier and Log-Rank tests, which are standard methods in survival analysis. However, the article did not mention whether corrections were made for possible confounding factors, such as age, gender, tumor stage, etc.

The baseline characteristics of the study participants are presented in Table 1. The low and high PA groups were generally well balanced, except for a higher proportion of men with PA above 4.6° compared to women. Standard variables for clinical studies on patients with mCRC were included as potential confounding factors. Univariate regression was performed for each variable: age, sex, ECOG, phase angle, resection of the primary tumor, RAS status, tumor sidedness, liver, and peritoneal metastases. Statistical significance was observed only for phase angle (HR 0.69, 95% CI 0.48-0.99, p=0.045) and primary tumor resection (HR 0.54, 95% CI 0.34-0.85, p=0.007). These findings were confirmed by multiple regression analysis. The results of multiple regression are detailed in Table 2.