

**Point-to-Point Response**

**Tuesday, July 29, 2025**

**Response to Editor**

**Response:** Dear Editor & Reviewers,

We appreciate your time and your comments which have been approved to be productive and significant in improving the manuscript. The concerns raised, have been addressed carefully, and incorporated into the main manuscript under Track Changes in MS Word. Moreover, attached is the point-to-point responses to reviewer's comments.

Thanks

## **Reviewer's comment**

### **Reviewer 1**

The authors suggested that further functional in vivo validation will strengthen the findings of this study. They commend that the authors have acknowledged that the lack of animal models limits a comprehensive understanding of immunological dynamics in the TME. Future investigations should focus to explore how HDAC10 suppression influences CD8+ T cell activation and checkpoint blockade efficacy in vivo. Similarly, investigating specific immune cell subsets such as T-regulatory cells or myeloid-derived suppressor cells could clarify its impact on the TME. The suggestion is reasonable, with that the reader could better understand the HDAC10 functions in the TME. Secondly, the authors suggested to investigate the direct pharmacological inhibition of HDAC10 using selective inhibitors such as DKFZ-748 and subsequent biological response assessments would elucidate role of HDAC10 in chemoresistance and strengthen the therapeutic rationale for its selective targeting in CRC. because of the transcriptional and immune-modulatory effects compared to pan-HDAC inhibition. Therefore, it's reasonable that validating the oncogenic function of HDAC10 and evaluate its potential as a therapeutic target in vivo studies, which would provide critical mechanistic insights into its role in tumor progression and therapy resistance. Thirdly, the authors suggested to investigate the HDAC10-p53-PD-L1 axis, which could clarify how HDAC10 modulates both tumor proliferation and immune escape, particularly through transcriptional and epigenetic regulation of PDL1. Since HDAC-mediated regulation of PD-L1 via p53 and STAT1/IRF1 has been described in multiple cancer types which suggests a broader transcriptional control mechanism beyond direct promoter binding. Therefore, assessing whether REG1A and CLDN18 expression correlates with specific CRC subtypes in clinical samples would strengthen the link between HDAC10-driven transcriptional changes and tumor heterogeneity. In conclusion, this letter to editor is comprehensive and reasonable, as the authors said the future studies will build upon this work by incorporating in vivo models, targeted HDAC10 inhibition as suggested, and deeper mechanistic insights into how HDAC10 regulates tumor immunity.

**Response:** Thank you for your thoughtful and positive comments. We appreciate your evaluation and valuable suggestions for future investigations.

## Reviewer 2

**Comment 1.** The letter could briefly comment on whether HDAC10-selective inhibitors might have clinical advantages in MSS or MSI-H CRC subtypes which can enhance the practical clinical relevance of the commentary.

**Response:** Thank you for the suggestion. We have briefly discussed the relevance of HDAC10-selective inhibitors in MSS and MSI-H CRC subtypes to enhance the clinical applicability of the commentary.

“Moreover, Colorectal cancers (CRC) are further classified into microsatellite instability-high (MSI-H) and microsatellite stable (MSS) subtypes based on DNA mismatch repair status [1]. MSI-H tumor responds well than MSS inhibitors to immune checkpoint inhibitors due to high neoantigen load [2]. The distinct immune landscapes of MSS and MSI-H colorectal cancer subtypes would be worthwhile to explore whether HDAC10 inhibition offers great therapeutic advantages in MSS tumors.”

**Comment 2.** While Figure 1 is helpful, the inclusion of question marks as placeholders (e.g., for Treg activation or MDSCs) might benefit from clearer labelling such as “hypothetical role” or “requires validation.”

**Response:** The figure legend has been revised to clarify the question marks indication “Red question marks indicate hypothetical mechanisms that remain to experimentally validate.”

**Comment 3.** The authors may consider briefly elaborating on the potential advantages of isoform-selective inhibitors such as DKFZ-748, particularly in terms of improved specificity and reduced off-target toxicity compared to pan-HDAC inhibitors.

**Response:** We have elaborated on the therapeutic advantages of isoform-selective inhibitors like DKFZ-748 in comparison to pan-HDAC inhibitors.

“Isoform-selective such as DKFZ-748 offers significant therapeutic advantages over pan-HDAC inhibitors. They provide higher therapeutic precision to minimize off-target effects and systemic toxicity associated with pan-HDAC

inhibitors [3]. This target approach may improve clinical applicability and enhance patient safety.”

## References

1. Wang, H., et al., *Subtyping of microsatellite stability colorectal cancer reveals guanylate binding protein 2 (GBP2) as a potential immunotherapeutic target.* J Immunother Cancer, 2022. **10**(4) [PMID: 35383115 DOI: 10.1136/jitc-2021-004302].
2. Deng, Z., et al., *Pathological response following neoadjuvant immunotherapy and imaging characteristics in dMMR/MSI-H locally advanced colorectal cancer.* Frontiers in Immunology, 2024. **15**: p. 1466497 [PMID: 39399495 DOI: 10.3389/fimmu.2024.1466497].
3. Stewart, T.M., et al., *Histone deacetylase-10 liberates spermidine to support polyamine homeostasis and tumor cell growth.* Journal of Biological Chemistry, 2022. **298**(10) [PMID: 35988653 DOI: 10.1016/j.jbc.2022.102407].