Utilizing Drug Intelligence Science (DIS®) for tumor type selection and molecular characterization of HFB200603, a best-in-class B and T Lymphocyte Attenuator (BTLA) monoclonal antagonist

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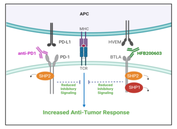
BACKGROUND

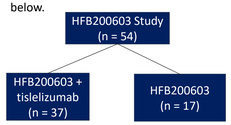
- HiFiBiO has developed an innovative approach to drug development through its Drug Intelligence Science (DIS®) platform, which leverages the power of multi-modal artificial intelligence (AI). By integrating both predictive and generative models, the DIS® platform is designed to:
  - Precisely match subjects with the most suitable therapies
  - Accelerate the drug development timeline
  - Improve the probability of success in clinical trials
- The DIS® platform has been applied to select tumor types most likely to respond to HFB200603, a best-in-class antagonistic monoclonal antibody targeting BTLA as monotherapy and in combination with tislelizumab (anti-PD-1) in an ongoing Phase 1 clinical trial (NCT05789069).
  - BTLA is a co-inhibitory immune checkpoint molecule primarily expressed on B cells, T cells, and dendritic cells.
  - Dual blockade of BTLA and PD-1 is expected to result in stronger activation of T effector cells.
- Here, we report progress on applying the DIS® platform to infer predictive signatures of clinical benefit, supporting indication and subject selection in ongoing and upcoming trials.



HFB200603 PHASE 1 TRIAL

Study Design & Tumor Sample Analysis

- This trial is evaluating HFB200603, a best-in-class BTLA antagonist, in 54 subjects with advanced, treatment-resistant solid tumors: 17 subjects received monotherapy HFB200603, and 37 subjects were treated with HFB200603 in combination with tislelizumab (anti-PD-1) every three weeks (Q3W).
- Bulk RNA-seq and multiplex immunofluorescence (mIF) data was generated from tumor samples collected at screening (baseline) and C2D8. Subject counts for each modality of tumor data are shown below.

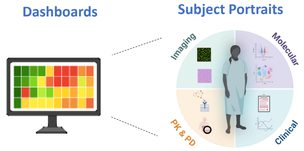


Indication	N subjects	Tumor bulk RNA-seq	Tumor mIF
ccRCC	10	1	1
Gastric	10	5	6
Melanoma	5	0	0
CRC	20	8	8
NSCLC	9	2	3

AI-READY CLINICAL DATABASE AND BIOMARKER DISCOVERY

Dashboards for real-time clinical monitoring and subject portrait generation

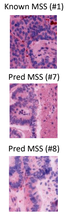
- **Real-time Dashboards:** Accelerate the transition from data collection to actionable insights, facilitating clinical monitoring and expediting biomarker discovery.
- **Deep Learning:** Generates precise subject features from HiFiBiO dashboards, feeding predictive AI models to improve decision-making and clinical outcomes.
- **Subject Portraits:** Integrate multi-modal biomarker and clinical data to create comprehensive profiles, enabling more personalized therapeutic approaches.



- **Augmented Subject Portraits:** Can be generated by integrating data from public databases. To illustrate this, we assigned MSI/MSS status to CRC tumors using deep learning foundation models trained on Hematoxylin and Eosin (H&E) images from CRC patients in public databases. These predictions were independently verified by an expert pathologist.

MSI/MSS status assignment using digital pathology

Patient	Medical Record	Digital Pathology Prediction	Prediction Probabilities
Patient #1	MSS	MSS	(93 – 100%)
Patient #2	MSS	MSS	(91 – 100%)
Patient #3	MSS	MSS	(99 – 100%)
Patient #4	NA	MSS	(87 – 100%)
Patient #5	NA	MSS	(93 – 100%)
Patient #6	NA	MSS	(99 – 100%)
Patient #7	NA	MSS	(76 – 100%)
Patient #8	NA	MSS	(97 – 100%)



Representative example images and predictions

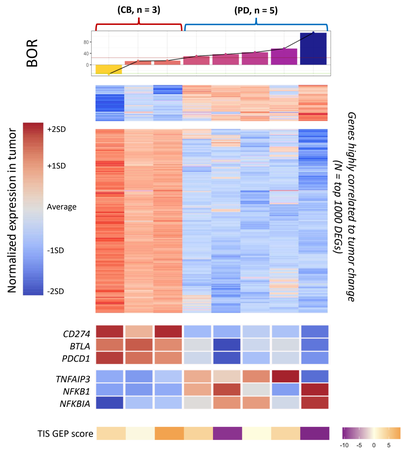
DIS® INTEGRATES GENOMIC, PHENOTYPIC AND CLINICAL DATA TO DISCOVER PREDICTORS OF CLINICAL BENEFIT

Discovery of a Potentially Predictive Tumor Signature for MSS/MSI-low CRC subjects

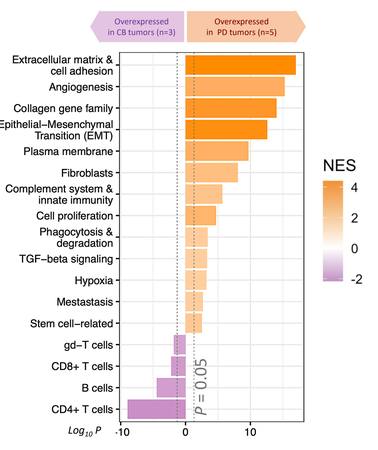
- CRC subjects (N=8) progressed on 4-5 prior lines of therapy, had liver metastases, and were confirmed to be MSS/MSI-low using deep learning digital pathology.
- Bulk mRNA-seq of baseline tumor biopsies identified an expression profile associated with **changes in tumor size (BOR)**. This profile was characterized by:
  i) Overexpression of lymphocyte activity related genes, including *BTLA*, *CD274* (PD-L1), and *PDCD1* (PD1), alongside lower expression of inflammation markers *NFKB1* (NF-kB), *NFKBIA* (IkBa), and *TNFAIP3*, which were associated with **clinical benefit (CB).**
  ii) Overexpression of poor prognosis related genes (e.g., TGF-beta signaling, EMT) was associated with **progressive disease (PD).**

  Notably, tumor inflammation signature (TIS) GEP score, predictive of responses to anti-PD-1, does not correlate with clinical benefit

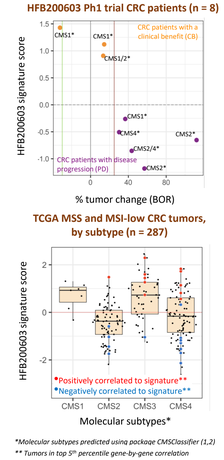
Tumor expression signatures in CRC patients (baseline, N = 8)



Gene Set Enrichment Analysis (GSEA)



Clinical benefit signature application to identify likely responders in a public dataset



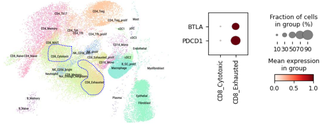
**Molecular subtypes predicted using package CMSclassifier (1,2)*
*** Tumors in top 5th percentile gene-by-gene correlation*

HFB200603 DEMONSTRATES PROOF OF MECHANISM IN TUMOR TYPES PRIORITIZED USING DIS®

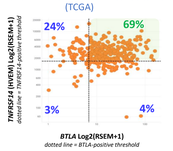
DIS® identifies Phase I indications for predicted MoA

- *BTLA* and *PDCD1* (PD-1) are co-expressed in exhausted CD8+ T cells in anti-PD-(L)1-refractory tumors
- *BTLA* and its ligand *TNFRSF14* (HVEM) are found co-expressed in different tumor types in public databases, specifically clear cell renal carcinoma (ccRCC), melanoma, colorectal (CRC), gastric, and non small cell lung (NSCLC) cancers. These tumors generally express PD-L1.

BTLA and PD-1 expression by CD8 T cells in tumors refractory to IO (HiFiBiO Disease Cell Atlas)

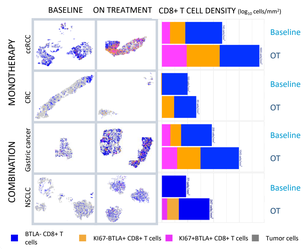


BTLA and HVEM expression in ccRCC tumors (TCGA)

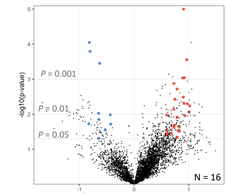


HFB200603 promotes on-mechanism proliferation of intratumoral BTLA+ T cells

Intratumoral CD8+ T cell density in digitally reconstructed tumor mIF slides



Volcano plot for on-treatment vs. baseline comparison of mIF cell density features



In red: Ki67+BTLA+ T cell : Ki67-BTLA- cell ratios
In blue: Ki67-BTLA- cell : Ki67+BTLA+ T cell ratios

SUMMARY & FUTURE DIRECTIONS

- HiFiBiO's DIS® platform is being used to prioritize indications and subjects in ongoing Phase 1 clinical trials. This approach integrate AI and predictive modeling with real-time data from the ongoing Phase 1 trial to enrich clinical enrollment for the tumor and subject populations most likely to respond to HFB200603.
- HFB200603 treatment, both as a monotherapy and in combination with tislelizumab, results in expansion of intratumoral BTLA+ CD8+ T cells.
- Powered by integration of tumor biopsy and near real-time clinical data, the DIS® platform enabled the generation of a preliminary clinical benefit prediction signature for HFB200603 based on RNA expression Phase 1 data from metastatic MSS/MSI-L CRC tumors.
- These results underscore HiFiBiO's commitment to accelerate development timelines and improving the probability of success by prioritizing subject selection for the most suitable therapies.

Acknowledgments and references

We extend our thanks to the patients, their families, and the investigators and staff members who made this trial possible.

1. Guinney et al. 2015. The consensus molecular subtypes of colorectal cancer. Nat Med 21:1350-6 (10.1038/nm.3967)
2. *https://github.com/Sage-Bionetworks/CMSclassifier*

For additional information, please email contact@hifibio.com or visit HiFiBiO.com