Reconfigurable Separation (MARS®) system – combines novel acoustic cell processing and in-flow immuno-magnetic separation technologies with automation of the entire purification workflow for downstream cellular growth, modification, and analysis prior to being administered to patients.

Methods As a fully automated system, the MARS® system can isolate T-cells with high purity from lysed whole blood in as little as 11 minutes with up to 98% recovery and 98% viability without the need for Ficoll or centrifugation. The process is scalable to 10ML of blood, with complete purification requiring 1 hour. The system is designed to fit into a culture hood for sterile cell handling and has been used to isolate T-cells for expansion for downstream T – cell uses.

Results The tunable microfluidic cell processor is a functional module capable of washing and concentrating various sample types including all white blood cell types from whole blood, bone marrow, and apheresis. Additional uses for thawed frozen PBMC, cultured cells and solid tumor dissociation have also been demonstrated. Comparing with conventional centrifugation process, cell preparation by MARS has demonstrated high level of debris removal (>97%), minimal cell loss (> 90% recovery) and high cell viability with full automation. MARS is the first-to-market fully automated system to integrate sample preparation and cell isolation into a single platform and is designed to be a versatile tool for downstream cell analysis workflows.

Conclusions MARS is the first to market automatic sample preparation system and is designed to be a versatile tool for downstream cell analysis platforms. The MARS® system is an ideal instrument to prepare CAR-T cells due to its ability to isolate and purify these cells from whole blood with high viability in a completely automated process.

REFERENCES

1. https://medcitynews.com/2018/10/car-t-therapies-face-logistical-bottleneck-as-market-set-to-see-major-growth/
2. https://www.statnews.com/2018/07/17/car-t-bottleneck-cell-collection-centers-feel-crunch/

http://dx.doi.org/10.1136/jitc-2020-SITC2020.0002

3 BUTYROPHILIN-3A IS EXPRESSED IN MULTIPLE SOLID TUMORS: TRANSLATIONAL RESEARCH SUPPORTING THE EVICTION STUDY WITH ICT01, AN ANTI-BTN3A MAB ACTIVATING VG9VD2 T-CELLS

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Background Butyrophillin-3A (BTN3A) three isoforms (3A1/3A2/3A3) are widely expressed on a variety of tumors.[1] BTN3A1 plays a key role in phosphoantigen activation of Vg9Vd2 T-cells, key mediators of innate and adaptive anti-tumor immunity.[2] Vg9Vd2 T-cell infiltration into tumor tissues is associated with a positive prognosis across multiple cancers,[3] which makes BTN3A an interesting target for enhancing anti-tumor immunity. ImCheck Therapeutics is developing ICT01, an anti-BTN3A mAb that specifically activates Vg9Vd2 T-cells. ICT01 is currently in an international, multi-center Phase 1/2a clinical trial (NCT04243499, EVICTION Study). The level of BTN3A expression required for a clinical response to ICT01 is not known. Therefore, we developed novel immunohistochemistry (IHC) methods to enable a precision-medicine based approach to target population selection for dose escalation and potentially guiding patient selection in the expansion cohorts of the ongoing EVICTION study.

Methods A panBTN3A IHC staining that recognizes the three isoforms was developed on Fresh frozen (FF) tissues, while BTN3A2- and BTN3A3-specific IHC methods were developed on formalin-fixed paraffin embedded (FFPE) tissues. BTN3A1-specific staining is still under development. Transfected knock-out/knock-in cell lines and positive tissues were used to assess antibody specificity. BTN3A expression was then analyzed on both normal and associated tumor tissue using tissue microarrays (TMA) and selected frozen blocks from tumor biopsies. FACS analyses were also performed on dissociated lung and pancreatic cancer biopsies to determine BTN3A (3 isoforms) membrane expression on tumor-infiltrating immune cells and cancer/stromal cells.

Results In normal tissues, BTN3A2 and BTN3A3 specific IHC signals were granular cytoplasmic in epithelial cells, with positive mononuclear and endothelial cells. Higher expression in lung, colon, and small intestine tissues was observed. Regarding panBTN3A expression, inter-indication and inter-patient heterogeneity was observed among head and neck, lung, melanoma, bladder, colon, pancreas, breast, and prostate cancer tissues, with both cytoplasmic and membranous localizations. The major finding was higher expression of BTN3A2 on malignant cells in melanoma, lung, colon, and prostate cancers, as compared to normal tissue. Finally, FACS analyses of lung and pancreatic cancer tissues revealed stronger expression of all BTN3A isoforms at the cell surface of infiltrated immune cells compared to its expression on stromal cells.

Conclusions These validated IHC methods supported the selection of cancer indications for the EVICTION trial and will potentially help identify specific tumor subtypes and patients that will most likely benefit from ICT01 treatment.

REFERENCES

1. Compte E, Pontarotti P, Collette Y, Lopez M, Olive D. Frontline: characterization of BT3 molecules belonging to the B7 family expressed on immune cells. *Eur J Immunol* 2004;**34**:2089–99.
2. Juan-Luis Blazquez, Audrey Benyamine, Christine Pasero, and Daniel Olive. New Insights into the Regulation of ?d T Cells by BTN3A and Other BTN/BTNL in Tumor Immunity. *Front Immunol* 2018;**9**:1601.
3. Andrew J. Gentles, Aaron M. Newman, Chih Long Liu, Scott V. Bratman, Weiguo Feng, Dongkyoon Kim, Viswam S. Nair, Yue Xu, Amanda Khuong, Chuong D. Hoang, Maximilian Diehn, Robert B. West, Sylvia K. Plevritis, Ash A. Alizadeh. The prognostic landscape of genes and infiltrating immune cells across human cancers. *Nat Med* 2015 Aug; **21**(8):938–945.

http://dx.doi.org/10.1136/jitc-2020-SITC2020.0003

4 MOLECULARLY GUIDED MULTIPLEXED DIGITAL SPATIAL ANALYSIS REVEALS DIFFERENTIAL GENE EXPRESSION PROFILES IN THE WNT-β-CATENIN PATHWAY BETWEEN MELANOMA AND PROSTATE TUMORS

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Background The canonical WNT-β-catenin signaling pathway is vital for development and tissue homeostasis but becomes strongly tumorigenic when dysregulated. and alter the transcriptional signature of a cell to promote malignant transformation. However, thorough characterization of these transcriptomic signatures has been challenging because traditional methods lack either spatial information, multiplexing, or sensitivity/specificity. To overcome these challenges, we

J Immunother Cancer: first published as 10.1136/jitc-2020-SITC2020.0003 on 9 November 2020. Downloaded from http://jitc.bmj.com/ on February 26, 2024 by guest. Protected by copyright.