MAR 21, 2024

DOI:

Protocol Citation: Liz McDonough 2024. HuBMAP | GE HealthCare/UPitt Cell DIVE™ and scRNA Modality Overview. **protocols.io**

Protocol status: Working
We use this protocol and it's working

Created: Mar 21, 2024

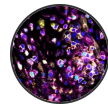# 🌐 HuBMAP | GE HealthCare/UPitt Cell DIVE™ and scRNA Modality Overview

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ABSTRACT

This is an overview of all protocols currently in use for the 2024 GE HealthCare/UPitt collaboration for the Human BioMolecular Atlas Program (HuBMAP). It includes links to each of the individual protocols that make up this project workflow.

Obtaining Donors and Tissue Blocks

1 Confirm donor acceptance criteria for inclusion.

Donor Acceptance Criteria for GE/UPitt HuBMAP Inclusion

2 5 mm skin biopsies obtained from donor subjects are divided into two parts, with one half for formalin fixed and paraffin embedded (FFPE) processing and the other half for skin digestion for further scRNA analysis.

scRNA Analysis

3 Perform skin digestion and scRNA analysis.

HuBMAP | Digestion and scRNA Analysis of Skin

Cell DIVE Multiplexed Immunofluorescence (MxIF)

4 Prepare formalin fixed and paraffin embedded (FFPE) blocks and tissue sections.

HuBMAP | Formalin Fixation and Paraffin Embedding of Tissue Samples
HuBMAP | Sectioning of FFPE Specimens

5 Deparaffinize and rehydrate slides.

Cell DIVE™ Platform | Slide Clearing and Antigen Retrieval

6 Characterize antibodies (primary/secondary, direct conjugates, and zenon labelled antibodies) and determine any antigen effects from the Cell DIVE dye inactivation process.

Cell DIVE™ Platform | Antibody Characterization for Multiplexing
Cell DIVE™ Platform | Antibody Staining & Imaging

7 Prepare direct conjugates for study.

Cell DIVE™ Platform | Antibody Purification Chemistry
Cell DIVE™ Platform | Ab Conjugation: Initial Conjugation & Scale up Conjugation

8 Perform Cell DIVE™ multiplexed data acquisition on the final cohort.

> **Note**
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> Staining is done using the Leica Bond MAX and images are acquired on the IN Cell Analyzer 2200.