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HuBMAP | Digestion and scRNA Analysis of Skin

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We use this protocol and it's working

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Abstract

This method details digestion and scRNA analysis of the HuBMAP tissue skin specimens.



- 1 The fresh skin sample is digested enzymatically using **Miltenyi Biotec Whole Skin Dissociation Kit, human** for 2 hours and further dispersed using the Miltenyi gentleMACS Octo Dissociator.
- 2 The resulting cell suspensions are filtered through 70 μ m cell strainers twice and resuspended in phosphate-buffered saline (PBS) containing 0.04% BSA.
- 3 Resulting cell suspensions are then loaded into 10 $\times$ Genomics Chromium instrument (Pleasanton, CA) for library preparation. V1 and V2 single-cell chemistries were used per manufacturer's protocol.
- 4 The resultant libraries are sequenced (~200 million reads/sample), using the Illumina NextSeq-500 platform.
- 5 The output sequencing reads were then examined by quality metrics, transcripts mapped to reference human genome (GRCh38) and assigned to individual cells according to cell barcodes, using Cell Ranger (10 $\times$ Genomics).