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Human Lung Digestion: Deriving a single cell suspension

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1 Works for me This protocol is published without a DOI.

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SUBMIT TO PLOS ONE

ABSTRACT

Human Lung Digestion: Deriving a single cell suspension

Digestion solution: (add PBS (without Ca,Mg) up to 50mL)

- 84mg Collagenase Type I (Life Tech, cat# 17100017, powder 285U/mg)

- 5mL Dispase (Fisher, cat# CB40235, 50U/mL)

- 3mL DNase1 (Roche, cat# 10104159001/165U/ml)

FACS Buffer:

- 1L dPBS (Cell Center, cat# 14190136, without Ca++ & Mg++)

- 2mL EDTA (Invitrogen, cat# 15575020, UltraPure, 0.5M, ph 8.0, 100mL)

- 10mL of 10%FBS (Invitrogen, cat# 10437-028, 500mL)

0.04% PBS+BSA (resuspending sample for sequencing)

- 10mg BSA (Jackson,cat# 001-000-162, IgG free, protease free)

- 25mL PBS (Cell Center, cat# 14190136, without Ca++ & Mg++)

MACS Accessories: (Miltenyi Biotec)

GentleMACS Octo Dissociator w/ heaters (cat# 130-096-427)

GentleMACS C Tubes (cat# 130-096-334)

MACS- Smart Strainers 70uM (4x25pk) (cat# 130-110-916)

ATTACHMENTS

Morrisey Human Lung Digestion Protocol March .pdf

PROTOCOL CITATION

Morrisey Lab 2021. Human Lung Digestion: Deriving a single cell suspension. **protocols.io** https://protocols.io/view/human-lung-digestion-deriving-a-single-cell-suspen-btntnmen

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