

LAMINA PROPRIA ISOLATION

Number of mice

0.5

Wash medium

HBSS	45 mL
Pen/Strep (100 µg/mL)	0.45 mL
FBS (1%)	0.45 mL

Epithelial separation medium

HBSS	10 mL	
Pen/Strep (100 µg/mL)	0.1 mL	
2-Mercaptoethanol	0.01 mL	
EDTA (1 mM---0.5M)	0.02 mL	
FBS (1%)	0.1 mL	25

HBSS + Pen/Strep + EDTA + FCS

HBSS	10 mL	11
Pen/Strep (100 µg/mL)	0.1 mL	
EDTA (1 mM---0.5M)	0.02 mL	
FCS (1%)	0.1 mL	

Digestion Medium

MEM α*	10 mL	0.5 mice	→	MEM media + NEAA (100X) + sodium pyruvate (100x)
Pen/Strep (100 µg/mL)	0.1 mL			
2-Mercaptoethanol (1000X)	0.01 mL			
FCS (5%)	0.5 mL			

Dnase (1000X)	6.25 µL
Collagenase V (0,85 mg/mL)*	4.25 mg
Collagenase D (1,25 mg/mL)*	6.25 mg
Dispase (1 mg/mL)* 없음	5 mg

*Minimum Essential Medium (MEM) α Medium 1X (32561-029 INVITROGEN GIBCO)

*Deoxyribonuclease I from bovine pancreas (10104159001, ROCHE) (Biochemistry, -20°C Martina)

*Collagenase from Clostridium histolyticum Type V (C9263-1G, SIGMA-ALDRICH)

*Collagenase D from Clostridium histolyticum (11 088 882 001, ROCHE)

*Dispase II (17105-041, GIBCO)

Digestion Medium	0.5 mice
MEM α*	10 mL
Pen/Strep (100 µg/mL)	0.1 mL
2-Mercaptoethanol (1000X)	0.01 mL
FCS (5%)	0.5 mL

Dnase (1000X)	6.25 µL
Collagenase V (0,85 mg/mL)*	4.25 mg
Collagenase D (1,25 mg/mL)*	6.25 mg
Dispase (1 mg/mL)* 없음	5 mg

Digestion Me	0.5 mice
MEM α*	10 mL
Pen/Strep (100 µg/mL)	0.1 mL
2-Mercaptoetha	0.01 mL
FCS (5%)	0.5 mL

Dnase (1000X)	6.25 µL
Collagenase V	4.25 mg
Collagenase D	6.25 mg
Dispase (1 mg)	5 mg