Appendix A

List of media used in this study

1. The component of PBI and M16

component	PBI	M16
	(mM)	(mM)
NaCl	136.00	94.66
KCl	15.42	4.78
CaCl ₂	0.89	1.71
KH ₂ PO ₄	1.47	1.19
MgSO ₄ .7H ₂ O		1.19
MgCl ₂ .6H ₂ O	0.44) ⁻
NaHCO ₃	-	25.00
Na ₂ HPO ₄	1.41	-
Sodium lactate	-	23.28
Sodium pyruvate	- 607	0.33
Glucose	5.56	5.56
Bovine serum albumin	0.4%	0.4%
Penicillin G	100 unit	100 unit
Streptomycin sulfate	-	38.85 unit
Phenol red		
Osmolarity (mOsm/kg)	280 ± 5	280 ± 5
pН	7.35-7.40	7.35-7.40

Appendix B

List of the chemicals and materials used in this study

Chemicals/Materials

Source

0.2m filtered unit

7x cleaning solution

Bovine serum albumin

Calcium chloride

D(+)-Glucose

Lactic acid

Magnesium chloride 6-hydrate

Magnesium sulfate 7-hydrate

Mineral oil

Penicillin G sodium salt

Phenol red

Plastic petri dish

Potassium chloride

Potassium phosphate monobasic anhydrous

Pyruvic acid

Sodium bicarbonate

Satorius, Germany

ICN Biomedical, USA

Sigma chemical Co., USA

Nunclon, Denmark

Sigma chemical Co., USA

Sigma chemical Co., USA

Sigma chemical Co., USA

Sigma chemical Co., USA

Sodium chloride

Sigma chemical Co., USA

Streptomycin sulfate

Sigma chemical Co., USA

Syringe

Nipro, Thailand

Appendix C

1. Preperation of capillary pipette

In constructing capillary pipettes, the thin portion of a Pasteur pipette was held in a gas flame and rotated for a few seconds. When the glass became soft, it was quickly removed from the flame and immediately pulls out a predetermined distance without breaking the capillary segment. The ends were held steady for a few seconds, then bent it and broken at about 2 cm from the shoulder of pipette. Examination of the pipette under the stereomicroscope was made to be certain broken cleanly to give a perfect flat tip. Aperture diameter was controlled by initial pull when the glass was soft. For 2-cell mouse embryos, an aperture of approximately 120 µm was most desirable. This was achieved with an initial pull of about 12 inches.

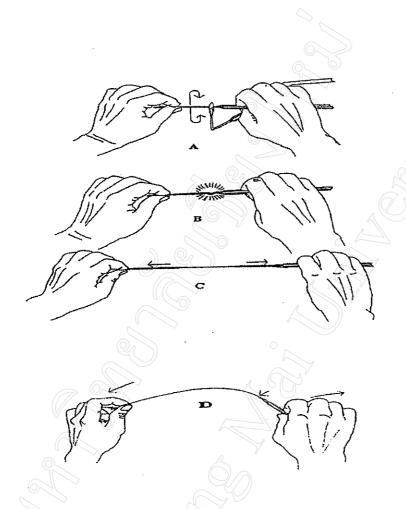


Figure 21 Procedure for drawing Pasture pipette. A Pasture pipette was rotated in a gas flame (A) Until it was softened locally and quite easy to deform. It was pulled from the flame (B) and immediately drawn out a predetermined distance (C)The ends were held steady for a few seconds, then bent the capillary segment and broken at about 2 centimeters from the shoulder of of pipette (D) (Keen and Rafferty, 1970)

Appendix D

List of instruments used in this study

Instrument

Beam balance

Dissecting microscope

Laminar flow work station

Lyophilizer

Osmometer

pH meter

CO₂Water-jacketed incubator

Source

Satorius, Germany

Olympus, Japan

Gelman Sciences, USA

Leybold-Heraeus, Germany

Gonotec, Germany

Orion, England

Nuair, USA

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