SOP: Propagation of PANC1

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Ordering Information

PANC1 can be ordered from ATCC as a frozen ampoule.

Name: PANC1, epithelioid carcinoma (pancreatic)

ATCC #: CRL-1469

Notes:

This is an adherent cell line.

Materials List

- 1. DMEM with glucose, L-glutamine and sodium pyruvate (Cellgro Cat# 10-013-CM)
- 2. Fetal Bovine Serum (Cellgro Cat# 35-016-CV)
- 3. Sodium Bicarbonate (Cellgro Cat# 25-035-CI)
- 4. T75 & T225 culture flasks
- 5. Graduated pipets (1, 5, 25mL)
- 6. Penicillin-Streptomycin Solution (100X) (Cellgro Cat# 30-002-CI)
- 7. Hemocytometer
- 8. Micropipet w/ P20 tips
- 9. Microscope

Growth Medium for PANC1

DMEM with glucose, L-glutamine and sodium pyruvate

10% FBS

Pen-Strep (1X)

Sodium Bicarbonate 1.5g/L

Bovine Insulin 0.01mg/ml

Procedure

A. Receipt of frozen cells and initiation of cultures.

- 1) Immediately place frozen cells in liquid nitrogen storage incubator.
- 2) Quickly thaw ampoule in 37°C water bath.
- 3) Transfer thawed cells to a T75 flask with 40mLs of warm growth media.
- 4) Allow cells to recover over night in 37°C, 5% CO₂ humidified incubator.
- 5) Pour off medium the next day, replace with fresh medium and return to incubator.

B. Sub-culture

- 1) Propagate cells until density reaches 70-80% confluence.
- 2) Decant medium.
- 3) Wash cells with warm 1X PBS.
- 4) Add 8mLs of Accutase and return to incubator for 10-15 minutes.
- 5) Immediately remove cells and pellet at 500 xg for 5 minutes (4°C)
- 6) Wash cells 2X with 1X PBS.

- 7) Gently re-suspend cell pellet in warm medium.
- 8) Perform 1:2 to 1:4 cell split as needed.
- 9) Record each subculture event as a passage.

C. Maintenance

1) Change media the day after seeding and every 2-3 days thereafter. Use ~50ml of medium per T225 flask.

D. Harvest

- 1) Do not use cells that have been passed more than 8 times.
- 2) Remove cells from flasks according to protocol described above under 'subculturing'.
- 3) Let cells grow 48 hours past confluence.
- 4) Examine viability using trypan blue staining (SOP).