Biosafety level: 2

Organism: Homo sapiens (human)

Tissue: kidney
Growth Properties: adherent
Morphology: epithelial

Comments: The 293T/17 cell line is a derivative of the 293T

(293tsA1609neo) cell line. 293T is a highly transfectable derivative of the 293 cell line into which the temperature sensitive gene for SV40 T-antigen was inserted. 293T cells were cloned and the clones tested with the pBND and pZAP vectors to obtain a line capable of producing high titers of infectious retrovirus, 293T/17. These cells constitutively express the simian virus 40 (SV40) large T antigen, and clone 17 was selected

specifically for its high transfectability.

Subculturing: Remove medium, and rinse with autoclaved PBS containing 0.5

mM EDTA (0.1 or 0.2- μ m-rated sterilizing-grade filtration will not usually retain mycoplasma). Remove the solution and add trypsin-EDTA solution (1 ml/10-cm dish). Incubate the culture dish at room temperature until the cells detach (for 5-10 min). Add fresh culture medium, aspirate and transfer into a tube. Centrifuge the tube at 171 x g for 3 min, remove medium and tap the bottom of the tube to dissociate the cell pellet. Add fresh culture medium and dispense the cells into a new culture dish.

Medium: DMEM (GIBCO, 11965) with MEM Sodium Pyruvate Solution

100 mM (100x) (GIBCO, 11360), Penicillin-Streptomycin (50x) (GIBCO, 15070), MEM Non-Essential Amino Acids Solution 10

mM (100x) (GIBCO, 11140) and 10% fetal bovine serum

Split Ratio: A subcultivation ratio of 1:3 to 1:30 is recommended. Subcultures

at a 1:20 split ratio achieved confluency within another 3 days. Subcultures at a 1:30 split ratio achieved confluency within

another 4 days.

Fluid Renewal: 2 to 3 times per week

Freeze Medium: Complete growth medium supplemented with 10 % (v/v)

dimethyl sulfoxide (DMSO) (1 ml of freeze medium per 10-cm dish per tube)

Note: A confluent 10-cm dish has $2-3 \times 10^7$ cells.