## **Application consideration**

Please keep the following points in mind:

## RCB1185:TT1TKB Lot. 3

• For this lot, seed the cells to one 60mm dish per tube.

The cells will take 1 day to stabilize their adhesion after thawing. (See photos #1, #2)
\*Do not change medium during this period.

This cell line grows slowly. (See photos #3, #4)
\*Keep observing the cells for 7 to 14 days after starting culture.
\*Change medium every 3 to 4 days as appropriate.

• When passaging the cells, it is difficult to disperse into single cells completely, passage them as a few to several dozen of "colony clusters". (See photos #5)

\*Photos are on the backside of this page.

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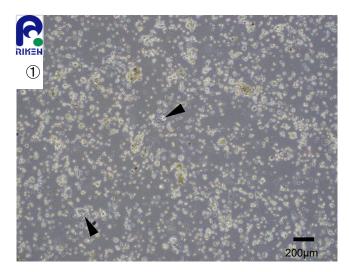


Photo #1 (x40) : Next day after thawing Almost no adherence

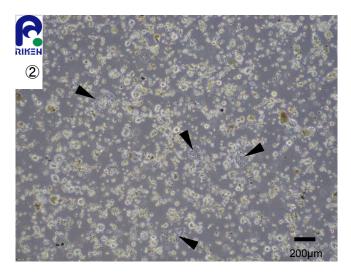


Photo #2 (x40) : Day 2 after thawing Gradual increase of adherent cells

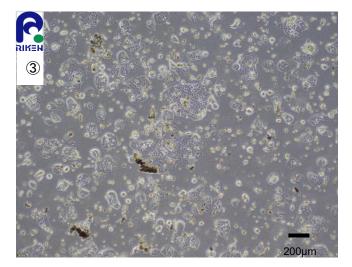


Photo #3(x40) : Day 6 after thawing Form colonies and proliferate ※Medium change at Day 4

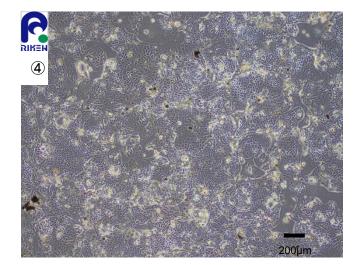


Photo #4 (x40) : Day 12 after thawing (Passaging the cells) The cells adhered to 80-90% of the entire Dish and layered in some places.

Medium change every 3 to 4 days

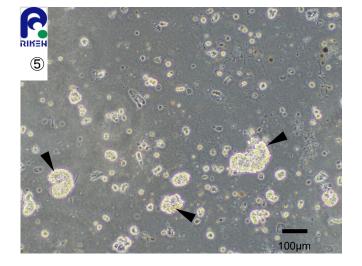


Photo #5 (x100) : Just after passaging the cells The cells are seeded in colony clusters.