

Application consideration

RCB 3522 : HAM2

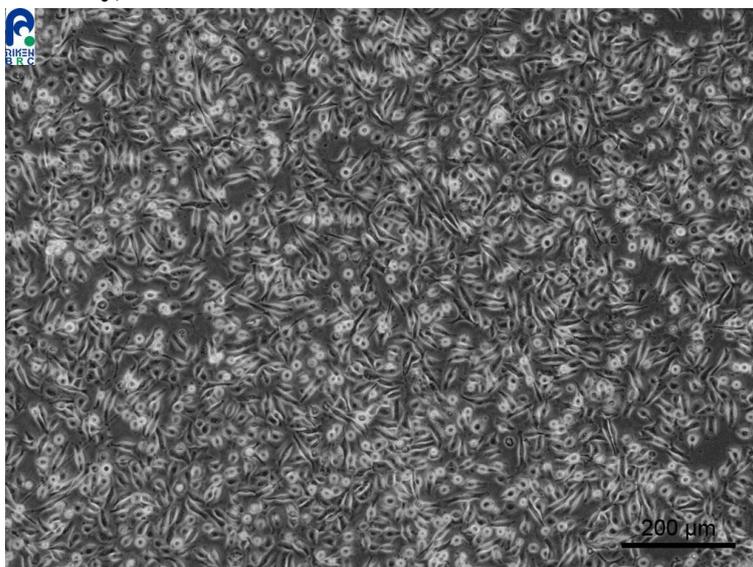
Please keep in mind following points:

Special care should be required in subculturing this cell line since it is Serum-Free cultured.

<A Method of subculturing this cell line>

1. Remove medium and wash cells once with PBS(-).
2. Add trypsin-EDTA and rotate dish gently to cover the entire cell surface, then remove it leaving small amount of trypsin-EDTA. Incubate for 2 min at 37 degrees Celsius.
3. Tap on side of dish to release cells with being careful not to splash any medium into the cover of dish. Check cells under microscope to ensure 80% of cells are detached. If cells are not sufficiently detached, incubate for an additional minute.
4. Add medium and pipette cells up and down.
5. Prepare centrifuge tube containing the equivalent of 10 volumes of medium or PBS(-), and resuspend detached cells
6. Centrifuge at 1,000rpm (about 200 x g) for 3 min at room temperature.
7. Repeat step 5 to 6 once or twice when necessary.
8. Subculture cells in 1:2 or 1:3 split ratio.
9. Check cells under microscope on the next day for attached cells.

NOTE: If you cultivate serum-free cells for the first time, please do not subculture cells of all the dishes at one time. Only after confirming the presence of some attached cells in a dish on the next day, subculture cells of the rest of dishes.



【Technical support】 e-mail : cellqa@brc.riken.jp