Application consideration

RCB 3521 : HAM1

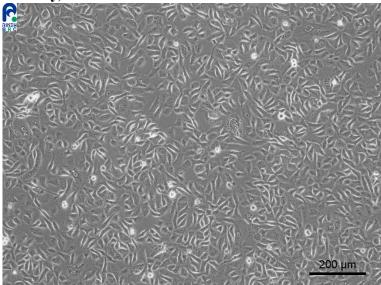
Please keep in mind following points:

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 tryspsin-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 pippet 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.



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