Appatiolicn consideration

RCB1538 : 32Dcl3

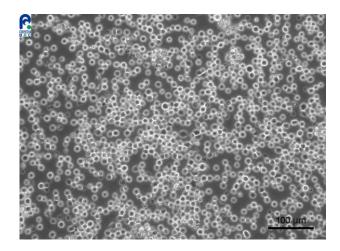
Please keep in mind following points:

This cell line needs to be added a Conditioned Medium for culturing.

- < A Method of preparing WEHI-3 (RCB0035) Conditioned Medium>
 - 1. Start cultivating WEHI-3 cells at 37 degrees Celsius and cultivate cells for 3 to 5 days after subculturing.
- 2. Collect all cell supernatant when cells reach almost confluence.
- 3. Centrifuge cell supernatant and remove dead cells and cellular debris. (Skip this step if there are few floating debris.)
- 4. Pass cell supernatant through 0.22 um syringe filter (by using low binding type).

 -If the syringe filter clogs, use 0.45 to 1.0 um filter prior to 0.22um filter.
- 5. Store it at 4 degrees Celsius. For long term storage (more than a month), keep it in -20 degrees Celsius.

NOTE: Culture filtrate (Conditioned Medium) should be stored in small aliquots to avoid repeated freeze-thaw cycles.



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