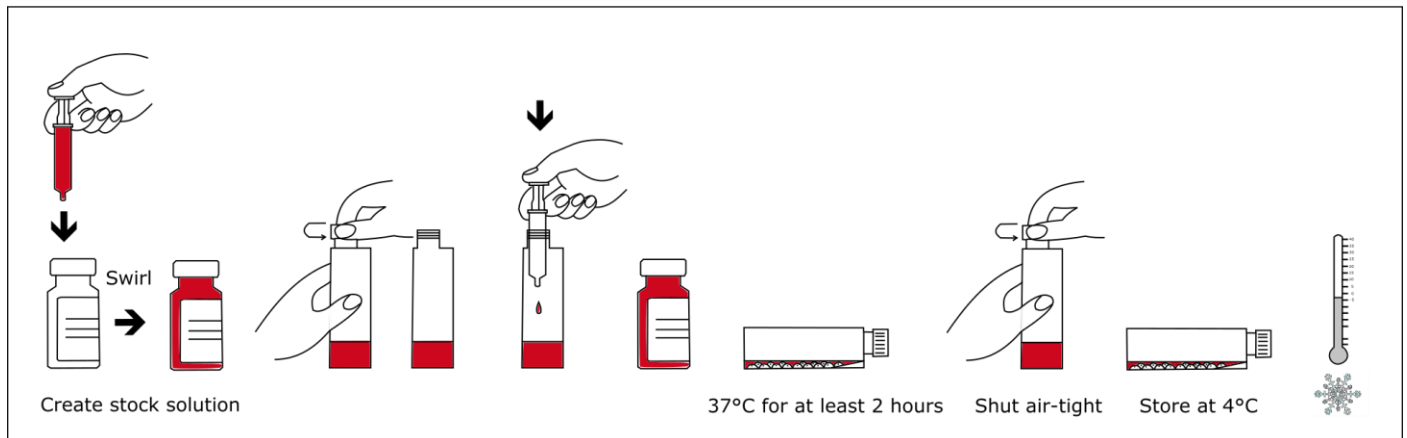


ROKEPIE®

CELL PROTECTION BY HIBERNATION

Directions for use



Directions of Use

This following protocol is an example for use in adherent mammalian cells. The cell culture medium in this example uses bicarbonate as a pH-buffer. Cells are cultured in a flask with a standard phenolic style screw cap (i.e. without a membrane) which can be closed air-tight*. This enables 5% CO₂ enriched air to remain present in the flask during storage, preventing a sharp increase in pH due to a drop in CO₂.

1. Clean the outside of the container with your disinfectant of choice.
2. Dissolve the lyophilized ROKEPIE-FD01 in 2.0ml solvent or culture medium of your choice.
3. Add ROKEPIE®-FD01 stock solution in a 1:25 ratio (confluent monolayer) or 1:50 ratio (semi confluent monolayer) to either fresh or already present cell culture medium.
4. Incubate the cells in the supplemented medium for a minimum of two hours at 37°C.
5. Close the flask air-tight and place the cells in a cold-room or refrigerator (2-8°C).
6. After cold-storage, place cells in a humidified incubator with a slightly opened cap and replace the supplemented cell culture medium once cells have re-attached**. This normally takes about 6 – 24 hours. If cells have not detached during cold-storage, the supplemented cell culture medium can be replaced immediately.

**) If no standard phenolic style caps are available, the filter can be covered by parafilm in order to seal the flask air-tight. Wells or dishes can be covered entirely with parafilm, or all around the edges of the lid.*

****) Detachment of cells can occur in some cell lines, but does not necessarily indicate they are apoptotic/necrotic. ROKEPIE®-S01 is not toxic at working concentrations and 24 hour exposure to cells at 37°C should not result in loss of viability.*

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