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ProductInformation

CLONING CYLINDERS

Product No:	C 1059	8 x 8 mm	150 Fl	15 cylinders/dish
	C 2059	10 x 10 mm	250 Fl	10 cylinders/dish

Store: Room Temperature

Application: For isolating individual colonies of transfected cells.

SUGGESTED PROTOCOL

1. Clones should be at least 100 cells before isolation.

2. Remove media from 100 mm plate and wash once with 5 ml PBS without calcium or magnesium (D 5650). Add 5 ml PBS without calcium or magnesium to each plate.

3. Using sterile, curved forceps, center the cylinder over a colony of cells. Using the flat side of the forceps, press gently but firmly on the cylinder to create a seal between the plate and the cylinder.

4. Fill the cylinder with PBS w/o Ca and Mg but do not overflow. Aspirate the solution and add the appropriate amount of dissociation solution (Trypsin, EDTA, etc.)

5. Aspirate the dissociation solution and monitor the colony under a microscope. As soon as the cells have detached, add growth media to resuspend the cells.

6. Transfer the cells to an appropriate sized plate containing growth media. It is better to transfer the cells to a small surface area until they become established, and then transfer to a larger vessel.

7. Examine the area within the cylinder to verify that the colony has been removed.

8. After all of the desired clones have been picked from the plate, remove the PBS and add fresh growth media. The remaining clones will continue to grow. The cylinders may be left on the plate or removed and discarded.

9. To measure secreted products in isolated clones:
-Remove the growth media from the plate and center the cylinder over the colony as described in step 3.
-Add media for pulse to each cylinder and examine for leaks. -Add growth media to plate and incubater for assay. Assayed clones can then be picked as described.