HARVESTER 96®
APPLICATION NOTES

CELL REMOVAL FROM MICROPLATES

A Cell Harvester works on the following principles: Room air at atmospheric pressure is sucked into a vacuum source. The speed of this air flow varies with:

a. depth of vacuum (inches Hg)

b. the restriction in the pathway

By obstructing the air flow with cells suspended in liquid, and a glass fiber filter, the cells are trapped in the filter. However, for the cells to be transported, they must be in suspension in the liquid the air is moving. This note addresses some alternative actions in those cases where the cells are adherent or otherwise not in suspension.

PUTTING CELLS IN SUSPENSION

There are several conventional methods of breaking away adherent cells and putting them in suspension. The individual operator is the best source of which method is most suitable and compatible with the protocol used.

AN ADDITION TO THE WASHING REAGENT

One report describes adding a small amount of EDTA, Ethylene Diamine Tetra Acetate, to the wash reagent. It was very successful for that application. **Caution:** Check for compatibility.

USING THE PULSE WASH PROGRAM

The programmability of the Harvester 96® allows for a pulse wash program. In this type of program, the well is filled and emptied several times. This increases the erosion effect by creating more action in the well. Harvesters shipped after January 1993, incorporate a new "Slow Pulse" program (please contact Tomtec for and upgrade if you believe this will improve your results). In the new machines, cycle # 3 can be set to repeat the same sequence a number of times. The use is to set the wash and wash/aspirate times to just fill the well, then aspirate it to empty, repeating the sequence as desired. This provides considerable action in the well to aid the removal of cells.

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USING CORNING EASY WASH PLATES

Corning provides a flat bottom tissue culture plate (Corning P / N: 25870) that has a beveled surface at the corner instead of a sharp, right angle. This plate design provides better washing action.

HAVE ALL CELLS BEEN REMOVED?

The quickest method of observing if all cells have been removed is microscopic examination. If cells are still present, run the same cycle again to determine if they can be removed by additional washing action. If so, increase the wash times (**Caution**: Check for flooding), or repeat the program, which ever is more suitable.

If a MicroBeta counter is available, you may try counting what is left behind. Harvest the plate, add scintillation cocktail and count the empty plate for residual counts. Another method is to cut up the harvested (and empty) plate and put the pieces in scintillation vials. Add cocktail and count conventionally.

TELEPHONE SUPPORT FROM TOMTEC

If the results are not as desired, the following is a check list of the variables that can be influenced. Before you call Tomtec for a solution, please make a few notes identifying the present settings.

CHECKLIST:

- Wash rate into the well: psi setting
- Wash time into the well
- Vacuum during and directly after the aspirate cycle
- Aspirate time
- Filter thickness
- Soak time
- Using pre-wet cycle or not