



## Splitting Routine/Maintenance of the cells

### Material

- Culture medium
- PBS
- Cell culture plastic ware
- Trypsin/EDTA solution (TE)
- Sterile Pasteur Pipettes

### Preparation

- Check the status of the cells microscopically
- If the cells are 80-90 % confluent, split the cells

### Splitting (see *Table 1* for corresponding volumes of Medium/PBS/TE)

- Aspirate the cultivation media of the cells with a sterile Pasteur Pipette.
- Wash the cells once with PBS.
- Aspirate the PBS.
- Add TE to the cells.
- Incubate the cells with TE at room temperature or at 37°C until the cells start to detach (check microscopically).
- Resuspend the cells with medium or PBS.
- Transfer an aliquot of the cell solution to a new cell culture dish/flask.
- Add cultivation medium to the cells.
- Cultivate the cells at 37°C.

Plastic ware	Area in cm <sup>2</sup>	Culture Medium	PBS	TE
T75	75	8-10 ml	8-10 ml	2 ml
T25	25	4-5 ml	4-5 ml	1 ml
6 well	9	1,5-2 ml	1,5-2 ml	0,5 ml
12 well	4	1 ml	1 ml	0,2 ml
24 well	2	0,5 ml	0,5 ml	0,2 ml
96 well	0,32	0,1 ml	0,1 ml	0,05 ml

*Table1: Recommended Volumes of Medium/PBS/TE*