rotated about the long axis (1 to 5 rpm). Cells adhere to the walls of the bottle and are exposed to liquid 25% of the time and to gas (e.g., 5% CO_2 and 95% air) 75% of the time. When exposed to the liquid, nutrients can be transported into the cell, and when in the gas phase, aeration takes place. The roller-bottle system has an advantage over T-flasks because of increased surface area, agitation of the liquid, and better aeration. Roller bottles are not typically used for large-scale production because of high labor requirements and bottle-to-bottle variability. However, a highly automated facility making extensive use of robots uses roller bottles for commercial production of erythropoietin (a therapeutic protein). Roller bottles are also used for production of some vaccines.

The use of microcarriers for the cultivation of anchorage-dependent mammalian cells is an attractive approach. For example, microcarriers such as DEAE-Sephadex beads, which provide large surface per unit volume of reactor (~70,000 cm^2/l), allow high cell concentrations in the medium (~10^7 cells/ml). The microcarrier beads with cells are suspended in a stirred bioreactor. Other microcarrier beads, such as polyacrylamide, polystyrene, cellulose fibers, hollow glass, and gelatin-coated dextran beads, have been developed. At present the most widely used microcarriers are dextran- and DEAE-based (DEAE-Sephadex, DEAE-polyacrylamide). The surfaces of these microcarrier beads can be modified by addition of compounds, such as collagen, to promote cell adhesion and enhance cell function. Cells grow on the surfaces of microcarriers, usually in the form of monolayers and sometimes as multilayers. Microcarrier culture methods provide a large surface area for growth and a rather homogeneous growth environment. Microcarrier beads can be placed in a gently agitated reactor vessel, in a fluidized bed, or in an airlift (bubble-column) fermenter. Bead-to-bead contact and abrasion of the surfaces can be a problem, and the nonporous nature of these carriers limits the available surface area. Agitation of large-scale vessels with microcarrier cultures is difficult, owing to balancing the needs for aeration and mixing against the shear sensitivity of cells attached to microcarriers. Shear forces are more harmful to cells attached to microcarriers than to suspended cells because attached cells cannot "tumble," and shear tends to "rip" them off the carrier surface. Macroporous microcarriers can be used; cells which enter the pores are shielded from shear effects. However, diffusional limitations and heterogeneous growth conditions may cause undesirable results. Because cells are easily retained in the bioreactor, microcarrier cultures are well suited for perfusion operation.

Conventional stirred-tank bioreactors have been modified to reduce shear rates on cells in suspension. Sail-type and axial-flow hydrofoil agitators have been developed and used for suspension cultures. The agitation rate in these reactors is on the order of 10 to 40 rpm, providing low shear rate. Many animal cells can be cultured as suspensions in bioreactors up to 10,000 l in size. Because cells are significantly smaller than the turbulent eddies in these bioreactors, cell lysis is minimized. Only cells that are at the interface of an eddy and another eddy or another surface (e.g., reactor wall) are likely to experience damage. Cells at the gas–liquid interface are particularly prone to damage. The breakage of air bubbles is particularly destructive to cells that accumulate at the interface of a gas bubble and medium.

Many shear protecting agents (such as serum or Pluronic F-68) work by preventing cells from accumulating at the gas–liquid interface. While gas exchange can be accomplished with membranes or silicone rubber tubing, sparging of gas is the simplest method and is acceptable with many cell lines when shear protectants are used. Cell lines differ in