

Another important category is the culture of hybridomas. Hybridoma cells are obtained by fusing lymphocytes (normal blood cells that make antibodies) with myeloma (cancer) cells. Lymphocytes producing antibodies grow slowly and are mortal. After fusion with myeloma cells, hybridomas become immortal, can reproduce indefinitely, and produce antibodies. Using hybridoma cells, highly specific, monoclonal (originating from one cell) antibodies can be produced against specific antigens.

To produce hybridoma cells, animals (mice) are injected (immunized) with certain antigens (see Fig. 12.3). As a reaction to antigens, the animals produce antibodies. The antibody-producing cells (e.g., spleen B lymphocytes) are separated from blood sera and fused with certain tumor cells (e.g., myeloma cells with infinite capacity to proliferate and derived from lymphocytes). The resulting cells are hybrid cells (hybridomas), which secrete highly specific antibodies (monoclonal Ab's) against the immunizing antigen.

Mammalian cell culture to produce proteins other than antibodies usually involves genetically engineered cells. Although many host cells can be used, Chinese hamster ovary (CHO) cells are particularly popular. Most of the vectors for genetic engineering are derived from viruses (usually primate). Some inducible promoters exist. However, expression levels are relatively low, often in the range of 1% to 5% of the total cellular protein.

A typical growth medium for mammalian cells contains serum (5% to 20%), inorganic salts, nitrogen sources, carbon and energy sources, vitamins, trace elements, growth factors, and buffers in water. Serum is a cell-free liquid recovered from blood. Examples are FBS (fetal bovine serum), CS (calf serum), and HS (horse serum). The exact composition of any serum product is not known. However, serum is known to contain amino acids, growth factors, vitamins, certain proteins, hormones, lipids, and minerals. A list of major

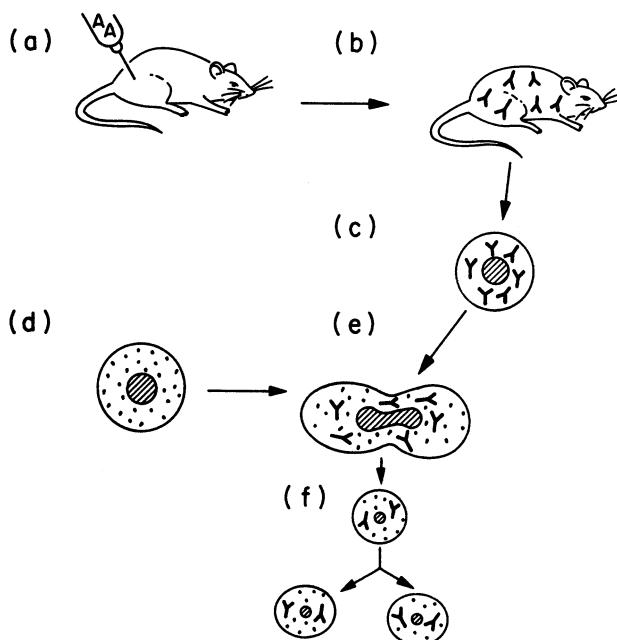


Figure 12.3. Formation of a hybridoma for making a monoclonal antibody. (a) Antigen is injected into a mouse; (b) lymphocytes in the mouse are activated to produce specific antibodies to the antigen; (c) lymphocytes are collected from the mouse; these lymphocytes grow poorly in tissue culture; (d) myeloma (cancer) cells growing in tissue culture are produced; (e) myeloma cells are fused with lymphocytes; (f) the hybrid cell grows well in tissue culture and makes a single monoclonal antibody. Progeny are called hybridomas and can be propagated indefinitely.