IMPROVED MONOCLONALS

MAKING CHIMERIC ANTIBODIES EVEN MORE HUMAN

LONDON—Researchers are anxiously awaiting clinical results of therapy using chimeric monoclonal antibodies (which contain rodent antigen-binding sites but are otherwise of human origin). Nevertheless, U.K. commercial attention is already turning toward even more extensively “humanized” antibodies that contain a bare minimum of rodent sequence. Celltech (Slough, Berkshire, U.K.) is actively pursuing this approach, while Wellcome Biotechnology (Beckenham, Kent, U.K.) has a particular interest in the recent humanization of a rat monoclonal that it already has in clinical trials.

The latest step in reshaping rodent monoclonals to minimize their recognition as foreign by the human immune system has been pioneered at the Medical Research Council's (MRC) Laboratory of Molecular Biology (Cambridge, U.K.). Instead of containing a complete rodent antigen-binding site—in the form of the variable domains of the light and heavy chains of the antibody molecule—the reshaped monoclonals contain only the three short “hypervariable” sequences of rodent variable domains (see diagram). These regions are engineered into a human variable-domain framework, which then constitutes about 80 percent of the whole variable domain.

In reality, the technology is not so much the humanization of rodent antibodies as the site-directed mutagenesis of human antibodies; oligonucleotides are used to provide the recombinant immunoglobins with the hypervariable region (or complementarity determining region) sequences of rodent antibodies.

Several recent papers from Greg Winter's MRC group have demonstrated that the functional properties of the rodent monoclonals are retained by such antibodies. In the March 25 issue of *Science* (239:1534, 1988), the team reported that when the hypervariable regions of the heavy chain of human myeloma antibody were replaced by the hypervariable regions of a mouse monoclonal against lysozyme, the resulting antibody was able to bind lysozyme. And a paper in the March 24, issue of *Nature* (332:323, 1988) describes the construction of a largely human antibody that is at least as effective as the rat CAMPATH antibody (from which its hypervariable sequences are derived). This chimeric antibody—directed against the CAMPATH-1 antigen that is expressed specifically on

human lymphocytes and monocytes—causes the complement-mediated and cell-mediated lysis of human lymphocytes.

Celltech's interest in adopting this technology currently focuses on a mouse monoclonal being developed for cancer imaging and therapy in collaboration with American Cyanamid (Wayne, NJ). At least 0.5 mg of the antibody must be given for imaging and about 5 mg for therapy. Unfortunately, a single imaging dose is sufficient to invoke a HAMA (human against mouse antibody) response, says Celltech medical director Alastair Riddell. Consequently, Celltech plans to push a chimera containing only the antigen-binding site of the mouse antibody into clinical trials later this year; the human testing of a more humanized version is anticipated by the end of 1989.

of research.

Other companies that intend to license MRC's antibody humanization technology include Unilever (Sharnbrook, Bedfordshire, U.K.), Behringwerke (Marburg, F.R.G.), Scotgen (Aberdeen, Scotland), and Wellcome. Unilever and Behringwerke will apply the technique to cancer, while Scotgen wants to treat human cytomegalovirus infection.

For Wellcome, there is an immediate interest in the humanized CAMPATH antibody because the rodent monoclonals are already in trials that will assess their clinical value in destroying the excess lymphocytes in advanced lymphocytic leukemia, and the unwanted donor lymphocytes in bone marrow and kidney transplants. But, says Herman Waldmann of Cambridge University's department of pathology, some patients rapidly

Other antibodies that Celltech intends to humanize include one against tumor necrosis factor (that the company hopes will find a place in the therapy of septic shock) as well as various hybrid effector molecules. In these hybrids, the antigen-binding portion of an antibody is grafted to the catalytic domain of an enzyme—a fibrin binding-site attached to a tissue plasminogen activator domain, for example. The reshaping of antibodies to make them as human as possible “will become a habit for us,” predicts Ed Lennox, Celltech's director

produce neutralizing antibodies to the rodent antibody. Humanized versions should induce only weaker antibodies (directed just against the idio-type of the hypervariable sequences).

“We are excited by the potential of humanized antibodies,” says a Wellcome spokesman, “but will await their preliminary clinical trials by the Cambridge group before deciding whether to push ahead with producing them.” One problem could be manufacturing sufficient quantities in existing expression systems.

—Peter Newmark