CANCER DIAGNOSIS

'SECOND ANTIBODIES' LEND A HAND IN IMAGING

NEW YORK—Researchers have long used antibodies attached to radioisotopes for the *in vivo* imaging of human tumors. One problem, however, has always been that some of the labeled antibody remains free-floating in the blood stream, thereby raising background levels of radiation and partially obscuring the desired image.

A number of strategies have been developed to circumvent this difficulty, and the latest to reach moderately large-scale human testing is second-antibody mediated clearance. The idea, which actually dates back to the mid-1960s, is to administer a second antibody which specifically binds to the primary imaging antibody. These second antibodies form aggregates with unbound imaging antibody; the body's reticuloendothelial system then clears the aggregates quickly and efficiently from circulation.

David M. Goldenberg, president of the Center for Molecular Medicine and chairman of Immunomedics Inc. (both in Newark, NJ), has now tested the technique in over 40 patients using ¹³¹I-iodine-labeled goat anti-CEA (carcinoembryonic antigen) IgG followed 24 hours later with donkey anti-goat IgG. As he reported at a recent meeting on "Cancer Progress" sponsored by Communitech (Yorktown Heights, NY), the newly patented method succeeded in improving signal-to-noise ratio in tumor imaging without deleterious effects on the patients. In fact, the technique occasionally pinpointed tumors that had been missed previously.

One of the potential strengths of

second-antibody clearance is that it might be used in combination with other methods of eliminating background radiation. For example, Goldenberg has also pioneered a technique known as subtraction. Here, another compound—such as technetium-99m-labeled human serum albumin—is administered to model the free-floating, unbound antibody. When this image is computer-subtracted from that of the labeled antibody, the "noise" from nonspecific background radiation is decreased drastically. Goldenberg says he is currently trying subtraction *after* second-antibody clearance to boost the signal-to-noise ratio even further.

NeoRx (Seattle, WA), a competing start-up company, uses a proprietary ligand to bind technetium-99m irreversibly to antibody fragments. President Robert Abbott reports that NeoRx doesn't need a second-antibody clearance approach because the body clears the firm's smallest antibody fragments to the kidney in as little as two hours, compared to about 24 hours for whole antibodies. Even so, says Goldenberg, applying the second-antibody approach could improve sensitivity with fragments as well.

Abbott cautions that some important questions must be answered before second-antibody clearance can be deemed effective. "Aggregates are very much more immunogenic than monomeric molecules," he stresses, so work must be done to determine how patients' immune systems react to the procedure.

Goldenberg admits that second-

antibody clearance is not perfect. The main fault he sees is that the body's reticuloendothelial system clears the radioactive material to the spleen. This is not a serious problem for imaging most solid tumors, he says, because these cancers rarely spread to the spleen. But it would be a drawback in detecting lymphomas.

"You haven't eliminated the material from the system" with second-antibody clearance, stresses Thomas McKearn, vice president of R&D at Cytogen Corp. (Princeton, NJ). Thus he prefers Cytogen's site-specific binding technique that attaches label to a non-binding portion of the antibody. In this way, more of the antibody remains capable of binding to tumor cells, so less is left in the circulation.

Many imaging procedures also have potential application in antibody-mediated cancer treatment. Goldenberg calls second-antibody clearance "an exciting approach to enhance the selective delivery of immunoconjugates in the therapy of cancer." McKearn, however, worries about damage these therapeutics might do to the cells of spleen—or wherever the antibody aggregates collect. "It's a toxic waste problem," he says. "One would prefer to use some method to concentrate antibody at the site of the tumor."

Goldenberg counters that immunoconjugates are cleared from the spleen in a matter of days, so side effects might not be too serious. Answers should be forthcoming from his current animal studies.

—Arthur Klausner

Left: Anterior view of abdomen. Only nontargeted ¹³¹I anti-CEA monoclonal is seen in blood-pool regions. Center: 24 hours after second-antibody injection, the kidney shows increased radioactivity. Right: 48 hours post second antibody, the radioantibody is more prominently localized to the kidney—and to the spleen where the complexes are retained.