

JANUARY 30, 2001

Novel Route to Lupus-Like Disease in Mice

Researchers have knocked out a gene that codes for an enzyme involved in modifying sugar molecules on the surface of cells, producing a disorder in mice that resembles the human disease, systemic lupus erythematosus.

Their finding represents the first time that an autoimmune disease has been linked to defects in cell-surface carbohydrate chains called glycans. According to the researchers, their work suggests that faulty glycan construction may play a role in the onset of human autoimmune diseases such as lupus. Autoimmune disorders, which are caused when the body's immune system attacks its own tissues, affect about five percent of people worldwide.

"We don't know if there are examples of human systemic autoimmune disease and lupus out there that may be due to mutations in this gene. But what concerns us is that there are currently very few clinical diagnostic tests for carbohydrate-based disease."

— Jamey D. Marth

The researchers, led by Howard Hughes Medical Institute investigator Jamey D. Marth at the University of California, San Diego, published their findings in the January 30, 2001, issue of *Proceedings of the National Academy of Sciences*.

"Until now, it's been known only by association that various autoimmune syndromes are shadowed by changes in glycosylation," said Marth. He cited, for example, that antibodies that recognize glycans are a central part of the immune system's ability to "see" foreign invaders. "However, it wasn't known whether changes in glycosylation cause autoimmune disorders," said Marth.

Past efforts to model systemic autoimmune diseases in animals—by genetically altering the function of white blood cells, for example—have not completely mirrored the human disorder. "In contrast, human autoimmune disease is often a long-term or chronic affliction and does not appear to be

associated with similarly overactive lymphocytes," Marth said. "Patients may live many years with the disease and some do rather well, but the disease waxes and wanes. And in human lupus, intrinsic defects in the immune system, such as the presence of dysfunctional lymphocytes, do not appear."

To determine whether defects in glycosylation play a role in autoimmune responses, Marth and his colleagues knocked out the gene in mice that produces α -mannosidase II, an enzyme that prunes mannose sugars from growing glycan chains on cell surfaces. Pruning enables more complex growth by glycosylation—in much the same way that pruning a tree alters its growth pattern. Without the enzyme, the cells show abnormal surface glycan formation. In earlier studies with mice, Marth and his colleagues had observed abnormalities that suggested the enzyme might be involved in an autoimmune reaction.

While the resulting knockout mice did not show acute symptoms of immune disorder as they aged, they did develop inflamed and scarred kidneys and autoreactive antibodies indicative of lupus-like abnormalities seen in the human disease. "The disease produced in these mice was chronic and long term, waxing and waning," said Marth. "And it doesn't appear to be due to defects intrinsic to lymphocyte development or activation, as these parameters were normal.

"As in humans with these disorders, these animals have a varied life span," said Marth. "They can live the human equivalent of sixty to seventy years, although their kidney function falters and some of them die much younger due to kidney failure."

Also intriguing, said Marth, was that the production of glycans in some tissues appeared to proceed via a pathway that did not require the α -mannosidase II enzyme. Exploring the differing pathways in the different tissues should give further insight into the role and machinery of glycosylation, he said. The clinical implications of the discovery of the effects of knocking out the α -mannosidase II gene are unclear, said Marth.

"We don't know if there are examples of human systemic autoimmune disease and lupus out there that may be due to mutations in this gene," he said. "But what particularly concerns us is that there are currently very few clinical diagnostic tests for carbohydrate-based disease. It's only been in the last five years that such syndromes have been discovered. And those were found serendipitously by a test for alcoholism that detects abnormal glycosylation in the liver due to alcohol intake," he said.

Marth emphasized that the role of glycosylation defects in producing autoimmune disease will not be known until clinics begin routine testing for such disorders—especially in the 50 percent of children who show symptoms of unknown inherited metabolic diseases.

Additional studies are needed to understand how knocking out α -mannosidase II produces an autoimmune disease, he said. The abnormal glycans might directly trigger the immune system, or they might indirectly

cause cell malfunction and death, that overactivate the immune system, creating chronic inflammation, said Marth.

Also, he noted, compounds that inhibit α -mannosidase II have shown therapeutic effect as an anti-cancer drug. "In animal studies, these compounds have inhibited tumor growth and metastasis," said Marth. "These findings raise the possibility that inhibiting the enzyme might modulate the immune-autoimmune threshold, perhaps resetting the rheostat enough to induce the immune system to suppress tumor growth," he said.