# BIOLOGY

*Julie Ann Miller reports from Washington at the International Symposium on Aging and Cancer*

## Drug resistance: Add-a-gene

Tumor resistance to drugs is a frustrating problem of cancer therapy. The explanation for that phenomenon may lie in the ability of cancer cells to produce and incorporate multiple copies of a gene, says Robert T. Schimke of Stanford University. Schimke first found evidence in laboratory-grown mouse and hamster cells (SN: 12/16/78, p. 421) that the cells that are resistant to the drug methotrexate contain as many as 200 times the normal number of genes for the enzyme dihydrofolate reductase. That enzyme is the target of the anti-tumor drug. Now Schimke reports preliminary evidence that the tumors of patients treated with methotrexate also can become resistant by making extra copies of the gene. The extra genes seem to be situated first on small, two-part, self-replicating units of DNA, which are called "double minutes." The extra genes can become more permanently incorporated into the cell's genetic information by attaching to one of the cell's chromosomes. Gene amplification is likely to be a common mechanism for the development of drug resistance, Schimke says.

## Back to the basic insulin gene

Rats have two insulin genes that make equal amounts of slightly different varieties of insulin. A surprising difference between the genes was revealed when they were analyzed with the powerful new techniques that allow scientists to determine the exact content of genetic material. One insulin gene is interrupted by a region of DNA that never makes a contribution to insulin or to either of its precursors. The other gene contains two such intervening sequences (SN: 7/7/79, p. 13). Is the second intervening sequence a relatively new addition to the rat genetic material or is it an old element recently lost from one rat insulin gene?

Howard Goodman of the University of California at San Francisco may have found the answer. He reports that both the human insulin gene and the chicken insulin gene have two intervening sequences in the same places as the interruptions in the second insulin gene of the rat. Therefore, the first rat insulin gene probably lost its second intervening sequence during rather recent evolution. Goodman proposes that one rat insulin gene arose from duplication of the other, followed by loss of the second intervening sequence. Future comparison of the two rat genes may give clues to a major puzzle of recent genetics: What is the function of the intervening sequences?

In related experiments with Paul Berg at Stanford University Goodman used a monkey virus, SV 40, to move one rat insulin gene into monkey cells that were growing in laboratory culture. He found that the cells make a protein that is identical by physical and immunological analyses to the insulin precursor called proinsulin. The monkey cells correctly trim an earlier insulin precursor, preproinsulin, and excrete the proinsulin appropriately.

## A story of skin

Cancerous skin cells and normal skin cells are growing on laboratory plates. Under proper conditions the normal cells form remarkably normal "skin." Howard Green of the Massachusetts Institute of Technology has studied the differentiation of those cells—the way they change from the layer of small, proliferating cells to the specialized cells of the skin surface. Green finds that the normal cells in culture follow all the biochemical steps observed in intact animal tissue.

In cells taken from human skin tumors, however, James G. Rheinwald of the Sidney Farber Cancer Institute in Boston has observed a partial defect in the process called "terminal differentiation." That process normally limits cell proliferation and regulates tissue size and function. While they are growing in a laboratory culture, the cancer cells exhibit some differentiation into mature skin cells, Rheinwald finds. But when he puts the cancer cells into a liquid medium, they retain the ability to proliferate far longer than do the normal precursors of skin cells. For example, in three hours, half of a sample of normal skin-forming cells loses the ability to grow from a single cell into a new cell colony, while half of the cancer cells from some tumors retain that ability for a week. Rheinwald says that in normal development it is crucial that precursor cells lose their proliferation potential at the proper rate and become committed to differentiate into mature cells. He and Green predict that these skin culture methods will yield new techniques for early diagnosis of skin malignancies.

## Getting older in 7,000 ways

Thousands of genes may influence the aging process, says George M. Martin of the University of Washington. He calculates that 7,000 genetic loci contribute to the aging of a person. That estimate is based on a survey of inherited diseases that seem to affect processes associated with normal aging. Martin says that adults with Down's syndrome and patients with an inherited form of Alzheimer's disease, for instance, have microscopic changes in their brains that are indistinguishable from those of ordinary senile dementia. Martin describes other rare diseases that grotesquely mimic aspects of aging. For example, a patient with Hutchinson-Gilford syndrome, or progeria, begins losing hair at age three and looks like a wizened elder by the age of 13. Such patients usually die of heart attacks in their early teens. Another disease resembling early aging is Werner's syndrome. Patients have accelerated aging in blood vessels, bones, skin, ocular lens and many other tissues. Werner's syndrome patients often develop diabetes and many kinds of cancer. Martin says that cells taken from such patients will reproduce fewer times in culture than will normal cells. The cells have many chromosomal rearrangements and deletions, including translocations rarely seen in normal subjects. Comparison of the genetic and physiological bases of these and similar disorders is expected to make important contributions to understanding of normal aging processes.

## Cultivating immune system components

The large number of cells involved in an immune response makes it difficult to assess their roles and employ them in disease therapies. "The task is fishing out any lymphocyte from the enormous sea in which it lives," says William E. Paul of the National Institutes of Health. He believes laboratory growth of distinct, homogeneous sets of cells will allow scientists to determine properties and functions. Paul has grown colonies of mouse cells of the type called T-lymphocytes. Those cells react to specific foreign compounds called antigens. The cells need a growth factor to proliferate in culture, and even then they don't grow with the exuberance of cancer cells, Paul explains. The descendants of a T-lymphocyte grown in culture all respond to the same antigen. Another group of immune system cells, called B-lymphocytes, have been grown under laboratory conditions for the first time, Paul reports. Cell cultures derived from mouse and from human cells have now been maintained for more than 6 months. The cells depend on a growth factor and can secrete "substantial amounts" of immunoglobulin, the material of antibodies. So far Paul has not formally proved that his B-lymphocyte colonies are homogeneous.