Reflections on Research and the Future of Medicine

With incidental reflections on the state of the art in molecular genetics, drug discovery, and other matters.

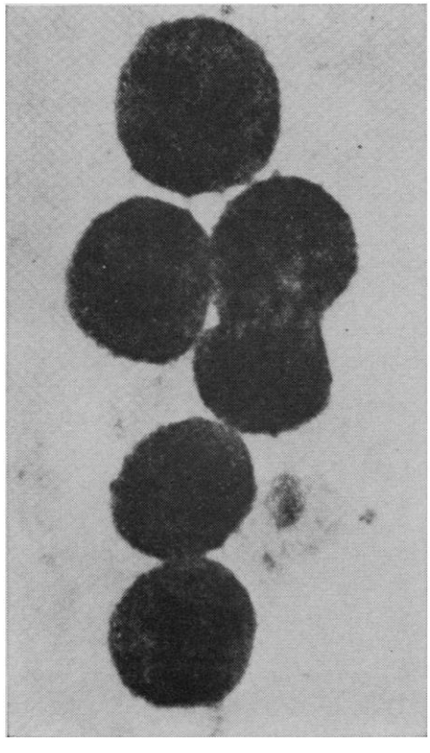
When the new commissioner of the Food and Drug Administration told a recent meeting of drug manufacturers that they were putting profits ahead of human lives, the assembled company presidents seemed unable to rally from the shock of the blunt language in time to agree on an effective answer.

A more thoughtful answer came in late May from one of the oldest and most prestigious of U.S. pharmaceutical companies. The occasion was the dedication of a new $7 million research building for the Merck Sharp & Dohme Research Laboratories at Rahway, New Jersey, and the method was a report to some 700 outstanding life scientists, invited to a gala affair, on recent achievements of research in the life sciences, without any narrow claim for the role industry has had in some of these but with some sober consideration of how the academic-government-industry combine can continue to jog along.

Ten Nobel laureates (some of whom had a working relationship with Merck) participated in the 2-day program, whose audience included presidents or deans of 12 neighboring colleges and 13 top-rank scientists from 10 other countries.

The meeting began with a symposium arranged by the medical faculty of Columbia University and was held in the alumni auditorium recently finished at the College of Physicians and Surgeons in New York. Columbia's president Grayson Kirk and Houston Merritt, dean of the College of Physicians and Surgeons, were on hand to open the symposium, joined by Nobel laureate Dickinson Richards. Chairman of the morning session was Robert Loeb, former member of the National Science Board, Columbia's Bard professor of medicine emeritus, co-author of the classic *Textbook of Medicine*, and pioneer of the group practice plan now rapidly spreading in the U.S. as a solution to the problem of how to distribute scientific medical care. Loeb's successor as chairman of Columbia's department of medicine, Stanley Bradley, was chairman of the afternoon session.

Nature



Future engineering of human genes for wanted characteristics is suggested by finding by Henry Harris, University of Oxford, that in culture, mammalian cells, even from different species, can be caused to fuse by exposure to unidentified component of certain viruses.

In this brilliant setting, reflections to match were expected and the speakers did not let their hosts down. For some time life scientists have been warning the community about the imminence of planned interference with human genetics: Nobel laureate Edward Tatum offered what amounted to an engineering blueprint for this, making this fateful power seem much closer than it had before. Nobel laureate Sir Macfarlane Burnet presented recent advances in understanding of autoimmune disease. Francis Schmitt, Massachusetts Institute of Technology, described some recent studies of neural mechanisms. All three speakers left the audience with a renewed sense of the unity of biochemical mechanisms operating in diverse disease entities and an increased respect for immunochemical techniques as a means of exploring them.

There was general agreement that the goal of medicine should not be, as it has recently seemed, to prolong the life span, but to prolong the middle years that are the prime of life, as Columbia's W. Henry Sebrell said. Sebrell spoke as commentator on the address by Sir George Pickering, Regius professor of medicine, University of Oxford, who said that contemporary society refuses to recognize the biological function of death. "Insofar as man is an improvement on monkeys, this is due to death," Pickering said. "A new species, for better or worse, can only begin with a new life."

Pickering said that the present goal of medicine seems to be "indefinite life, perhaps in the end with somebody else's heart or liver, somebody's else's arteries, but not with somebody else's brain"—for the adult brain seems an inaccessible pinnacle even to the most enthusiastic organ transplant specialists. If other transplants succeed, as they now give promise of doing, "those with senile brains will form an ever increasing fraction of the inhabitants of the earth. I find this a terrifying prospect."

By the time the guests had wound their way through dinner at the Plaza and traveled next day by bus along the New Jersey Turnpike to the dedication program and picnic lunch under festive green-and-white-striped tents run up alongside the new laboratories at Rahway, they had been exposed to a number of other themes. Among these: a vote of confidence in private enterprise from Vannevar Bush, America's senior statesman of applied science and former chairman of Merck's board of di-

Warman

Speakers in press conference were, from left, Ernst Chain, Sir George Pickering, Edward Tatum, Francis Schmitt, Sir Macfarlane Burnet.

rectors; a strong hint from U.S. Surgeon General William Stewart that the federal fund givers are looking now for research that will promise immediate clinical results; the opinion of Nobel laureate Ernst Chain, chemical father of penicillin, that today's study of enzyme systems has not, and will not at any time soon, yield clues to useful drugs.

Genes to Order

Edward Tatum used a summary of recent progress in molecular genetics as a foundation for what one listener called "the most astounding prospect so far suggested by science."

The prospect, as sketched by Tatum, is for genetic engineering: synthesis of human genes to order. "The time may soon come when a few molecules of such synthetic genes, or of genes isolated in a pure state from nature, will be replicated enzymatically in vitro," Tatum said. "With more complete knowledge of the biological processes and techniques involved in DNA uptake and integration, these DNAs can be incorporated into chromosomes."

Tatum pointed out that microbial life offers precedents for the scientist seeking to remodel man's own genes. In bacterial transformation, isolated DNA is taken up and integrated into a recipient genome. In transduction, DNA is transferred from one bacterium to another by a virus. Researchers are already trying to copy these processes, using mammalian cells in culture.

"The first successful genetic engineering will be done with the patient's own cells, for example, liver cells, grown in culture. The desired new gene will be introduced by directed mutation, or from normal cells of another donor by transduction, or by direct DNA transfer. The rare cell with the desired change will then be selected, grown into a mass culture, and reimplanted in the patient's liver."

Directed mutation looks possible, Tatum said, because experiments have already shown that substitution of one base for another in the DNA triplet can be forced. In nature, certain substitutions have been found to be more frequent than others, and "can selectively be made still more frequent by the incorporation into DNA of base analogs which change base pairing specificities."

Ultimately actual synthesis of the desired gene should be possible (synthesis of a biologically active RNA has already been accomplished), Tatum said, from specifications that will be clear when the genetic code is completely filled in.

Along these lines, the genetic engineering of human cells would evolve, supplying genes needed to make enzymes whose absence or short supply is responsible for a variety of deformities and disease states. Tatum said that a number of diseases will probably soon be added to the list of those now identified with specific enzyme defects. He also said it is reasonable to expect that the basic cause of cancer will be established within the next few decades.

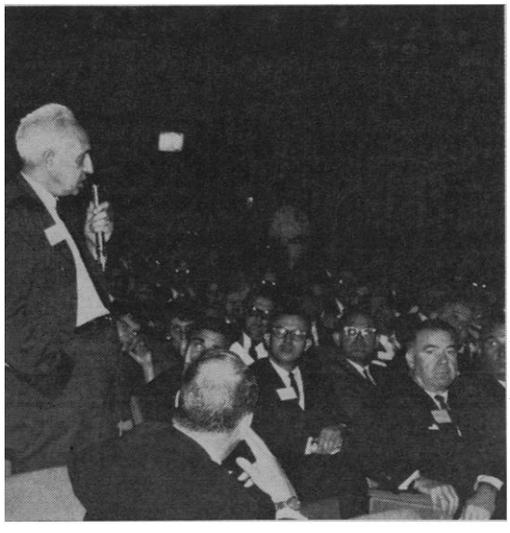
"All suspected causes, viral, mutational, or regulatory failures, center on cell genetics, and on nucleic acid structure and function. Hence we can predict effective preventive and curative therapy . . . by modification and regulation of gene activities, or by means of gene repair or replacement."

There is increasing evidence "that many forms of cancer in animals are indeed due to particular viruses in a phenomenon basically analogous to lysogenesis in bacteria." When viral genes are integrated into a host genome, the new cells produced by the "transformed" DNA show new structure, changed metabolism, and loss of contact inhibition.

Control of Gene Expression

We are already in an era of "euphenic engineering" without clearly recognizing it, Tatum said. He defined "euphenic" as improvement of the phenotype of the individual by measures that affect the expression of given genes, as distinguished from "genetic engineering" to change the character of the genes themselves. Dietary restric-

Vannevar Busch greets Mrs. Mildred Barry Hughes, New Jersey state senator.



Nobel laureate Severo Ochoa speaks from floor at daylong Columbia symposium.

From left, C. W. Mushett, director of international scientific relations for Merck laboratories; Dwight J. Ingle, University of Chicago; Bernard Halpern, Collège de France, Paris.

tions (as in phenylketonuria) and supply of vitamins and of hormones such as insulin are examples of euphenic engineering.

"Hormone therapy may actually represent gene regulation," Tatum said. He expects great development of this sort of euphenic engineering, with many new compounds yet to be discovered that will repress harmful dominant genes or turn on and off inactive genes, "even *in utero* at critical periods of development."

"Recent elegant experiments by Kidson and Kirby (*1*)" suggest that sex hormones regulate gene activity by derepressing production of messenger RNA. But other suggested modes of hormone action, for example, on cell membrane permeability, have not been ruled out.

Breakdown in the Body's Fail-Safe System

Many diseases whose cause is now unknown will probably be shown to result from an abnormal immune response directed against the body's own components, Sir Macfarlane Burnet, University of Melbourne, told the meeting. Such an immune response occurs when there is a breakdown in the body's "fail-safe" system for tolerating its own chemical components while rejecting those of any other organism.

"I am confident that there is still one primary mechanism of tolerance and that failure here is at the back of most of those forms of broken tolerance which manifest as autoimmune disease," Burnet said.

In Burnet's view, the thymus, while elaborating the cells of normal immune defense, also eliminates any cells carrying immune patterns capable of reacting with antigenic determinants carried by the body's own components—thus discharging "the censorship function that is responsible for natural tolerance."

On this model, autoimmune disease would arise from a cell which despite the fact that it was capable of immune reaction against a body antigen had escaped the censorship process. Such a cell would proliferate to form what Burnet calls a "forbidden" cell clone. Such a clone would not be automatically pathogenic. "There are strong suggestions that second level homeostatic controls exist," Burnet said.

The model also suggests that autoimmune disease has some analogy with conditioned neoplasms. Multiple myeloma (malignant tumor of the bone marrow) is an example of what Burnet calls "conditioned neoplasms." In such neoplasm, a "somatic cell line may be proliferating under what might well be called a hormone specific to itself, for this is precisely the function of antigen in relation to the immunologically competent cell," Burnet said.

"There is strong but not yet absolutely established evidence that inheritance plays an important part in allowing susceptibility to autoimmune disease in human beings," Burnet said, citing a recent study by P. R. J. Burch in which this worker concludes that in northwestern Europe about 50 percent of individuals are genetically susceptible to rheumatoid arthritis.

Evidence for genetic determination of autoimmune disease comes from certain studies in mice, Burnet said. He described his study over the last 8 years of a strain of New Zealand mice developed by Marianne Bielschowsky and their F1 hybrids "in which the genetic character of autoimmune disease is completely evident."

The hybrids are susceptible to kidney disease "which has all the characteristics of an autoimmune process" and which, when examined histologically, resembles a wide range of human kidney disease states, Burnet said. If untreated, all these mice die of kidney disease within 400 days.

In association with Burnet, Pamela German and Margaret Holmes have recently shown that cyclophosphamide arrests kidney disease in these mice. Like other alkylating agents, this drug is thought to act essentially to inhibit mitosis, particularly of rapidly dividing cells. This mode of action, Burnet said, suggests that kidney lesions in the mice strain "are based on the proliferation of immunocytes being stimulated by some intrinsic antigen."

Questioned after his lecture, Burnet said he had found no reason to modify his clone theory of antibody selection in any major way. "For 8 years immunologists have been trying to think up experiments to disprove this hypothesis," Burnet said, "and so far nobody has succeeded." Burnet thinks proponents of the "instructional" theory of antibody specificity have yet to explain the system that maintains nonantigenicity for the body's own components and the occasional breakdown in this system that results in autoimmune diseases.

As usual at meetings, not all the news was on the program. Immunologists present caught up in private conversations with the work of Bernard

Halpern, who holds Claude Bernard's chair in the Collège de France. Halpern has been studying the reticuloendothelial system for 15 years and considers it the "key of the natural defense." In a search for substances that might stimulate the system, Halpern recently found such a substance in a nonpathogenic mycobacterium. When injected with the substance, all animals inoculated with tumors survived. Halpern said the substance is not related to antibodies or to anything so far known of the bodily defenses and that it acts against bacteria and viruses as well as against tumors. "Several American pharmaceutical firms have isolated this substance for me and it is now in the purification stage. I think it may have great importance."

Molecular Recognition in Nerve Cells

Neuron circuits may be activated by some molecular "recognition" process like the selective interaction of antibody with antigen, Francis Schmitt, chairman of MIT's neurosciences research program, said.

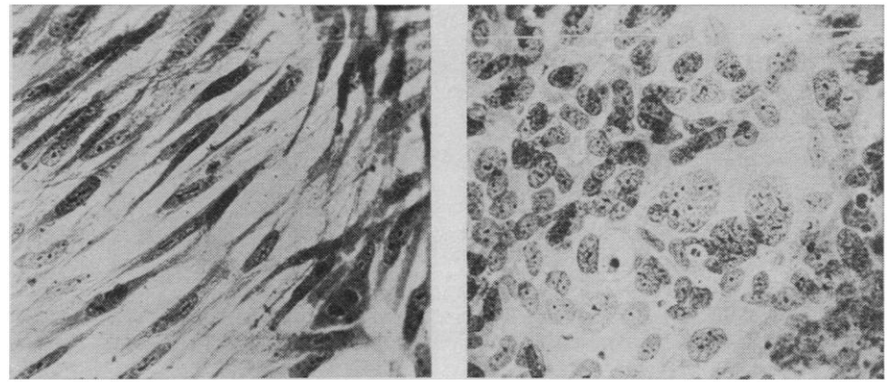
One important line of current study is the search, in both neuron and glial cells, for a protein functioning in the way that the immunochemists have demonstrated for the immunoglobulins. Just as a small part of the immunoglobulin peptide chain selectively interacts with just one of the thousands of different antigens entering the body, so such a neural protein would provide the structural basis for a similarly specific read-in and read-out of neurophysiological and psychological information.

Researchers in the Schmitt group are currently looking for a protein that may initiate the nerve impulse by action at the axon membrane. Schmitt calls this the "electrogenic protein."

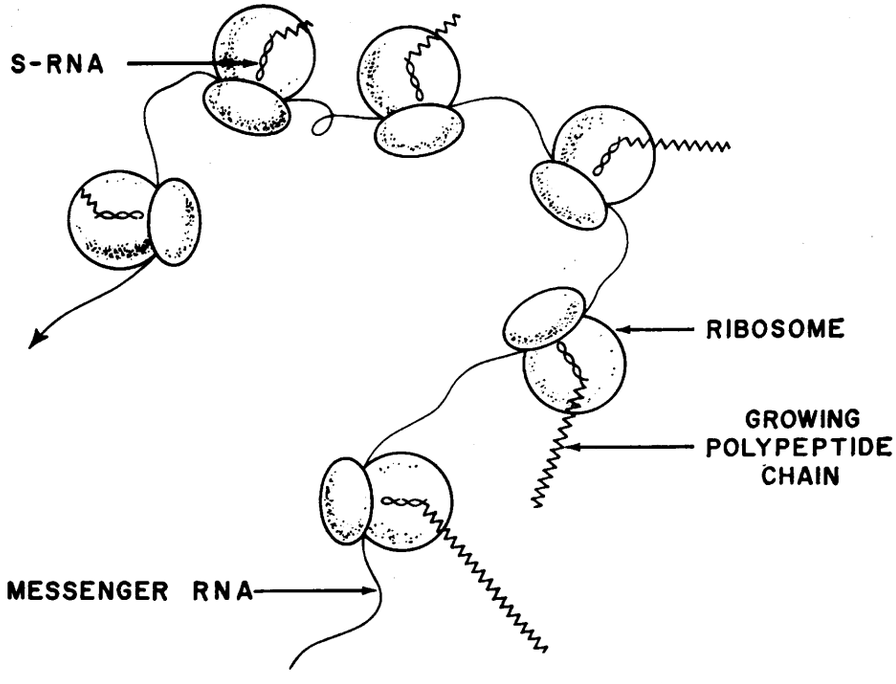
The view that a protein acts to initiate the action potential by changing membrane permeability to sodium ion finds some support in recent experiments in the Schmitt laboratories. These show that trypsin and other proteases destroy nerve excitability when introduced in fluid perfusing the giant nerve fiber of squid.

A related discovery, made by Huneeus-Cox at the Schmitt laboratory station in Chile, is that reagents combining with sulfhydril groups rapidly block the action potential when applied to squid axon. The blockade can be reversed by SH reagents such as mercaptoethanol.

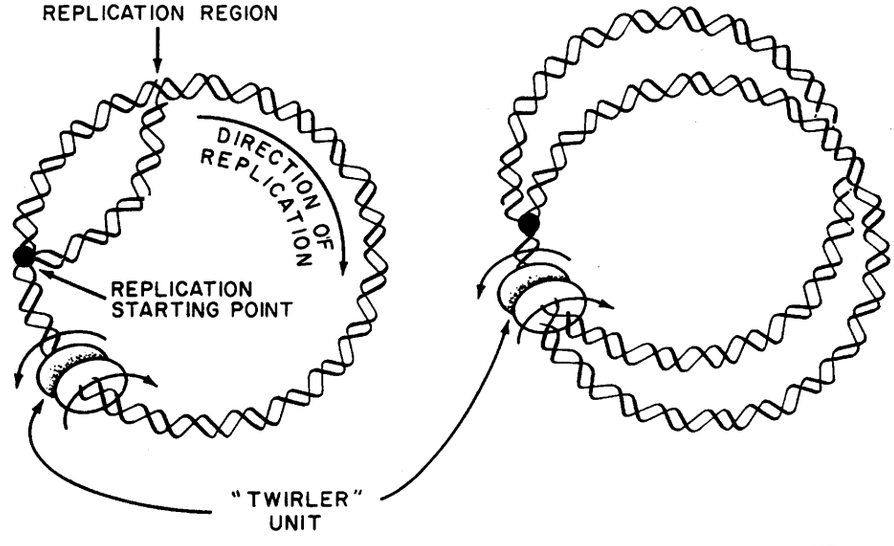
On the basis of this and other work,



Transformation of normal cells in culture, left, to cells whose shape is like that of tumor cells has been done experimentally by adding human and animal viruses. Transformed cells also show changed growth requirements and loss of contact inhibition. Such experiments have been done by Dulbecco, Dubos, and Trentin among others. Sometimes "helper" virus seems needed.



Messenger RNA may link ribosomes for protein synthesis, above. DNA molecule is thought to be circular, with replication moving from one end to the other. Because ends of circle are joined, a swivel has been proposed by John Cairns as mechanism for spinning both parent molecule and daughters as replication moves along. Drawings are from a book, "The New Genetics," by the late Engel, being edited by Edward Tatum for Doubleday publication.



Schmitt proposed the hypothesis that change in permeability of the axon membrane is the result of a reversible and fast change in molecular conformation. This may be either an intramolecular, tertiary conformational change or an intermolecular quarternary change like that by which actin molecules are thought to act in muscle contraction.

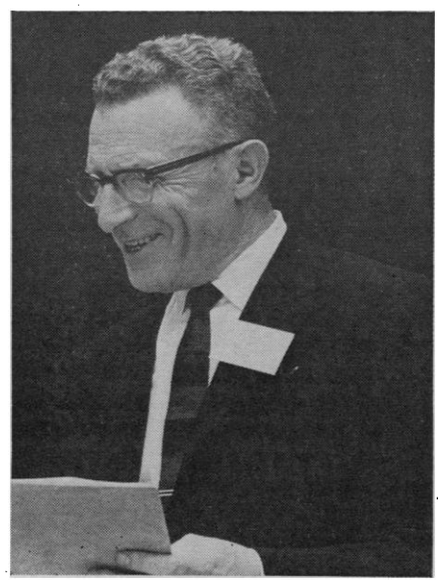
Immunochemical technique may make it possible to identify the electrogenic protein among the 15 proteins shown by electrophoresis to be present in squid axoplasm. Antiserum prepared in rabbits against whole squid axoplasm blocked the action potential. Next step is to fractionate the proteins. Then an antiserum prepared against each one will be tested for possible activity in blocking nerve conduction. Schmitt said that Huneeus-Cox and associates have made preliminary tests with one antiserum against the neurofilament protein, the major protein of axoplasm, and found that it does not affect the action potential. The function of the ubiquitous neurofilament (which hypertrophies in certain nerve diseases) is still completely unknown, but antiserum will provide a powerful tool for its study.

End of a Golden Age?

After this brilliant display of the resources of current molecular investigators, visitors were somewhat surprised to hear Ernst Chain, University of London, say at the dedication ceremony on the following day that the "dazzling success and rate of development which have characterized chemotherapy in the last few decades cannot be maintained in the future . . . unless a flow of new ideas based on the discovery of new biological phenomena occurs."

Will it be possible, on the basis of the molecular biology approach, to design tailor-made drugs which act as inhibitors or accelerators of enzyme action? Chain said he thought this approach is not worth serious consideration in the present state of knowledge.

"The solution to the problem of the structural configuration of the active centers of enzymes has proved elusive and seems to recede, like a Fata Morgana, as we approach it closer. . . . We have as yet not even a name for all the forces which are responsible for the tertiary structure of proteins. . . . The enzyme systems in which we are interested in relation to metabolic disorders . . . are not simple water-soluble entities, but complex multienzyme systems,



Max Tishler chose industrial research.

consisting sometimes of 50 enzymes and more in which the geometrical arrangement in space of the different components is one of the determining factors for their activity. . . ."

Those searching for new biodynamically active substances, Chain said, should pay attention to the biological and biochemical study of unusual animal species, hibernating animals, insects, slime moulds, and others and to interaction between biological systems. A fundamental problem which needs "a detailed systematic study with novel experimental approaches is that of permeability of different substances into different tissues."

Chain urged a large-scale, publicly financed effort to determine the amino acid sequence of a "thousand proteins with specific metabolic function," knowledge from which "we could derive some general structural rules which could be of use in the search for new biodynamic substances."

Coming out resoundingly for private enterprise in drug development, Chain said he knew of no government that would have taken the risks associated with the development of the antibiotics and that in 50 years not one really novel drug of interest has been developed in the Soviet Union.

Merck & Co. president, Henry Gadsden, said at the dedication that his purpose was "to put to rest . . . the concept that there is any subject, or any field, or any point in time where the interests of research and management should diverge." But the eccentric minds that even in the present era of Big Science are still conceded to be a necessary ingredient of research would probably take fright at Gadsden's confidence that "When you have a group, you must have a manager"—a confidence irresistibly reminiscent of right thinking in the better Yale groups, class of 1933.

Some might be further put off by Gadsden's remark that each member of the research team must be "prodded if need be" and that "scientific management is management in much the same way as business management is managment." But no doubt it is fairer to consider Gadsden's statement that the scientific staff is free to publish and that "Dr. Tishler and his associates know that they are free to make, and are responsible for making, their own medical and scientific decisions."

Gadsden's administrative experience is in production and marketing; in research, the climate of the group seems different. Take, for example, Max Tishler, an organic chemist who left the Harvard faculty in 1937 to join Merck and the only staff member of a U.S. pharmaceutical firm to hold membership in the National Academy of Sciences. "Max is a wonderful guy" is an estimate spontaneously offered by even the most junior among the 300 M.D.'s and Ph.D.'s on the research staff—although Tishler has now risen to the august position of president of the Merck, Sharpe and Dohme research laboratories and sits on the board of directors of Merck & Co.

Tishler's view of the research group seems different. "Each man in the research group is an individualist," he said in a conversation after the meeting. "Each one is dependent on all the others."

It is the group approach that makes industrial research the most stimulating place to be, in Tishler's opinion. Interdisciplinary study of problems in chemistry or in the life sciences was virtually unknown when he left the Harvard faculty, Tishler said. And he thinks industry is still way ahead of the universities on this score.

"Sometimes university researchers come to us for help on something that could be solved by a fellow on the floor below their own lab."

Tishler chose industry because he likes problem-centered research, as compared to the observational, analytic research that he thinks is often found in the academic world. "Actually research

to solve a given problem is more difficult. In observational research, if one line doesn't pan out, the researcher can shift to another question, or even to another field. But when you've got a problem to lick, the problem doesn't change—only your thinking about it must change."

"Observation—say, measuring how many times a bird in flight flaps its wings—is important in science, especially when it starts free-swinging thinking, but less interesting to me."

While Tishler is not unaware that observations of the flight of birds probably helped to get the first airplane off the ground, it is obvious that any thought he might give to a bird in flight would start with a wish to get a man up there.

Also responsible for the character of the Merck "group" are Karl Beyer and Lewis Sarett. Beyer, senior vice president for research at the West Point laboratories, led the development of the thiazide diuretics and is president of the Federation of American Societies for Experimental Biology. Sarett synthesized cortisone for Merck within 2 years after he took his Ph.D. at Princeton and has since risen to vice president for research at Rahway. All three men have an unaffected directness more often met on the campus than in the board room, and their cheerfully modest air does indeed suggest that it would not be too onerous to be part of the Merck group.

$85,000 NMR Spectrometer

For the June Ph.D., deciding where to place his career, the instruments available in a well-heeled industrial laboratory like that of Merck are an inducement that many academic laboratories still cannot match. Take, for example, the $85,000 nuclear magnetic resonance spectrometer that will be important in the new addition to Merck research announced at the laboratory dedication: a biophysics department headed by Nelson Trenner.

The spectrometer is one of the latest models, equipped with a computer for time-averaging, and has a highly stabilized and homogeneous magnetic field controlled by a feed-back element linked to a natural frequency of vibration of the water molecule. Trenner says he is perfectly satisfied with the stability achieved; in his view, this is the first NMR instrument that makes study of complex biological interactions really feasible at physiological concentrations.

A request for an electron-spin resonance instrument is contemplated, Trenner said, since this is now essential for studying the free radical intermediates by which most enzymes are thought to operate.

Trenner hopes soon to be set up for x-ray crystallographic methods that will show what happens when a crystalline solid is allowed to interact with a molecule in solution.

"Someday," Trenner said, "it might be possible to inject low molecular weight substrates into a living cell and monitor what happens by the changing spectroscopic pattern. We hope to use these structural studies to discover new biodynamic substances. But even if we don't succeed in that we should, at a minimum, be able to eliminate many substances that are not biologically active and relieve the animal people of drug assays that require much time and are subject to the uncertainties that follow from the well-known differences of drug reactions among the different species."

T. L. CAMPBELL

AAAS

References and Notes

1. C. Kidson and K. S. Kirby, *Nature* **203**, 599 (1964).
2. All photographs by Morris Warman except where otherwise credited.

Forthcoming Events

August

22–24. **Computer and Information Sciences,** symp., Columbus, Ohio. (J. T. Tou, Communication Science Research Center, Columbus Laboratories, Battelle Memorial Inst., 505 King Ave., Columbus, Ohio 43201)

22–24. **Physiology,** 12th Scandinavian congr., Turku, Finland. (K. Hartiala, Dept. of Physiology, Turku Univ., Turku)

22–26. Society of **Photo-Optical Instrumentation** Engineers, 11th annual technical symp., St. Louis, Mo. (R. T. Hedden, 16 Harneywold Dr., St. Louis 63136)

22–26. **Poultry Science** Assoc., Utah State Univ., Logan. (C. B. Ryan, Dept. of Poultry Science, Texas A&M Univ., College Station 77843)

22–27. **Food Science and Technology,** 2nd intern. congr., Warsaw, Poland. (A. Borys, Inst. Przemyslu Miesnego, Rakowiecka 36, Warsaw 12)

22–27. Pan American Federation of Associations of **Medical Schools,** 1st general assembly, Bogota, Colombia. (E. Braga, Caixa Postal 26-ZC-39, Rio de Janeiro, GB, Brazil)

22–10. **Science,** 11th Pacific congr., Tokyo, Japan. (Pacific Science Assoc., Bishop Museum, Honolulu, Hawaii 96819)

23–25. **Biological Photographic** Assoc.,