

the

Record

U. S. DEPARTMENT OF
HEALTH, EDUCATION, AND WELFAREVolume XIII, No. 18
September 12, 1961PUBLIC HEALTH SERVICE
NATIONAL INSTITUTES OF HEALTH

Near-Pure Virus Extracted from Leukemic Rats

The extraction, by two National Cancer Institute scientists, of nearly pure virus from the blood of laboratory rats infected with a virus-caused leukemia was announced last week by Surgeon General Terry.

"This," Dr. Terry said, "is a highly important finding. The availability of a relatively simple way to extract purified virus from cancer-bearing hosts will greatly speed our efforts to learn whether viruses cause human leukemia."

Seeks Virus in Humans

Scientists at NCI are already searching for possible leukemia-causing viruses in the blood of human leukemia patients, according to the Institute's Director, Dr. Kenneth M. Endicott.

Dr. Endicott said that virus extracted from the blood of rats is nearly free of extraneous material and causes leukemia sooner, and in a higher percentage of animals, than does virus recovered from other tissues of the same rats.

A full description of studies resulting in the finding was presented September 6 at a scientific symposium in Bern, Switzerland, by Dr. Albert J. Dalton, who, with Dr. John B. Moloney, conducted

(See NEAR-PURE VIRUS, Page 6)

Prevalence of Mycoses Presents Challenge To Medical Research

The serious nature of the fungal disease problem was stressed by Dr. Chester W. Emmons, Chief of the Laboratory of Infectious Diseases, National Institute of Allergy and Infectious Diseases, in the Annual Lecture to a meeting of the Mycological Society of America at Purdue University last month.

Dr. Emmons said that although therapeutic use of antibiotics has almost eliminated many of the infectious diseases as causes of death in the United States, conquest of the mycoses (fungal diseases) remains a serious medical problem.

Fatalities Constant

He said that reported deaths resulting from mycoses have held almost constant since they have been reliably reported. (To illustrate the prevalence of mycoses, it is estimated that 30 million people now living have had histoplasmosis, a mycotic lung infection, often inapparent but sometimes fatal.)

In reviewing mycology, the science and study of fungi, and its historical association with medicine from the Greek and Roman eras down to the present day, Dr. Emmons demonstrated the role of fungi in man's prescientific grop-

(See MYCOSES, Page 2)

Dr. Ralph Knutti Is Named NHI Head; Dr. Watt Accepts International Post

Surgeon General Terry has announced the appointment of Dr. James Watt, Director of the National Heart Institute since 1952, as Chief of the Public Health Service's Division of International Health. The appointment became effective yesterday.

Dr. Watt's successor as NHI Director is Dr. Ralph E. Knutti, Associate Director for Extramural Programs, National Institute of Arthritis and Metabolic Diseases.

Soviet Microbiologist Works in NIAID Lab

The second Russian medical scientist to come to the United States for extended research under the 1960-61 Cultural and Scientific Exchange Agreement is working in the Laboratory of Infectious Diseases, National Institute of Allergy and Infectious Diseases.

Here for a "general exchange of information, methods, and thinking," Dr. Nikolai Petrovich Yelinov, Deputy Director of the Leningrad Chemical-Pharmaceutical Institute, plans a 4-month program of research studies on deep mycoses (fungus-caused diseases).

The Soviet scientist is interested in learning U.S. methods of diagnosis, treatment, and prophylaxis of such deep mycoses as coccidioidomycosis, histoplasmosis, and North American blastomycosis. These last two have never been identified in the Soviet Union.

Heads Microbiology Chair

Head of his Institute's Chair of Microbiology, Dr. Yelinov is also on the staff of the All Union Scientific Research Institute of Antibiotics in Moscow. He holds the degree of Candidate in Medical Science, a degree approximately equivalent to an M.D. degree in the United States.

Dr. Yelinov whose research field is the pathogenic (disease causing) microbes of man and animals—is also studying methods of laboratory production of specific antigens (substances which stimulate the production within the body of antibodies to combat disease).

He brought with him samples of Russian coccidio culture for com-

(See MICROBIOLOGIST, Page 7)

Dr. Knutti entered the PHS Commissioned Corps in 1951 as a Medical Director. Since that time he has been continuously associated with the NIAMD extramural programs and has held his present position since September 1960.

Before coming to NIH he was Director of Laboratories at the



Dr. Knutti

Dr. Watt

Los Angeles Children's Hospital.

He has also been an instructor in pathology and research at the Rockefeller Institute, the University of Rochester, and an associate professor of pathology at the University of Southern California.

A native of Palo Alto, Calif., Dr. Knutti received his M.D. degree from the Yale University School of Medicine in 1928, and served his internship at Lakeside Hospital, Cleveland, Ohio.

In addition to directing the Service's Division of International Health, Dr. Watt will be Dr. Terry's chief assistant and counsellor on international health affairs.

He will also be responsible for coordinating PHS programs on the international scene and for relating these programs to other Federal and worldwide activities in the health field.

Dr. Watt's experience in international health activities dates

(See DR. KNUTTI, Page 2)

Annual UGF Campaign Begins Oct. 2

The takeoff date for the 1961 United Givers Fund campaign in the metropolitan Washington area is October 2.

Federal agency campaign dates and dollar quotas for NIH Institutes and Divisions will be announced shortly, according to the UGF chairman for NIH, Chris A. Hansen, Chief of the Division of Research Services.

Mr. Hansen expects that the timing of the NIH campaign will be similar to that followed in 1960.

"We can expect the kickoff rally to be held early in October," he

reported last week.

Roy Perry, Chief of the Photography Section, Medical Arts and Photography Branch, DRS, is the campaign's information chairman. Campaign efforts throughout the Institutes and Divisions will be coordinated by Robert H. Handy, DRS Administrative Officer.

Dr. G. Halsey Hunt, Chief of the Division of General Medical Sciences, is vice chairman for the 1961 fund drive and will be chairman next year, in keeping with the practice of rotating the chair among the Institute Directors and Division Chiefs.

the Record

Published bi-weekly at Bethesda, Md., by the Public Information Section, Office of Research Information, for the information of employees of the National Institutes of Health, principal research center of the Public Health Service, U. S. Department of Health, Education, and Welfare.

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PERSONNEL TO PERSON

SCHEDULES for the fall secretarial-clerical training programs have been distributed to all NIH Institutes and Divisions by the Personnel Management Branch.

The program, which is conducted by the Employee Development Section, will offer a course in business English and two courses in shorthand—one for candidates for the Civil Service Stenographic Examination and a second for stenographers who wish to improve their writing skills.

Persons interested in these courses should discuss with their supervisors the possibility of attending.

All nominations must be received by Division or Institute Personnel Operations Officers by September 14.

EDUCATION COUNSELING

Services of William L. Fournier, Educational Counselor, George Washington University, will again be available this year to counsel NIH employees on their academic interests and needs.

Assistance and counsel will not be limited to any particular field, and Mr. Fournier will have curricular information from all the local colleges and universities.

Mr. Fournier may be seen by appointment on September 12 and 19, in Bldg. 1, Rm. 114, between 10:30 a.m. and 2:30 p.m.

To schedule appointments call the Employee Development Section, Ext. 2147.

CLAIM KITS

Aetna Life Insurance Company has issued "Claim Kits" for all employees enrolled in the Government-wide Indemnity Benefit Plan. The kits are designed for use in maintaining claim records.

The booklet, "How Your Benefits Are Paid," included in these

Outpatient Clinics Listed In Psychiatric Directory

Outpatient psychiatric services in the United States increased 16 percent from 1956 to 1959, according to a nationwide directory issued recently by the National Association for Mental Health in cooperation with the National Institute of Mental Health.

The directory, Outpatient Psychiatric Clinics, lists all regularly scheduled clinics in which a psychiatrist has medical responsibility for the welfare of patients. It also lists State and Federal hospitals for the mentally ill and mental defectives, State mental health associations, State departments dealing with mental health, and all regional offices of HEW.

Data Collected

The 1959 data on over 1,400 clinics were collected and prepared by NIMH Outpatient Studies Section, Biometrics Branch, from the 1959 Annual Report of Outpatient Psychiatric Clinics.

The Annual Report indicates that the number of clinics has increased from 1,234 in 1956 to 1,429 in 1959. Also, the number of professional man-hours available in the clinics has increased 41 percent to 265,000 man-hours per week.

Single copies of the directory are available without charge from the NIMH Publications and Reports Section, Rm. 317, National Bank Bldg., Bethesda, Md. The telephone extension is 4795.

kits contains two sets of instruments for filing claims: a supplementary sheet of instructions to be used prior to November 1961; and instructions for filing claims after the November 1961 revised plan becomes effective.

Employees enrolled in this Health Benefits Plan, may obtain a kit from the office of their Institute or Division Personnel Representative, upon presentation of their Aetna identification cards.

MYCOSES

(Continued from Page 1)

ing toward an understanding of the causes of disease and a search for defenses against illnesses. He said that fungi have been used, though perhaps unscientifically, throughout man's history for medicinal purposes.

Dr. Emmons pointed out that medical mycology is not an obscure specialty but an important area of investigation, one that is supporting a revolution in the area of infectious diseases. He noted that Fleming's observation of the antibiotic action of the mold that produces penicillin has led to a host of drugs based on molds which are more effective against the bacterial invaders of man's body than most of those which come from the chemist's laboratory.

Elderly Patients Threatened

He said that such therapeutic triumphs have not been achieved in the mycoses, although there are at least 50 types of fungi that cause disease in man. Two antibiotics, Amphotericin B, and the experimental drug X-5079C, have proved effective in treating systemic fungus diseases in some patients, but at present these are balanced by more frequent diagnosis of fungus infections as a result of improved methods. As the life span of man increases, fungi may parasitize older patients or those whose resistance has been weakened by other diseases.

In closing, Dr. Emmons said that diagnostic mycology is still a developing science requiring proper training and research ability.

Harry T. Aylor Wins Performance Award

Harry T. Aylor, a research technician in the Laboratory of Control Activities, Division of Biologics Standards, received a citation for



Mr. Aylor

sustained superior performance and a check for \$200 at an award ceremony held August 25 in the DBS Conference Room. An NIH employee for the past 23 years, Mr. Aylor was specifically commended for outstanding work in the DBS control testing program relating to "his knowledge and experience with a wide variety of highly technical skills, and his ability to supervise and train personnel in specialized control testing techniques."

Mr. Aylor is co-author of two published papers pertaining to work on smallpox vaccine.

4 New Consultants on Environmental Health Added to SEB Staff

The Sanitary Engineering Branch, Division of Research Services, has announced the recent appointments of four consultants on environmental health problems.

Dr. Harold J. Fournelle, formerly with an International Cooperation Administration team in Colombia, South America, has been named head of the Bacteriology Unit of the Sanitation Section.

He will be responsible for microbiological studies in environmental health investigations conducted by the SEB. Such investigations will include studies of the bacteriological quality of air in the Clinical Center surgical and patient areas and bacteriological analyses of food, milk, and water.

The other new employees are James M. Cox, Herbert H. Jones, and Albert S. Gates, Jr.

A PHS Reserve Corps Sanitarian on active duty in the Sanitary Engineering Branch, Mr. Cox will be responsible for the organization and planning of training courses in basic sanitation and the review from a sanitation standpoint, of plans for new construction and alteration projects at NIH.

Before coming to NIH, Mr. Cox was an instructor at the University of North Carolina.

Mr. Jones is a PHS career officer who was formerly in the Cincinnati office of the Division of Occupational Health, Bureau of State Services. He will use his experience in industrial hygiene engineering to seek solutions to environmental health problems.

Mr. Gates, a mechanical engineer previously with the Bureau of Ships, United States Navy, will have chief responsibility for equipment development in environmental engineering at NIH.

DR. KNUTTI

(Continued from Page 1)

from the start of his career in the PHS Commissioned Corps in 1938 when he was assigned to the Mexican border States to investigate outbreaks of enteric diseases.

At the request of the Chinese Government he went to Chungking in 1945 to investigate and develop control measures for a serious outbreak of cholera, and subsequently, at the request of the Pan-American Sanitary Organization, went to Brazil and other Latin American countries for similar epidemiological activities.

In 1945, he received the Bailey K. Ashford Award of the American Society of Tropical Medicine for his research in tropical medicine.

Antinuclear Sera React With Chromosomes of Man, Other Mammals

For several years it has been known that sera from patients with certain diseases, particularly lupus erythematosus, have antinuclear activity. When a patient's serum is incubated with human or other mammalian cells, gamma globulin in the serum reacts with the nuclei of the cells. If the cells then are incubated with fluorescently labeled antihuman globulin, the nuclei will fluoresce under ultraviolet light.

Antinuclear Activity Noted

The discovery that certain sera having antinuclear activity also react with human and other mammalian chromosomes, is reported in *Science* by Dr. Robert S. Krooth, working at the National Institute of Allergy and Infectious Diseases on loan from the National Institute of Neurological Diseases and Blindness, Drs. John E. Tobie and Howard C. Goodman of the National Institute of Allergy and Infectious Diseases, and Dr. J. H. Tjio of the National Institute of Arthritis and Metabolic Diseases.

Sera from patients with lupus erythematosus, Sjogren's syndrome, and nephrosis first were tested for antinuclear activity by the investigators. All sera reacted with mouse liver nuclei when tested by the fluorescent antibody technique.

The sera were then allowed to react with chromosomal preparations made from human peripheral blood cells and a variety of other human and subhuman mammalian tissue culture cells; subsequently, the chromosomes were stained with fluorescein-labeled horse antihuman globulin.

Fluorescence Observed

Serum from five of six patients with systemic lupus erythematosus (SLE) reacted with chromosomes, as indicated by chromosomal fluorescence. Most of the sera possessing such antinuclear activity apparently had reacted with all of the chromosomes of the cells, since 46 discrete fluorescent chromosomes were usually observed. When, after fluorescent staining, the same cell was restained with acetic acid-orcein, every chromosome stained with fluorescent antibody was also stained with orcein.

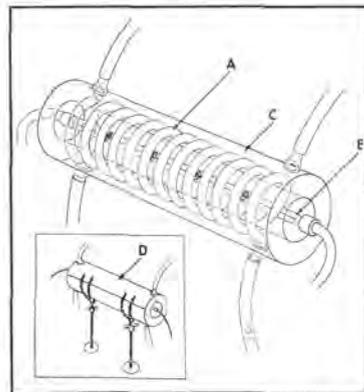
Serum from one of two patients with nephrosis also reacted with chromosomes of all of the mentioned cell types when examined by this method. However, serum from the three patients with Sjogren's syndrome could not be demonstrated to react with chromosomes, even though clear evidence

DBS Scientist Develops New Method For Selective Inactivation of Viruses

A new method of inactivating potentially harmful organisms in live virus vaccines has been developed by Dr. C. W. Hiatt, of the Division of Biologics Standards.

The method could have an important application to the manufacture of live attenuated poliomyelitis vaccine, since the presence of viruses of monkey kidney origin at various stages in the manufacturing process constitutes a major production problem.

A paper on the results of his studies, "Differential Rate Methods for the Selective Inactivation of Viruses," was presented by Dr.



Apparatus for continuous-flow irradiation of liquids. Liquid is pumped through glass tubing (A) around light source (B). The coil is surrounded by a water jacket (C) and metal reflector (D).

Hiatt at the 7th International Congress of Biological Standardization in London last month.

His method is one of several differential rate methods in which the extraneous virus is inactivated more rapidly than the poliovirus.

The method represents a new concept of an old technique—that of photodynamic inactivation—first reported in 1898. It involves the effect of visible light on living organisms in the presence of trace amounts of certain dyes.

Dr. Hiatt discovered that while some animal viruses combine with the dye and thus become sensitized to ordinary light, others including

that it possessed antinuclear activity could be demonstrated by its positive reaction with mouse liver cell nuclei.

In the light of these findings, the authors suggest that perhaps certain sera may be found which are more specific in their chromosomal reactions. The authors also suggest additional studies directed to the determination of the nuclear antigens involved in these reactions so that the significance of antinuclear factors, found in the sera of patients with diseases such as lupus erythematosus, can be better understood.

poliovirus, are highly resistant to this treatment.

To date, Dr. Hiatt has successfully inactivated five of the 40-odd simian viruses known to be present in monkey kidney tissue cultures. Simian viruses 1, 5, 12, 15, and 40 have been found to be susceptible to photodynamic action, while SV₁₆, SV₁₈, and SV₁₉ are resistant, indicating that the latter probably have some characteristics similar to the poliovirus.

The Coxsackie and ECHO-1 viruses, true enteroviruses affecting the intestinal tract, are also resistant.

Non-Toxic Dye Used

Dr. Hiatt's method consists of the addition of a minute, non-toxic quantity (one to ten parts per million) of the thiazine dye, toluidine blue, to the monkey kidney tissue culture fluid to be treated.

The fluid is then pumped through a continuous flow apparatus, consisting of a coil of glass tubing surrounding an intensely brilliant incandescent bulb giving off visible white light. The viruses that are susceptible to photosensitization by the dye become direct targets for the destructive bombardment of light energy.

The incandescent bulb used in this process is a new type, developed by General Electric for special uses, such as the illumination of airports. Its pencil-thin, tubular shape is ideally suited to the continuous-flow method apparatus developed by Dr. Hiatt.

The method has been successfully adapted to pilot plant operation, and is feasible for processing commercial volumes of the vaccine.

NIAID Scientist Gives R. R. Parker Address

Dr. G. Robert Coatney, Chief of the Laboratory of Parasite Chemotherapy, NIAID, delivered the R. R. Parker Memorial Address August 22 during the Sixteenth Annual International Northwest Conference of Diseases in Nature Communicable to Man at Colorado State University. His subject was "Simian Malaria; a New Zoonosis."

The address is presented annually in honor of Dr. R. R. Parker, Director of the NIAID Rocky Mountain Laboratory in Hamilton, Mont., from 1928 to 1949.

In the 1920's Dr. Parker did research in the Bitterroot Valley, Mont., on Rocky Mountain spotted fever, at that time a fatal disease with no known treatment. His research was responsible for much of the work that culminated in an effective vaccine.

Calcium-High Diet Found to Offset Bone Weakening

Scientists of the National Institute of Arthritis and Metabolic Diseases have found that diets high in calcium may offset the thinning of bones in arthritis patients treated with cortisone and other corticosteroids.

Osteoporosis, a "bone thinning" disease marked by excessive calcium loss, principally from the spine, affects 30 percent of women past the menopause and is probably more common in older women with rheumatoid arthritis, according to the NIAMD researchers.

Although the bone disease is known to worsen in patients who receive corticosteroid hormone therapy for relief of their arthritis, the mechanism by which bones are weakened through hormone action is not clear.

Hormone Effects Studied

To investigate the effects of corticosteroids such as cortisone and other hormones, Dr. S. Donald Whedon, Chief of NIAMD's Metabolic Disease Branch, and his associates, Dr. Leo Lutwak and Preston Smith, performed 13 studies, each lasting several weeks, to determine the amounts of calcium lost or gained in 11 patients given different hormone preparations.

Dr. Whedon said his group found that the hormones increased calcium excretion from the body in two, and decreased its loss in three. Despite this varying response, he said these results generally support the belief that corticosteroids cause calcium loss.

Dr. Whedon reports that large increases of dietary calcium—equal to more than a quart of milk a day above ordinary diet levels of calcium, by supplements of milk products and calcium lactate tablets—produced significant calcium storage in two studies even during corticosteroid hormone administration.

Prevents Calcium Loss

Similar diets given to seven patients with post-menopausal osteoporosis who were not receiving hormones, not only prevented calcium loss in six of the patients but also enabled four of them to begin storing calcium, according to the NIAMD scientists.

This suggests that abundant calcium in the diet continued during adult years may protect the skeleton, although many factors appear to be involved in the body's

(See *CALCIUM*, Page 4)

Virus-Cancer Program Given Branch Status; CCNCS Reorganized

Dr. Kenneth M. Endicott, Director of the National Cancer Institute, has announced the recent establishment of a new branch in the Viruses and Cancer Program, and the reorganization of the Cancer Chemotherapy National Service Center.

The new Virology Research Resources Branch, under the direction of Dr. Harvey I. Scudder, will administer and further develop the NCI program for promoting the study of viruses as a possible cause of human cancer.

Disseminates Information

It will have the responsibility for letting contracts to support the development of special material needed for virus-cancer research, such as tissue culture cell lines, virus detection agents, special experimental animals, and normal and malignant human tissue.

The new branch will also disseminate information on progress in the virus-cancer field.

The CCNCS has been reorganized into six branches. With their chiefs they are:

The Drug Development Branch, Dr. Howard W. Bond; Drug Evaluation Branch, Dr. Joseph Leiter; Clinical Branch, Dr. T. Phillip Waalkes; and Endocrine Evaluation Branch, Dr. Erwin P. Vollmer.

New Sections Established

Also, the Research Communications Branch, Dr. Jonathan L. Hartwell; and Operations Branch, George A. Brandner. In addition, a Biometrics Section headed by Dr. Edmund A. Gehan has been established in the CCNCS.

Both the Viruses and Cancer Program and the CCNCS are under the supervision of Dr. Stuart M. Sessoms, NCI's Associate Director for Collaborative Research.

CALCIUM

(Continued from Page 3)

utilization of calcium.

"There are indications," Dr. Whedon said, "that calcium requirements vary considerably among individuals and are probably higher than normal in individuals who develop osteoporosis." He reported that the studies support previous findings that high calcium diets are not harmful.

Dr. Whedon said the studies suggest that a high calcium diet is now the most reasonable clinical procedure to protect arthritic patients against the bone-depleting effects of corticosteroid therapy.

3 New Members Join Virology Branch Staff

Dr. Harvey I. Scudder, Chief of the Virology Research Resources Branch, National Cancer Institute, has announced the appointments of three new staff members.

Dr. Randall L. Thompson will join Dr. Marvin M. Harris in developing guidelines for the production, standardization, certification, and distribution of human viral typing reagents of importance to research workers in cancer virology.

Assists in Program

Dr. Leslie C. Murphy, a Lieutenant Colonel in the Veterinary Corps, U. S. Army, will assist Dr. Robert Holdenreid in directing a laboratory animal program leading to the production of specialized animals for experimental use.

Dr. Theodore Malinin has been assigned to the cell culture and tissue procurement areas of the program. He will work with Dr. Robert E. Stevenson in stimulating cell culture research and in establishing general principles and procedures for procuring human tissues needed for cancer virological research.

Conducts Medical Research

Dr. Thompson, formerly Head of the Microbiological Section of Sterling-Winthrop Research Institute, Rensselaer, N. Y., holds B.S. and M.S. degrees from the University of Washington, and a D.Sc. degree from Johns Hopkins University. He received his M.D. degree from the University of Chicago. He has taught in several medical schools and conducted research, including studies on the chemotherapy of viral diseases.

Dr. Murphy has a B.S. degree from the University of Idaho and a D.V.M. degree from Washington State College. His most recent tour of duty was at Ft. Detrick, Frederick, Md., where he was Chief, Branch IV, VR Division, Biological Laboratories and Staff Veterinarian. Dr. Murphy's thesis is in preparation for an M.S. degree in the Department of Microbiology of the University of Maryland.

Trains in England

Dr. Malinin is a 1960 graduate of the University of Virginia Medical School. He received additional training in pathology at the University of Cambridge, England, where he was a National Foundation Fellow during the summer of 1959, and at the John Hopkins University Hospital in 1960-61 where he served as an intern.

He is presently on active duty in the PHS Commissioned Corps assigned both to NCI and Bethesda Naval Hospital.

Complex Enzyme Structure Found Ingeniously Designed

Enzymes are the protein executives that expedite nearly all of the body's biochemical business, according to Dr. Christian B. Anfinsen, Chief of the Laboratory of Cellular Physiology and Metabolism, National Heart Institute.

In the assembly of their often very large and complicated molecules from twenty basic amino acid building blocks, enzymes must meet an extremely complex, interlocking set of requirements.

The operation of the evolutionary processes of mutation and natural selection has resulted in enzyme molecules so ingeniously designed that very little is left to chance in their synthesis.

Speaking at the Fifth International Congress of Biochemistry held in Moscow last month, Dr. Anfinsen cited evidence that he and his NHI colleagues had gained from studies on the enzyme ribonuclease.

Follows Automatically

Their findings suggest that once the protein chain of this enzyme has been forged, the coiling of the chain to form the active center and three-dimensional conformation required to confer enzymatic activity follow automatically and reproducibly. Apparently all of the genetic information needed for this process is contained in the amino acid sequence of the chain.

Enzymes appear to be made in the cell under the control of ribonucleic acid (RNA). This substance carries the genetic directions for choosing from among the twenty available amino acid building blocks, the proper number of the proper amino acids and for stringing them into the proper sequence to form the protein chain or chains of the enzyme.

Codes Instructions

The RNA, in turn, is probably constructed according to coded instructions carried by the deoxyribonucleic acid (DNA) of the cell nucleus. DNA, which can duplicate itself, appears to be the repository of a major portion of the body's genetic information.

While the RNA blueprint determines the basic sequential structure of the enzyme chain, the final step in enzyme synthesis—and the one necessary to confer enzymatic activity on the molecule—is the coiling or folding of the extended chain to form the enzyme's active center and its unique three-dimensional configuration.

This configuration must provide for "substrate fit," so that the substance processed by the enzyme can get at the enzyme's active center to the exclusion of other substances. The formation of the active center may require bringing

into close association amino acid residues that are widely separated in their linear arrangement along the chain.

The NHI studies on ribonuclease were concerned with whether this final step in its synthesis was also determined by the RNA blueprint or whether it might be predetermined, at least in part, by the amino acid sequence of the protein chain.

Ribonuclease consists of a single chain of 124 amino acids coiled into a roughly spherical shape. It is held in this configuration by four sulfur-to-sulfur (disulfide) bridges connecting portions of the chain at four points.

'Uncoils' Chain

Dr. Anfinsen and his colleagues "uncoiled" this chain, using chemical reagents that selectively break these disulfide bridges, but not the chain itself. The uncoiled molecule exhibited none of the enzymatic activity of the native ribonuclease.

In the presence of molecular oxygen and suitable conditions of alkalinity, the uncoiled chain was able to reorganize spontaneously, regaining both its original configuration and its lost activity.

This spontaneous reorganization indicated that the amino acid sequence of the chain carried all of the genetic information needed to determine the secondary and tertiary structure of the enzyme. The NHI scientists next explored the biochemical nature of this information.

Alignment Important

Of primary importance was the linear alignment of the amino acid links of the chain so that the coiling process would bring the right amino acids into the right spatial arrangement to form the active center. The process must also bring together in pairs the eight sulfur-containing cystine units, so that these can link up to form the four disulfide bridges.

The formation of these bridges was also dependent upon the operation within the chain of complex electrical and chemical forces.

One of these was the distribution of electrical charge in the molecule, which is affected in turn by

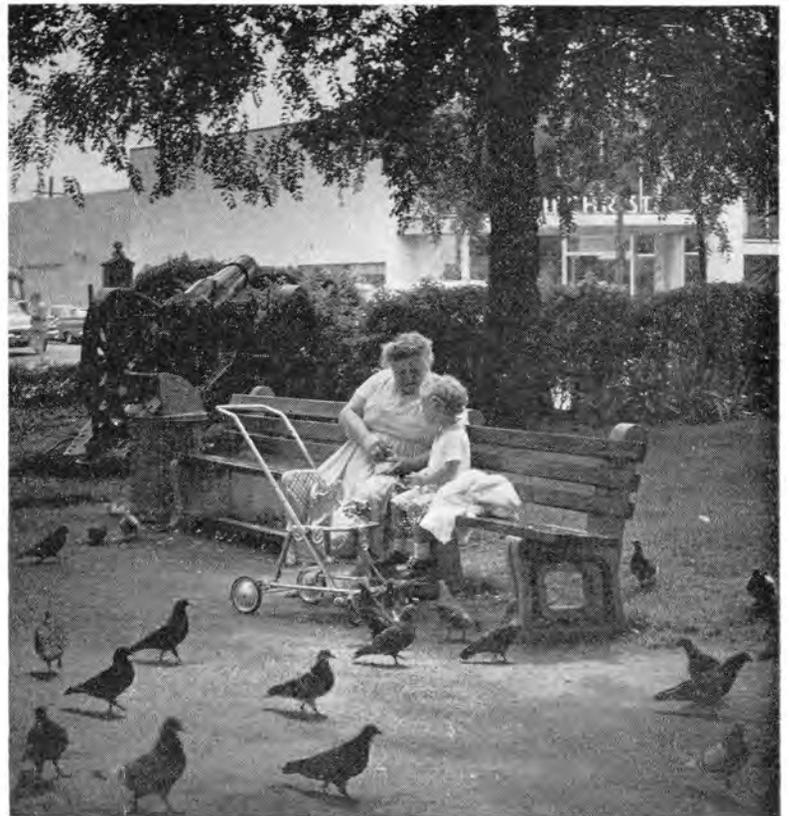
(See ENZYME, Page 3)



Dr. Thomas R. Dawber (right), Medical Director of the Heart Disease Epidemiology Study in Framingham, is shown with Dr. C. George Tedeschi, Chief of Pathology at the Framingham Union Hospital.

The Heart of a Town

For the past 12 years a little town of 28,000 inhabitants in Middlesex County, Massachusetts, has been cooperating with the National Heart Institute in a long-range study of heart disease. One-third of the residents of Framingham between the ages of 30 and 60—the age bracket most likely to develop heart ailments—have volunteered to submit to periodic examinations by NHI investigators and local physicians to determine what roles heredity, environment, diet, and other factors play in the incidence of coronary disease. The results of these investigations are expected to contribute significantly to the war against America's number one killer.



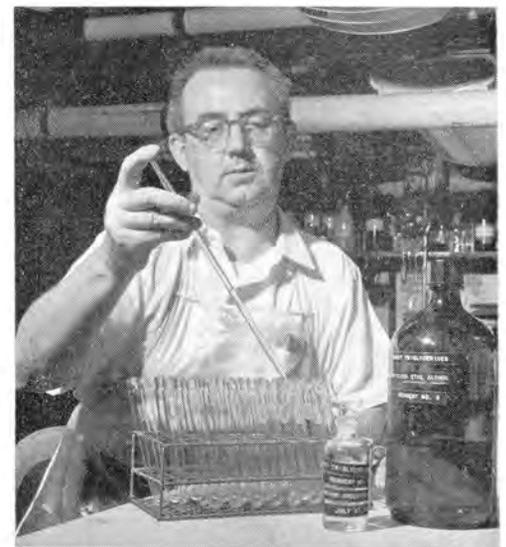
The young and old—as pictured on Framingham's village green—will someday benefit from the NHI cooperative study.



Arthur I. O'Brien, a study participant, talks to staff nurse Terry Ceredonna as he prepares for an examination.



Mr. O'Brien takes a maximal capacity test with the assistance of ECG technician Norma Buscone.



William E. Glennon determines total serum triglycerides, a measurement of blood lipids.

Photos by Jerry Hecht



Dr. William B. Kannel (left), Associate Director of the study, and J. William Claffey, x-ray technician, measure x-ray films.



Many employees of the Dennison Manufacturing Company, a major Framingham industry, are participants in the heart disease study.

Dr. Brackett Retires; Rekowned Biophysicist Serves NIH 25 Years

Dr. Frederick S. Brackett, Chief of the Section on Photobiology, Laboratory of Physical Biology, National Institute of Arthritis and Metabolic Diseases, retired last month after 25 years at NIH.

Dr. Brackett is internationally known for his work in the field of spectroscopy as applied to biological research. He first gained recognition in 1922 when his paper, "New Series of Spectrum Lines" (a series of hydrogen lines in the infrared spectrum), was published in *Nature*. Today these lines are known as the Brackett Series.

Paper Lays Groundwork

In 1928 his paper, "Characteristic Differentiations in the Spectra of Saturated Hydrocarbons," published in the *Proceedings of the National Academy of Sciences*, laid the groundwork for much important academic and industrial use of infrared spectra in the determination of molecular structure.

Dr. Brackett began his Federal career in 1917 at the National Bureau of Standards where he worked until 1919 as a laboratory assistant.

He spent a brief period—from June 1919 until October 1920—as an observer at the Carnegie Institute's Mount Wilson Observatory, where his observation of the infrared spectrum of the sun provided the groundwork for his subsequent discovery of the infrared spectral lines of the hydrogen atom.

Teaches in California

Dr. Brackett returned to the Bureau of Standards in 1921 as a part-time consultant while concurrently working on his physics doctorate. He has also been an assistant and associate professor of physics at the University of California.

In 1927 he joined the Department of Agriculture's Fixed Nitrogen Laboratory and has been associated



Dr. Frederick S. Brackett (left) receives a bound copy of his own works and a scroll of verse from Dr. Heinz Specht, Chief of NIAMD's Laboratory of Physical Biology. The gifts were presented at the time of his retirement last month.

with Federal laboratories ever since.

He has been a senior physicist at the Department of Agriculture, Director of the Division of Radiation and Organisms of the Smithsonian Institution, and Consultant to the Division of Cotton Economics of the Department of Agriculture.

In 1936 he came to the then National Institute of Health as director of biophysics research, and consultant and advisor on biophysics in cancer.

Dr. Brackett is one of a small number of men proficient in the design of optical equipment, a field which has extensive application in modern biophysics.

During World War II, Dr. Brackett suspended his NIH research to direct physical research in optics for the Armored Force Medical Research Laboratory. He received the Legion of Merit award for his research, design, and development of vision and fire control equipment for combat vehicles.

Born in Claremont, Calif., Dr. Brackett was graduated from Pomona College in 1918. He received his Ph.D. from Johns Hopkins University in 1922.

He will continue at NIH as a consultant in photobiology.

ENZYME

(Continued from Page 4)

conditions of acidity or alkalinity. The proper coiling and disulfide bridge formation in ribonuclease proceeded best under neutral or moderately alkaline conditions.

It would not occur at all under strongly acid conditions and not all of the amino acid chain components were essential. Folding would occur, though less efficiently, after as many as twenty amino acids had been chemically cut off one end of the chain.

The high degree of precision with which the complex processes

of enzyme synthesis are conducted and coordinated and the rugged durability of the finished product are due in part to the "design" of their molecules.

Dr. Anfinsen concluded that the apparent ingenuity involved in these designs cannot be properly attributed to any rational choices on the part of Nature.

The mutations that produced them came about purely by chance. They have persisted, not because they were rational or brilliant, but because they better fitted the organism to meet the random challenges that have thus far been posed by its environment.

NEAR-PURE VIRUS

(Continued from Page 1)

the work in NCI's Laboratory of Viral Oncology.

An electron microscope study by Dr. Dalton called the scientists' attention to the possibility of extracting virus from blood. With the microscope, Dr. Dalton saw many virus particles in the blood plasma of rats having so-called Moloney leukemia.

This leukemia was discovered three years ago in mice inoculated by Dr. Moloney with cell-free extracts of a solid tumor.

Uses Centrifuge

The procedure used to extract virus from leukemic rats consists essentially of spinning blood at high speeds in a centrifuge four times to separate the virus from blood cells and other debris.

In this way, the scientists obtain a small solid pellet consisting almost entirely of virus particles thus paving the way for the studying of the Moloney virus with greater precision than has hitherto been possible.

Dr. Dalton described his study at a meeting of the Electron Microscope Society of America in Pittsburgh last month.

He said that large numbers of virus particles usually were found in bone marrow and spleen megakaryocytes (cells which form platelets, an important portion of blood) of the leukemic animals.

The possibility that virus particles might also be present in the blood was checked by electron microscope inspection of pellets prepared by spinning the blood plasma (containing platelets but no red or white cells) of rats at relatively low speed in a centrifuge. These pellets were found to have virus particles in some of the platelets and in the spaces between platelets.

Describes Procedure

The Bern presentation by Dr. Dalton included a description of the procedure for extracting virus. He explained that blood is withdrawn from rats, treated chemically to break up clumps of cells and prevent clotting, and then spun at relatively low speed in a centrifuge.

The cells and other heavy material form a sediment that settles to the bottom of the centrifuge tube. Virus particles and other light-weight material remain at the top of the tube and are removed for additional spinning.

This is done three times more, at speeds up to 22,000 revolutions per minute (equal to 30,000 times the force of gravity in the centrifuge used in this work), so that the final sediment is a small solid

Snowshoe Hare Yields First Viral Isolate of California Encephalitis

Scientists from the Rocky Mountain Laboratory of the National Institute of Allergy and Infectious Diseases have made what is probably the first direct isolation of California encephalitis virus from a naturally infected vertebrate. The scientists, Dr. Willy Burgdorfer, Verne F. Newhouse and Dr. Leo A. Thomas, have reported on their work in the *American Journal of Hygiene*.

The viral agent was isolated from the blood of a snowshoe hare captured in the Bitterroot Valley of Western Montana and was identified serologically and by animal susceptibility tests as California encephalitis virus.

Two Tests Positive

In complement-fixation tests against immune sera of 35 vital agents, the isolate gave positive complement-fixation only in the homologous and California encephalitis virus systems. The behavior of the virus was also studied in tissue culture and embryonated chickens.

The finding adds weight to the presumption that the disease is maintained in nature in a cycle of mosquitoes, rabbits, and a ground squirrel, and possibly other vertebrates, including domestic animals. It also indicates that the distribution of the virus might not be as limited geographically as previously believed.

In Kern County, Calif., where the virus originally was isolated from two species of mosquitoes, inapparent infection in man has been shown to be common; clinical illness has also been linked to the agent.

pellet consisting almost entirely of virus particles.

Two pellets are made from the blood of each rat. (Rats are used instead of mice because of their greater blood volume.) Their virus content is studied by electron microscope inspection and by assay in animals.

Very thin sections of a pellet, magnified 10,000 to 20,000 times by the electron microscope, are seen to contain hundreds of particles with the fine structure characteristic of the Moloney virus. That is, the particles appear to consist of a somewhat dense, central core surrounded by outer rings that tend to converge. (The particle actually is spherical, but is seen in only two dimensions in a section cut for microscopic inspection.)

The second pellet prepared from the blood of each rat is mixed with appropriate fluid for reinjection into rats and mice.

NINDB Study Suggests Nerve Fiber Specificity May Occur in Mammals

A key to how the human body synthesizes long chain fatty acids—essential for the conversion of food to body fat—may be contained in a study recently completed by the National Institute of Neurological Diseases and Blindness.

The study was conducted by Dr. Roscoe Brady of NINDB's Laboratory of Neurochemistry, who reported his findings at the Fifth International Congress on Biochemistry held August 10-16 in Moscow.

Describes Sequence

The report contained the first description of the detailed sequence of biochemical reactions for the formation of long chain fatty acids in the cells of living animals.

This description, which contributes to basic knowledge of lipid (fat) synthesis, is potentially important in studies of various neurological and endocrine disorders.

Fatty acids combine with glycerine to form fats, and are essential components of myelin, the fatty nerve sheath which degenerates in such disorders as multiple sclerosis.

Converts Acetic Acid

In his study of purified enzyme systems from liver and brain, Dr. Brady has shown how acetic acid, a two-carbon unit, is converted to a 16-carbon chain fatty acid known as palmitic acid.

Early studies conducted by Dr. Brady and other scientists established that the presence of carbon dioxide was essential for fatty acid synthesis. Subsequent work by Dr. Brady indicated that the key intermediate in the biosynthesis is a derivative of malonic acid, known as malonyl coenzyme A.

Synthesizes Coenzyme

As a result, Dr. Brady was able to synthesize malonyl coenzyme A by adding carbon dioxide to acetyl coenzyme A, a derivative of acetic acid. By labeling these compounds, he discovered that one molecule of acetyl coenzyme A and seven molecules of malonyl coenzyme A, combined with 14 atoms of hydrogen, are required to build a molecule of palmitic acid.

The steps in the synthesis are as follows: When acetyl coenzyme A combines (condenses), the newly added carbon dioxide is displaced. When two atoms of hydrogen are added, the resulting compound can condense with another molecule of malonyl coenzyme A. This process is repeated until the appropriate

MICROBIOLOGIST

(Continued from Page 1)

parison with American cultures of the same microbe.

While at NIH Dr. Yelinov will not do actual bench work in his research. But he said at a recent press conference that "I will not avoid bench work (translated in Russian as 'working with one's hands') to learn new techniques."

Commenting on NIH he said,



Dr. Nikolai P. Yelinov, Russian exchange scientist, studies an NIH-developed aerosol exposure chamber for mice. The Soviet visitor is working in NIAID's Laboratory of Infectious Diseases.—Photo by Jerry Hecht.

"Although I have seen NIH only superficially, I have a favorable impression of this colossal scientific institution."

The first Russian scientist to do research at an American institution under the exchange program was Dr. Fedor Grigorevich Uglov, Head of the Chair of Hospital Surgery of the Pavlov Medical Institute in Leningrad, who spent nearly two months at Baylor University's College of Medicine last spring.

Dr. John Edgcomb of the National Cancer Institute was the first United States scientist to go to Russia for extended research. He left June 23 for a 6-month stay in the Soviet Union where he is presently engaged in collaborative cancer studies at the Institute of Experimental Pathology and Therapy of Cancer in the Academy of Sciences in Moscow.

chain length is reached.

To identify the intermediates which participate in these reactions, the required enzyme systems were isolated and purified. Each step in the sequence of reactions leading to fatty acid synthesis is in the process of definition by radioactive labeling of synthetic intermediary compounds.

3 Enzymes in Microbe Perform Same Action

One of the enzymatic assembly lines of the microbe, *Escherichia coli*, has three enzymes doing the same job. All three attach a single phosphate group onto the amino acid, aspartic acid.

This apparent triplication of effort may be essential to the efficiency of that particular assembly line, according to Dr. E. R. Stadtman of the National Heart Institute's Laboratory of Cellular Physiology and Metabolism, and Drs. G. N. Cohen and Gisele LeBras of the Pasteur Institute, Paris. The results of their collaborative study will appear in the *Journal of Biological Chemistry*.

The scientists point out that *E. coli* may need each of these enzymes because their product is a step in making three amino acids: lysine, threonine, and methionine.

The rate at which substances are synthesized by the cell for its own use is governed by two mechanisms called "feedback" and "repression." In "feedback," the surplus product slows production by stopping work of existing enzymes. In "repression," the surplus product blocks reproduction of enzymes.

Difficulties could arise for *E. coli* if only a single enzyme catalyzed the first step of this metabolic sequence leading to the formation of three amino acids. A surplus of any one under such a circumstance could also curtail production of the others and possibly lead to cellular shortages. The presence of three enzymes prevents such an occurrence.

Negative Charge Used To Inactivate Viruses Attacking Bacteria

Research in the Laboratory of Chemical Pharmacology, National Cancer Institute, has shown that specially-made chemicals can inactivate viruses ordinarily attacking bacteria. These studies are being made in NCI's Macromolecular Chemistry Section, headed by Dr. Peter T. Mora.

Dr. Mora reported on the work at the 18th International Congress of Pure and Applied Chemistry, held last month in Montreal.

He said that viruses permanently lost their ability to infect bacteria as a result of treatment with compounds having strong negative electrical charges. The compounds were polysaccharide molecules to which sulfate groups were linked chemically.

Bacterial viruses (bacteriophages) are tadpole-shaped organisms with head and tail made of protein. Packed tightly inside the head is deoxyribonucleic acid (DNA), the infective portion of the viruses.

The negatively charged compounds used in this work appeared to act by drawing certain positively charged compounds (amines) away from the viral DNA. The remaining negatively charged components of DNA repelled one another, thus expanding the tightly coiled DNA molecules inside the head of the viruses. As a result, DNA could not pass through the tail and into the bacterial cell wall.

These findings are being followed up to determine if polyoma virus, which induces tumors in animals, can be inactivated in the same way.

Dr. Daft Participates in Groundbreaking



Dr. Floyd S. Daft, NIAMD Director, lifts the first spadeful of soil at groundbreaking ceremonies for the new FASEB headquarters building on the Rockville Pike. Dr. Milton O. Lee, Federation Executive Officer, is at right.

Dr. Floyd S. Daft, Director of the National Institute of Arthritis and Metabolic Diseases, turned the first spadeful of soil at groundbreaking ceremonies August 14 for the new headquarters building of the Federation of American Societies for Experimental Biology.

The new 5-story Georgian structure will be located next to the present FASEB headquarters on the Rockville Pike just north of NIH. Plans for the building include a modern duplicating unit and a publications center. Completion is expected in September 1962.

Organized in 1912, the Federation is composed of six national biological research societies whose presidents serve as FASEB board chairman, on a rotating basis.

Dr. Daft, past president of the American Institute of Nutrition, a member society, is the current chairman.

NCI Investigators Reveal Leukemic Cells' Metabolic Differences

Research by scientists at the National Cancer Institute has revealed a difference in the way two types of leukemic cells conduct their metabolic, or energy-producing activity.

According to the Cancer Institute, knowledge of this kind is helpful in designing more effective drugs against leukemia.

Dr. Dean Burk of the Institute's Laboratory of Biochemistry reported this finding at a meeting of the Fifth International Congress of Biochemistry, which was held August 10-16 in Moscow.

WHO Provides Fellowships

The work was carried out by a group that included Dr. John Laszlo, formerly of NCI's General Medicine Branch; Dr. Berigoj Stambuk, formerly of the Laboratory of Biochemistry; Dr. Mark Woods, of the Laboratory of Biochemistry; and a Russian investigator, Dr. Joseph F. Seitz, who spent six months at the Institute under a World Health Organization fellowship.

Drs. Burk and Woods have long studied cell metabolism along guidelines proposed some 30 years ago by the German biochemist, Otto Warburg, who held that cancer-causing agents reduce a cell's ability to use oxygen to convert sugar into energy.

Requires No Oxygen

While normal cells rely almost exclusively on a process requiring oxygen (respiration), malignant cells, according to Warburg, also employ a process needing no oxygen (fermentation).

A few years ago a group headed by Drs. Burk and Woods was able to attribute the high level of fermentation in malignant cells to a breakdown in the hormonal mechanism that normally controls one step in metabolism. Suspecting that this defect could provide a target for cancer chemotherapy, the group then tested and confirmed the ability of several known anticancer agents to restore the missing control.

Leukemia Rate Differs

The results also furnished evidence of a difference in the rates of fermentation by both normal and leukemic cells of the lymph system and those of the bone marrow in the presence of oxygen. Supporting data were obtained by measuring the fermentation of cells maintained under conditions closely simulating those prevailing within

HOW TO BEAT THE SUMMER HEAT



Wearing a parka to protect herself from the 12° F. temperature of a National Heart Institute cold room, Carol O'Brien pours a chemical into a protein solution as part of an experiment made in the NHI Laboratory of Chemical Pharmacology. Carol, a student at Ursuline Academy, was one of seven high school science students who worked this summer in NHI and Naval Medical Center laboratories under Medical Research Fellowships awarded by the Montgomery County Tuberculosis and Heart Association. (NIH Record, June 6, 1961.)—Photo by Jerry Hecht.

NIMH Officials Conduct Seminar at Chapel Hill

Dr. Morton Kramer, Chief of the Biometrics Branch, and Earl S. Pollack, Chief of the Hospital Studies Section, NIMH, were guest specialists and conductors of a seminar at a recent mental health statistics training institute at the University of North Carolina.

The two-week course—supported by an NIMH project grant—was designed to provide basic instruction and review in statistical methods for persons working in State mental health programs in the South.

Observers from California, Oregon, and the American Psychiatric Association, who also attended the institute, anticipate the establishment of similar training programs in other parts of the country.

the body.

Though these findings indicate a difference between lymph and bone marrow cells, they reveal no distinction, with respect to fermentation, between normal and leukemic cells.

Further studies are under way to determine whether normal and leukemic cells differ with respect to respiration. A finding of such a difference might enable drugs to be designed that would attack leukemic cells only. The tendency of known anticancer drugs to damage normal as well as malignant cells

West Indies Scientist Is NINDB Lecturer

Dr. Eric Cruickshank, Professor of Medicine at University College of the West Indies, Jamaica, was guest speaker at an NINDB Epidemiology Branch lecture held August 24 in the Clinical Center.

Dr. Cruickshank's subject was "Epidemiologic Investigations of Nutritional Disorders Affecting the Nervous System in Jamaica." Using slides, he showed how neurological symptoms of nutritional disorders in Jamaica differed from those found in other parts of the world, notably Africa.

While no clear cause or causes of the Jamaican neurological disorder pattern have been proved, Dr. Cruickshank suggested that three factors may act together to produce the illness. These factors are toxic products from plants, especially a "bush tea" of which the natives are fond; nutritional deficiencies in the typically poor diet; and spirochete infection.

is one of the most difficult problems in cancer chemotherapy.

Drs. Burk and Woods are continuing this work at the Cancer Institute. Dr. Seitz is now at the Institute of Hematology and Blood Transfusion, Leningrad. Drs. Laszlo and Stambuk, respectively, are now at the Duke University Medical School, and the Veterans Administration Hospital, Chicago.

Deep Cold Found To Preserve Live Protozoa

An important new method of preserving live protozoa for use in medical research has been developed by Dr. Louis S. Diamond of the Laboratory of Parasitic Diseases, National Institute of Allergy and Infectious Diseases, in cooperation with Drs. Harold T. Meryman and Emanuel Kafig, Naval Medical Research Institute, National Naval Medical Center.

Dr. Diamond reported the success of the new method in tests performed with the parasite *Entamoeba histolytica*, an organism that causes amebic dysentery, in a paper presented last month at the meeting of the American Society of Parasitologists at Purdue University.

The scientists found that slowly freezing the one-celled animals and then cold-storing them in liquid nitrogen would preserve them intact for an indefinite period of time. When required, the frozen organisms need only be thawed and they are ready for use.

Fulfills Criteria

The criteria of successful preservation for such an organism are that it be capable of survival and reproduction when brought out of storage. The liquid nitrogen method fulfills these criteria.

In the past, several methods of providing live specimen protozoa for laboratory research have been used. One way was simply to preserve them alive in culture. This created two problems, both connected with their method of reproduction. First, since they reproduce by fission new generations of *entamoeba* quickly begin to crowd one another making it necessary that some be transferred to new cultures, a time-consuming operation. Second and more important, succeeding generations exhibit genetic changes and progeny may eventually become quite different.

Another, and the most common method of preservation also involves freezing and cold-storage, using dry ice as the refrigerant. In both the liquid nitrogen and dry ice methods some organisms are lost in the freezing process but those that are frozen intact and stored in liquid nitrogen will remain undamaged indefinitely, while those stored in dry ice will die after a period of time.

Development of the liquid nitrogen method provides laboratory investigators with an economical, efficient method of storing live protozoa, some of which cause severe, dangerous infections.